Report of the Veterinary Bacteriologist.

Division of Veterinary Science,
Pretoria, 12th August, 1904.

THE DIRECTOR OF AGRICULTURE.

SIR,

I have the honour to submit my report for the year 1903–1904. This is the first exhaustive report of the Veterinary Bacteriological Laboratories. In order to give a complete account of the steady progress of our investigations, I have found it necessary to incorporate the results of the experiments, which either have not previously been published or have appeared from time to time in the several departmental publications, as well as in various English and Continental journals.

The work may be divided into two parts; the first embraces the ordinary routine work of the laboratory, viz., the manufacture of rinderpest serum, biting-sickness virus, and calf vaccine; and the microscopical examination of specimens, the enumeration of which is given in tabulated form in the report. In connection with the office work of the station, it may be stated that during the past year 1,800 letters were received and 2,000 despatched; while 300 telegrams were received and 600 despatched. The second portion of the report comprises a series of articles dealing with experiments on the various South African diseases of domesticated animals. It was especially East Coast Fever that occupied my attention. The field experiments relating to this disease were undertaken in company with Mr. Stewart Stockman, Principal Veterinary Surgeon. The recorded experiments on horse-sickness date back to the year 1902. You will notice in the summary of the investigations of horse-sickness that the principle of a method of immunisation, at least for mules, is established; but that the inoculation cannot at present be carried out, owing to the haemolytic effect which the serum has on some animals. This difficulty will have to be overcome, before successful inoculation is practicable.

I regret to say that the premises in connection with the laboratory do not meet the increasing requirements of the station. Towards the close of the year three new stables and a forage room were built, which, however, are not sufficient to shelter all the animals.

It gives me much pleasure to certify to the good work done by my several assistants during the past year. At the beginning of the year, my staff consisted of Mr. B. Porta, Mr. E. Heron, Mr. A. von Bergen, Mr. P. R. Ferreira and Mr. D. Ferreira. Later, it was augmented by the following gentlemen :— Mr. Stevenson Cameron, Secretary; and Mr. J. C. Fletcher and Mr. Schneerger, Assistants. The work on East Coast Fever necessitated a permanent assistant in Nelspruit. Unfortunately, all the assistants in turn contracted malarial fever and had, consequently, to suspend work. Further we experienced a sad loss in the death of Mr. P. R. Ferreira, a most able and excellent official, who is greatly regretted by the staff and myself.

Datum/Date.....

Travelling.

The duties in connection with this office necessitated my absence from headquarters at different intervals. On August 25th, I proceeded to Potchefstroom to make arrangements with Mr. Alexander Holm, Manager of the Government Experimental Farm, for inoculating some imported cattle against redwater. The result of this inoculation is alluded to in the article, "The Piroplasma Bigeminum of the Immune Ox."

On September 18th, a journey was undertaken to Wit River in connection with an obscure disease amongst donkeys, which proved to be identical with that which I observed in 1897 during the first rinderpest outbreak. This disease is contagious, and consists of a dermatitis of the heels, complicated with inflamation of the coronary band and the lamina of the hoof.

The journey was continued to Delagoa Bay to inspect the landing arrangements for cattle which were daily expected to arrive from Texas, U.S.A. All necessary precautions to prevent tick infection were taken, and the close examination of several head of cattle living on the premises proved an absence of any ticks.

On the 19th and 30th of October, and 9th and 18th of November, and the 2nd and 14th of December, 1903, visits were made to Nelspruit, where an experimental camp was established in connection with East Coast Fever.

During the year 1904, visits to the same place were made on January 10th and 23rd, March 30th and May 13th. The results of the experiments are given in my different articles on East Coast Fever. Several visits were made to the bushveld and also to Bronkhorstspruit for the purpose of inspecting our serum cattle, which were kept in different lots and on different farms.

I attended the Conference on Diseases amongst Cattle and other Animals in South Africa, held in Bloemfontein on December 3rd, 4th, and 5th, 1903.

From May 25th to June 1st, I attended a similar Conference in Capetown.

On April 6th, I visited Johannesburg and read a paper on East Coast Fever before the South African Association for the advancement of science. It reviewed our knowledge of the disease up to that date; and was published in the *Transvaal Agricultural Journal* (No. 7). I wish to state that these visits were made in company with my colleague, Mr. Stewart Stockman, P.V.S., who co-operated with me in some of the investigations relating to East Coast Fever.

On the 27th October, 1903, I travelled for three days, together with Mr. Rose-Innes, Resident Magistrate for Pretoria, in order to meet the farmers north of the Magaliesberg in the Pretoria District. Three meetings were held at which I explained the nature of East Coast Fever.

Rinderpest Serum Statistics.

(*Production and Issue.*)

At the beginning of the financial year, 1903, rinderpest had almost disappeared from the Transvaal. There was only one outbreak of the disease in the Native Reserve of Zeerust, which was stamped out with the last supply of serum forwarded in June. At that period 938,775 c.c., were in stock. It

considered necessary, however, to increase this stock to a minimum of 000 doses at 100 c.c., in order to meet any emergency, should the disease ppear in our Colony. For this purpose, the production of serum was main- ed. We had, however, some difficulty in attaining the desired result in shortest possible time, inasmuch as pleuro-pneumonia, which was introduced a fresh supply of cattle, necessitated the inoculation of all our stock against disease, and prevented us from using the animals for serum until the reaction to that inoculation had subsided. In order to reduce the expenses in nection with feeding cattle, as much as possible, it was deemed advisable asture the serum herd. For this purpose a farm was hired at Wonderboom. fortified cattle were brought to the station for bleeding purposes. But he meantime, Rhodesian Tick Fever had made its appearance and it became ossible to move the stock along the road to the farm. We were, therefore, ged to erect a temporary laboratory on the farm itself, where the bleeding rations were successfully carried out. The production of serum ceased in ember, 1903. Some of the oxen were then sold. The majority, however, e kept in readiness, in case they might be required for serum.

The issue of rinderpest serum to other Colonies was made in accordance a resolution passed at the South African Conference on stock diseases, at Bloemfontein on the 3rd, 4th and 5th December, 1903, where it was ded :—

> "In the interests of South Africa generally, those Colonies having a serum reserve, should offer to supply at cost price those Colonies insufficiently provided with it, who require it to cope with the present outbreak."

The cost price of serum was calculated to be £2 10s., per litre, for which the serum was supplied to Natal, Mombassa and German West Africa.

The following list gives the supply of serum on hand at the beginning of the year; the production during the year; the issue and the stock, together with the value.

	Rinderpest Serum.	
1903–4.	*Issued.*	*Value.*
y	80 Litres	£200
ruary	100 Litres	250
rch	60 Litres	150
pril	500 Litres	1,250
Total	740 Litres	£1,850

ntity in stock July 1st, 1903	938·775 c.c.
e during the year	640·000 c.c.
	1,578·775 c.c.
ed during the year	740·000 c.c.
tock June 30th, 1904	838·775 c.c.

Pleuro-pneumonia Virus.

Article 43, letter C of the Government Notice No. 834 of 1903, in pursuance of Section 5 of Ordinance No. 17 of 1902, provides for the inoculation of cattle which have been in contact with others suffering from lung-sickness. This rticle necessitated the manufacture of pleuro-pneumonia virus in the laboratories nder my charge, as in practice it was not always possible to have the suitable

material on stock. It was left to the District Veterinary Surgeons to use the virus they were able to collect themselves from animals suffering from pleuro-pneumonia, and which were killed for this purpose. In many instances, it was not possible to procure the virus, owing to the fact that proper animals were unobtainable. In other cases, the pleuro-pneumonia virus was contaminated, owing to putrid mortification of the lungs. At that time the production of reliable virus became urgent. For this purpose, the method proposed by Pasteur, namely, the subcutaneous inoculation of virus into the connective tissue of a calf was first employed. But it was then found that a good many calves did not produce sufficient virus and, further, there was not always sufficient demand, so that the virus in stock became old and useless and, naturally, had to be destroyed. This was an expenditure which could not be justified, since material and time were wasted.

Thus the pure culture of the virus, as introduced by Nocard and Roux, was undertaken. A series of experiments was then conducted on the station which showed that pure cultures could be successfully substituted for virus taken from the pleural cavity of oxen or from the subcutaneous tissue of calves. Indeed, the pure cultures proved to be so virulent that the subcutaneous injection into the *regions defendues*, that is, the loose tissue of the trunk, was followed by death; whereas the injection into the tail gave the same local swelling as caused by virus taken freshly from a sick animal. The pure culture has the advantage that within a short period great quantities can be made. Its production is cheap and does not necessitate the slaughter of animals. A further advantage is that, in making such cultures every two weeks, as is now the rule at the laboratories, a fresh supply is constantly maintained. Accordingly, for the last five months only cultures were issued.

The following is the issue of pleuro-pneumonia virus during the year 1903-4, together with their relative values.

1903–1904.	*Issued.*	*Value.*		
	c.c.	£	s.	d.
July	681	£17	0	6
August	2,375	59	7	6
September	1,190	29	15	0
October	3,910	97	15	0
November	500	12	10	0
December	924	23	2	0
January	1,517	37	18	6
February	190	4	15	0
March	675	16	17	6
April	50	1	5	0
May	150	3	15	0
June	364	9	2	0
	12,526	£313	3	0

VACCINE.

1903–4.	Transvaal.	O.R.C.	Natal.	Portuguese Territory.	Swaziland	No. of Lots.
July	3,524	6,750	500	3,000	—	43
August ..	2,760	2,500	1,000	4,000	—	38
September ..	6,811	11,200	—	4,000	—	55
October ..	4,583	13,200	1,500	—	—	59
November ..	5,795	7,500	—	4,000	—	47
December ..	4,165	2,200	—	—	—	33
January ..	4,602	900	—	3,000	—	37
February ..	3,910	1,600	—	2,000	50	37
March ..	7,222	5,000	—	2,000	—	51
April	4,575	3,500	1,500	2,100	—	50
May	5,848	2,000	4,850	100	100	59
June	2,931	1,000	23,000	2,100	—	44
	56,726	57,350	32,350	26,300	150	553

TOTAL .. 172,776 Tubes at 2d. = £1,439 16s. 9d.

TABULAR STATEMENT OF MICROSCOPICAL EXAMINATIONS, JULY, 1903, TOGETHER WITH MONTHLY SUMMARY, DISTRICT SUMMARY, AND SUMMARY OF DISEASES.

JULY, 1903.

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
1-7-03	Oxen, 1 and 2 ..	D.V.S. Elder ..	—	—	Embabaan ..	1, African Coast Fever. 2, nothing seen.
2-7-03	Kidney, Liver, and Spleen of Ox	P.V.S. Stockman ..	—	—	Rietfontein..	Strong infection African Coast Fever and ordinary Red-water.
,,	Ox	D.V.S. Turnbull ..	—	—	Machadodorp	Strong infection African Coast Fever.
3-7-03	,, ...	,, Dunphy ..	—	—	Nylstroom ..	Nothing seen.
,,	,, ...	Inspector Nicholls..	—	—	Pretoria ..	Nothing seen.
6-7-03	Tubercular Gland	F. Fernandes ..	—	—	L. Marques .	Tuberculosis.
,,	Oxen, 6 Smears ..	D.V.S. Elder ..	—	—	Embabaan ..	Nothing seen.
,,	6 Smears ..	V.S. Dale	—	—	Pretoria ..	,, ,,
8-7-03	Oxen	D.V.S. Edgar ..	—	—	Pietersburg..	,, ,,
9-7-03	,, 2	,, Turnbull ..	—	—	Machadodorp	Both African Coast Fever.
,,	Ox	,, Garraway ..	Bronkhorst ..	—	Eland's River	African Coast Fever.
10-7-03	1, Blood, Heart; 2, Artery	,, Chalmers ..	—	—	Heidelberg ..	Nothing seen.
,,	Wool, Goat ..	,, ,, ..	—	—	,, ..	No Acariosis found.
,,	Oxen, ...	,, Walker ..	—	—	Ermelo ..	Both African Coast Fever
14-7-03	Pus	,, ,, ..	—	—	,, ..	Contain Saccharomy. Farcimi.
15-7-03	Blood 2, A and B	,, Gardener ..	—	—	Embabaan ..	African Coast Fever.
16-7-03	Mule Pus, 3 ..	,, Conacher ..	—	Vlakfontein ..	Lydenburg ..	Saccharomy. Farcimi.
,,	Pus, 2	,, Walker ..	—	—	Ermelo ..	,, ,,
,,	Portion of Lung..	,, Dunphy ..	—	—	Nylstroom ..	—
17-7-03	Ox	,, Garraway ..	Erasmus	Garstfontein ..	Pretoria ..	Nothing seen.
,,	Pus	,, ,, ..	—	—	,, ..	,, ,,
,,	Tongue and Mouth of Pig	,, ,, ..	—	—	,, ..	Measles.
18-7-03	Smears, Oxen, 3..	,, Edgar ..	—	—	Nylstroom ..	African Coast Fever.
22-7-03	Pus 3, BC, BM, BCM	V.S. Stonewall Jackson	Repatriation ..	Hill Side.. ..	Pretoria ..	Saccharomy. Farcimi.

,,	Smears, Ox ..	Inspector Nicholls ..	Mark's	Scheerpoort ..	,, .	Nothing seen.
23-7-03	,, ,, ..	D.V.S. Edgar ..	—	—	Pietersburg ..	,, ,,
,,	Pus, 3	,, Conacher ..	—	—	Lydenburg ..	All contain Saccharomy. Farcimi.
,,	Smears, Oxen, 3 ..	,, ,, ..	—	—	,, ..	All African Coast Fever.
24-7-03	,, Mule ..	,, ,, ..	—	—	,, ..	Piroplasma Equi.
27-7-03	Blood & Spleen, 2; Liver & Blood, 2	,, Garraway ..	W. Erasmus ..	—	Pretoria ..	Smears decomposed.
,,	Oxen, 1, 2, 3, 4, and T.	V.S. Dale	—	—	,, ..	Nos. 1, 2 and 3, African Coast Fever.
,,	Pus, 3	,, ,, ..	—	—	,, ..	Saccharomy. Farcimi.
29-7-03	Oxen, 3	D.V.S. Conacher ..	—	—	Lydenburg ..	African Coast Fever.
,,	Pus, Mule ..	,, ,, ..	—	—	,, ..	Unable to diagnose.
30-7-03	,, 2 ..	V.S. F. Scott ..	—	—	Johannesburg	Both Saccharomy. Farcimi.
,,	Pus, 1-6 ..	D.V.S. Turnbull ..	—	—	Machadodorp	3 and 4, ,, ,,
,,	Oxen, 1 and 2 ..	,, Edgar ..	—	—	Nylstroom ..	Nothing seen.
,,	Smears, 4, Donkeys	,, ,, ..	—	—	,, ..	,, ,,
,,	Kaffir Ox ..	,, ,, ..	—	Nooitgedacht ..	,, ..	,, ,,
,,	Ox, Blood K & S, 3	,, ,, ..	—	Weltevreden ..	,, ..	African Coast Fever and Ordinary Redwater.
31-7-03	Pus, 3	,, Cochrane ..	—	—	Middelburg ..	Saccharomy. Farcimi.
,,	Ox Smears ..	,, ,, ..	—	—	,, ..	Nothing seen.

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AUGUST, 1903.

1-8-03	Ox Smears, 4 ..	D.V.S. Turnbull ..	—	—	Machadodorp	All African Coast Fever.
,,	,, ,, 2 ..	,, Edgar, per Inspector Nicholls	—	—	Nylstroom ..	Nothing seen.
3-8-03	Pus, Horses, 2 ..	D.V.S. Cannon ..	—	—	Standerton ..	Both Saccharomy. Farcimi.
,,	Ox Smears, 2 ..	,, ,, ..	—	—	,, ..	Nothing seen.
4-8-03	,, ,, 4 ..	,, Turnbull ..	—	Kleinfontein ..	Lydenburg & Machadodorp	Two contain African Coast Fever.
,,	Pus, Mule ..	,, ,, ..	—	—	Machadodorp	Saccharomy. Farcimi.
,,	Donkey Smears, 8	V.S. Dale	Repatriation ..	—	Pretoria ..	Nothing seen.

AUGUST, 1903.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
6-8-03	Bottle Blood ..	J. R. Hamilton ..	—	—	Standerton ..	Nothing seen. Blood hæmolytic
8-8-03	Pipettes, Pus, 1-4	D.V.S. Turnbull ..	—	—	Machadodorp	1 and 2, Saccharomy. Farcimi.
10-8-03	Pus, Pony ..	,, Webb ..	—	—	Barberton ..	Saccharomy. Farcimi.
,,	Ox Smears, 1-4 ..	,, Elder ..	—	—	Embabaan ..	1 and 2, African Coast Fever; 3 and 4, nothing seen.
,,	Sheep, 1	Repatriation ..	—	—	Pretoria ..	Spleen examined, anthrax found.
11-8-03	Pus, Mare ..	D.V.S. Webb ..	—	—	Barberton ..	Saccharomy. Farcimi.
,,	Sheep & Goats, 6	,, Chalmers ..	—	—	Heidelberg ..	Nothing seen, scrapings not deep enough.
12-8-03	Mule Smear ..	,, Conacher ..	P.W.D.	—	Lydenburg ..	Nothing seen.
,,	Ox ,, ..	,, Webb ..	—	Kleinboy's Kraal	Barberton ..	Strong infection African Coast Fever.
13-8-03	Box Wool, Sheep	,, Chalmers ..	—	—	Heidelberg ..	Nothing seen.
,,	Smears, Pus, 1, 2, 3	V.S. Fletcher ..	—	—	Middelburg ..	2 and 3, Saccharomy. Farcimi.
,,	,, Ox ..	D.V.S. Edgar ..	—	Pilgrim's Hoof, 468	Nylstroom ..	African Coast Fever.
14-8-03	,, Mule ..	,, Conacher ..	—	—	Lydenburg ..	Nothing seen.
17-8-03	,, Oxen, 2..	Inspector Nicholls..	—	—	Pretoria ..	Too decomposed for examination.
,,	,, Horse ..	D.V.S. Cannon ..	—	—	Standerton ..	Nothing seen.
,,	,, Ox ..	P.W.D.	—	—	Pretoria ..	Slight infection African Coast Fever.
,,	,, Oxen 1,2,3	D.V.S. Elder ..	—	—	Embabaan ..	Nothing seen.
18-8-03	,, Ox ..	W. McLaghlan ..	—	Mooiplaats, 57 ..	—	African Coast Fever.
,,	Pus, Samples, 2..	D.V.S. Turnbull ..	—	—	Machadodorp	Saccharomy. Farcimi.
20-8-03	Smears 1, 2, 3, 4, 5, not labelled	V.S. Dale	—	—	Repartiation, Durban.	Nothing seen
,,	Bottle Blood, Heart, Ox	D.V.S. Dunphy ..	—	—	Krugersdorp.	,, ,,
22-8-03	Smears, Ox ..	,, Garraway .. (Inspector Nicholls)	—	—	Pretoria ..	Strong infection African Coast Fever.
24-8-03	,, ,, ..	P.W.D.	—	—	,, ..	African Coast Fever.

,,	Wool and Scab of Sheep	D.V.S. Chalmers ..	—	—	Heidelberg ..	Nothing seen.
25-8-03	Smears, Donkey ..	V.S. Dale	—	—	Pretoria ..	,, ,,
,,	,, Ox ..	D.V.S. Garraway ..	—	New Site	,, ..	—
26-8-03	,, Mule ..	,, Conacher ..	Dr. Franks ..	Weltevreden ..	Lydenburg ..	Nothing seen.
,,	Pus 1, 2, 3, 4 ..	,, Turnbull ..	—	—	Machadodorp	All Saccharomy. Farcimi.
28-8-03	Smears, Ox ..	,, Bush ..	—	—	Piet Retief ..	African Coast Fever.
,,	,, ,, ..	,, Conacher ..	—	Wildebeeste Hoek	Lydenburg ..	,, ,, ,,
29-8-03	,, Mule, 3 ..	V.S. Stonewall Jackson	Repatriation ..	Hill Side	Pretoria ..	No. 2, few Pirosoma.
31-8-03	,, Oxen, 1, 2, 3, 4, & 5	,, Dale ..	,, ..	,, ...	,, ..	Nos. 2, 3 and 4, N. Cells.
	,, Donkeys14	,, ,, ...	,, ..	,, ...	,, ..	Nothing seen.
,,	,, Oxen, 1, 2, 3 and 4	,, Conacher ..	—	—	Lydenburg ..	Nos. 1, 2 and 3, African Coast Fever ; 4 and 5, nothing seen

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SEPTEMBER, 1903.

1-9-03	Mules, 1 and 2 ..	V.S. S. Jackson ..	Repatriation ..	—	Pretoria ..	Nothing seen.
,,	Horse	,, Dale	,, ..	—	,, ..	,, ,,
2-9-03	Ox	D.V.S. Walker ..	N. Tooza ..	Rietfontein ..	Carolina ..	African Coast Fever.
,,	,, ...	,, Conacher ..	McGee	Sterkspruit, 159 ..	Lydenburg ..	Nothing seen.
3-9-03	Ox and Cow ..	D.I.T. Repatriation, S.E.	Repatriation ..	Stock Farm ..	Vaalbank ..	,, ,,
,,	Ox and Heifer ..	P.V.S.	,, ..	Farm	Middelpunt ..	,, ,,
,,	Ox	—	Van B.	Wonderboom ..	—	,, ,,
,,	Specimen in bottle	D.V.S. Conacher ..	—	—	Lydenburg ..	,, ,,
,,	Ox	,, Sturge ..	—	—	Rustenburg	Strong African Coast Fever.
4-9-03	Ox Lung	,, Dunphy ..	—	—	Krugersdorp.	Nothing seen.
,,	Cow	,, Conacher ..	De Sousa	Borchoek, 1341 ..	Lydenburg ..	,, ,,
5-9-03	Demodex Pus ..	Agricultural Dept. ..	—	—	Pretoria ..	,, ,,
7-9-03	Ox	D.V.S. Bush ..	P. Morgensen ..	Klein Vrij Staat ..	Piet Retief ..	Strong African Coast Fever.
,,	Oxen, 2	Inspr. Tomlinson ..	Native	Spekboom, 36 ..	—	,, ,, ,, ,,
,,	,, 2	D.V.S. Garraway ..	T. Swartz ..	229, Mayville ..	Pretoria ..	African Coast Fever.

SEPTEMBER, 1903.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
7-9-03	Donkeys, 7 ..	V.S. Dale	Repatriation ..	—	Pretoria ..	Nothing seen.
8-9-03	Ox Kidney ..	D.V.S. Gardener ..	—	—	Embabaan ..	Strong African Coast Fever.
9-9-03	Ox, H.K.L.S. ..	„ Conacher ..	McGee	—	Lydenburg ..	African Coast Fever and Ordinary Redwater.
„	„ „ ..	P.V.S.	J. Oeljse	Honingnest Krantz	Pretoria ..	African Coast Fever.
„	Lung	D.V.S. Turnbull ..	—	—	Machadodorp	Nothing seen.
„	Oxen, 1, 2, 3 ..	„ Garraway ..	Reid	Hermanstad ..	Pretoria ..	„ „
„	Ox	„ „ ..	Nel	Wonderboom ..	„ ..	African Coast Fever.
10-9-03	Mule Pus and P.W.D. Mule	„ Webb ..	—	—	Barberton ..	Both Saccharomy. Farcimi.
11-9-03	Smears, Mule 2 ..	V.S. Crawford ..	Repatriation ..	Hill Side.. ..	Pretoria ..	Nothing seen.
12-9-03	Cow and Bull ..	D.V.S. Garraway ..	Prinsloo, Vermeulen	Zoekoegat, 287 ..	„ ..	African Coast Fever.
„	Oxen, 2	„ „ ..	Grobler, Ras ..	Derdepoort ..	„ ..	Nothing seen.
„	Ox	„ Turnbull ..	Mr. Balveir ..	Geluk, 291 ..	Machadodorp	„ „
„	„	„ Sturge ..	—	—	Rustenburg	African Coast Fever.
„	„	Inspector Dunkley	A. Geyser ..	—	P. P. Rust..	„ „ „
„	„	„ „	N. Masebe (Native)	—	„ ..	„ „ „
„	Oxen, 2	D.V.S. Webb ..	W. D. Millar, Gwajos Kraal	Jamestown ..	Barberton ..	Both African Coast Fever.
14-9-03	Cow	„ „ ..	Gindes Kraal ..	Costinia Block ..	„ ..	Nothing seen.
„	Ox	„ Garraway ..	Engelhecht ..	Kamelzyn Kraal	Pretoria ..	Smears hæmolytic.
15-9-03	„	„ Edgar ..	Town Ox.. ..	—	Pietersburg..	Ordinary Redwater.
„	Oxen, 4	„ „ ..	—	Weltevreden, 140 Witkop	„ ..	2, African Coast Fever; 2, nothing seen.
„	„ 2	„ Turnbull ..	M. Grobler ..	Schoongezicht, 302	Machadodorp	Both African Coast Fever.
16-9-03	„ 6	„ Edgar ..	—	—	Pietersburg..	5 oxen, African Coast Fever; 1 ox, nothing seen.
„	Cow	Inspector Dunkley	Mr. Maggs ..	—	P. P. Rust..	African Coast Fever.
„	Oxen, 4	„ Nicholls ..	Plunkett, Potgieter	Bockenhoud, Middelfontein	Nylstroom ..	Nothing seen, smears not good.
„	Calf	„ Lester ..	Mr. Lombard ..	Rooidraai, 1242..	Lydenburg ..	Basic cells.

18-9-03	Oxen, 3	D.V.S. Bush ..	Liversedge (Native), M'Pendeese	Mooiplaats, Spekboom, Witcliff	Piet Retief ..	African Coast Fever.
,,	Heifer	Experimental Farm	Government ..	Experimental Farm	Potchefstroom	Ordinary Redwater.
19-9-03	Bull and Ox ..	D.V.S. Garraway ..	Mr. Erasmus ..	Sjambok Kraal, 52	Pretoria ..	Bull, nothing seen; ox, African Coast Fever.
,,	Ox	—	Native Plang ..	Mamagahalis ..	—	African Coast Fever.
20-9-03	Oxen, 2	Experimental Farm	Government ..	—	Potchefstroom	Both Ordinary Redwater.
21-9-03	,, 8	D.V.S. Conacher ..	Lena Settlement	—	Middelburg..	Nos. 1, 2, 3, 4, 5, 6, African Coast Fever; 7 and 8, nothing seen.
,,	Ox	Cond. Patterson ..	Repatriation ..	Bushveld ..	Rustenburg	Ordinary Redwater.
22-9-03	Oxen, 6	V.S. Crawford ..	,,	—	Nylstroom, 4; Pietersburg, 1; Bushveld, 1	Nothing seen.
,,	Scab Scraping, Sheep, 5	D.V.S. Chalmers ..	—	—	Heidelberg ..	,, ,,
,,	Oxen, 4	,, Conacher ..	Government ..	Experimental Farm	Potchefstroom	3 oxen, nothing seen; 1 ox, African Coast Fever.
23-9-03	,, 6	,, Garraway ..	S. Marks.. ..	Swaartkoppies ..	Pretoria ..	4 oxen, nothing seen; 2, African Coast Fever.
,,	Cow	,, Elder ..	—	—	Embabaan ..	African Coast Fever.
,,	Oxen, 3	,, Edgar ..	Native	Kalkbult, 131 ..	Pietersburg..	,, ,, ,,
			Land Dept. ..	—	—	Nothing seen.
			Mr. Bosman ..	Doornkraal ..	—	African Coast Fever.
,,	Mule Pus.. ..	V.S. Dale	—	—	Pretoria ..	Saccharomy. Farcimi.
24-9-03	Sheep Scraping..	D.V.S. Chalmers ..	Van der Merwe..	Groenfontein ..	Heidelberg ..	Nothing seen.
25-9-03	Oxen, 3	Inspector Dunkley	T. Smit	Tweefontein ..	Zoutpansberg	African Coast Fever.
			P. Schutte ..	Setaba Drift ..	,,	,, ,, ,,
			A. Becker ..	Broderstroom ..	,,	,, ,, ,,
,,	Pus, 2	D.V.S. Turnbull ..	Repatriation ..	—	Machadodorp	Both Saccharomy. Farcimi.
,,	Oxen, 2	,, ,, ..	Breytenbach ..	Schoonje, 38 ..	,, ..	1, African Coast Fever; 1, nothing seen.
,,	Piece Lung ..	,, Dunkley ..	—	—	Krugersdorp.	Nothing seen.
26-9-03	Ox	,, Webb ..	Mr. Nolingart ..	Louw's Creek ..	Barberton ..	African Coast Fever.
,,	Ox Heart, Liver, and Spleen	,, Edgar ..	F. Shiels	River Platz, 29 ..	Pietersburg..	Nothing seen; smears not good.
28-9-03	Mule and Ox ..	,, Sturge ..	Government ..	Witkop	Rustenburg .	Mule, biliary fever; ox, nothing seen.

SEPTEMBER, 1903.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
28-9-03	Sheep Scraping..	D.V.S. Chalmers ..	Mr. De Jager ..	Boschfontein, 271	Heidelberg ..	Acariosis.
,,	Oxen, 2	Inspector Dunkley	Mr. Tue	Wildeboschfontein	Pietersburg..	Nothing seen.
,,	Ox	D.V.S. Webb ..	Native, Tabanus	Goodehoep, 1176	Barberton ..	African Coast Fever.
,,	Ox Kidney, Liver, and Spleen	,, Garraway ..	J. Maritz ..	Van Dyk Spruit..	Pretoria ..	,, ,, ,,
29-9-03	Oxen, 2	,, Bush ..	,, ..	Witkoppies, 59 ..	Piet Retief ..	,, ,, ,,
,,	,, 2	,, ,, ..	G. Ferreira ..	Holfontein, Klein Vrij Staat	,, ,, ..	Nothing seen.
,,	,, 4	P.V.S., per D.V.S. Conacher	—	—	Pretoria ..	2, African Coast Fever; 2, nothing seen.
30-9-03	,, 3	D.V.S. Garraway ..	Mr. Dun ..	Roodeplaats ..	,, ..	Nothing seen.
			,, ,, ..	,, ..	,, ..	,, ,,
			,, Botha	Vorstfontein ..	,, ..	,, ,,
,,	,, 2	,, Turnbull ..	,, Gurntans ..	Rooidraai ..	Machadodorp	African Coast Fever.
			,, Breitenbach	Vlakfontein, 131	,,	Nothing seen, smears decomposed.

Number examined for September, 1903, 142.

OCTOBER, 1903.

1-10-03	Sick Ox	D.V.S. Elder ..	S.A.C.	—	Embabaan ..	African Coast Fever.
,,	Ox	,, May ..	—	Witkop, 1543 ..	Nylstroom ..	Strong African Coast Fever.
2-10-03	,, ...	,, Garraway ..	P. van Wyk ..	Silverton ..	Pretoria ..	Nothing seen.
,,	Oxen, 2	,, Cochrane ..	—	—	Middelburg..	,, ,,
,,	Cows, 2	,, Edgar ..	Mr. Monctford ..	Sterk Kloof, 91; Dairy Farm on Townlands	Pietersburg..	1 cow, nothing seen; 1, African Coast Fever.
3-10-03	Oxen, 2	V.S. Lee	—	—	Springs ..	Nothing seen.
4-10-03	Ox	D.V.S. Bush ..	J. Carelson ..	Town Lands ..	Piet Retief ..	Slight African Coast Fever.

5-10-03	Horse, Smear	Inspector Lester ..	Repatriation ..	—	Lydenburg ..	Nothing seen.
	Mule ,	,,	,,		,,	,, ,,
,,	Oxen, 2	D.V.S. Turnbull ..	,, ..	Kleinfontein, 1253	Machadodorp	Both African Coast Fever.
,,	Cows, 2	,, Webb ..	Barrett's Berlin G.M. Company	—	Barberton ..	Nothing seen.
,,	Cow	,, Lee	Bonswan ..	Casino	Pretoria ..	,, ,,
,,	Ox	,, Garraway ..	Mr. Horn, Jr. ..	Bockenhout Kloof, 263	,, ..	Strong African Coast Fever.
6-10-03	,, ...	,, May ..	Native, Marcus ..	Du Toit's Kraal, 282	Nylstroom ..	African Coast Fever.
,,	Bull	,, Webb ..	Temlut's Kraal ..	Twillo, 1006 ..	Barberton ..	Nothing seen.
,,	Oxen, 2	,, Edgar ..	—	Kalkbat, 132, ..	Pietersburg ..	1, nothing seen;
			(2) D. Buchanan	Goedehoek, 3 ..	—	2, African Coast Fever.
7-10-03	Ox	,, Garraway ..	Native, Isaac ..	Kruisfontein, 164	Pretoria ..	Nothing seen.
,,	Mule Pus, 5 ..	,, Turnbull ..	Repatriation ..	—	Machadodorp	1, 2, 3, 4, nothing seen; 5, Saccharomy. Farcimi.
,,	Oxen, 8	,, Cochrane ..	—	—	Middelburg ..	Nothing seen.
8-10-03	Ox	Inspector Dunkley	Native on J. Potgieter's Farm	Sterkspruit ..	Woodbush, Zoutpansberg	Basic cells numerous.
,,	,, ...	,,	P. H. Hill ..	New Agatha ..	Zoutpansberg	African Coast Fever.
,,	,, ...	D.V.S. Garraway ..	A. H. Malan ..	Boschkop, 313 ..	Pretoria ..	Strong combined Ordinary Redwater and African Coast Fever.
,,	Ox Liver & Kidney	,, Sturge ..	J. M. v. Rooyen ..	Kroom River, 590	Rustenburg	Nothing seen;
11-10-03	Ox	Inspector Dunkley	Le Roux & Celliers	Van Tonder's Hoek	Zoutpansberg	,, ,,
13-10-03	,, ...	D.V.S. May ..	Matabata ..	Ongegund, 2080 ..	Waterberg, Nylstroom	,, ,,
14-10-03	,, ...	Inspector Lester ..	Native, Tosolos ..	Klipfontein, 1162	Lydenburg ..	Strong African Coast Fever.
,,	,, ...	D.V.S. Garraway ..	J. v. Staden ..	Rietfontein, 280 ..	Pretoria ..	African Coast Fever.
,,	Oxen, 2	,, ,, ..	H. du Preez ..	,, ..	,, ..	1, African Coast Fever; 2, nothing seen.
,,	,, 2	,, ,, ..	J. G. Jones ..	Hartebeestepont, 498	,, ..	African Coast Fever.
			J. P. v. d. Venter	Uitzicht	,, ..	,, ,, ,,
15-10-03	,, 4	,, Webb ..	Tamlut's Kraal ..	Twillo, 1006 ..	Barberton ..	Nothing seen.
			,, ,, ..	,, ,, ..	,, ..	Strong African Coast Fever.
	Dead Ox	—	Mapang's Kraal ..	,, ,, ..	,, ..	,, ,, ,, ,,
			,, ,, ..	,, ,, ..	,, ..	,, ,, ,, ,,

OCTOBER, 1903.—(*continued*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
16-10-03	Liver, Spleen, and Muscle	D.V.S. Garraway ..	G. v. Staden ..	Klipfontein, 482..	Pretoria ..	Strong African Coast Fever
,,	Piece Lung ..	V.S. Pye	—	—	,, ..	Saccharomyeosis.
17-10-03	Sheep, Scab Scraping	D.V.S. Chalmers ..	Native, Wolf ..	Marais Drift, 4 ..	Heidelberg ..	Acariosis.
19-10-03	Ox	,, Turnbull ..	N. Oosthuizen ..	Gelok, 29.. ..	Machadodorp	Nothing seen.
,,	Pus Pipettes, 3 ..	,, ,, ..	—	—	,,	All Saccharomy. Farcimi.
20-10-03	Heifer, Blood, Kidney, Spleen, and Liver	,, Cochrane ..	Land Settlement	Schiedpad ..	Middelburg..	Strong African Coast Fever.
21-10-03	Ox	Inspector Lester ..	Van der Merwe..	Doornkop, 1438..	Lydenburg..	African Coast Fever.
,,	,, ...	,, Dunkley..	P. N. Hill ..	New Agatha ..	Hænertsburg	Nothing seen.
,,	,, ...	D.V.S. Sturge ..	Combrink ..	Town Lands ..	Rustenburg	Strong African Coast Fever.
22-10-03	Horse, Smears, Heart & Lungs	,, Webb ..	Military	—	Barberton ..	Putrid infection of lungs.
,,	Sick Cow.. ..	,, May ..	Repatriation ..	—	Nylstroom ..	Nothing seen.
23-10-03	Oxen, 2	Inspector Lester ..	Native, William..	Town Lands ..	Lydenburg ..	,, ,,
,,	Ox	—	F. McLaghlan ..	—	,, ..	,, ,,
24-10-03	,, ...	D.V.S. Edgar ..	P.W.D.	—	Pietersburg..	,, ,,
28-10-03	,, ...	,, Watkin ..	M. Tina	Buffelspruit, 35..	Ermelo ..	,, ,,
,,	,, ...	Lieut.-Col. Flintoff	—	—	O.R. Colony .	Blood hæmolytic infected.
,,	Cow	Inspector Lester ..	Native, William	Town Lands ..	Lydenburg ..	Slight African Coast Fever.
29-10-03	Ox	D.V.S. Edgar ..	—	Palmietgat, 645..	Pietersburg..	Strong infection Ordinary Redwater and African Coast Fever.
,,	Cows, 2, Smears & Scab Scraping	,, Lindsay ..	J. Steyn	Vyffontein, 3 ..	Heidelberg ..	Acariosis found, no Piroplasma.
30-10-03	Ox	,, Garraway ..	J. J. Fourie ..	Wildebeeste Hoek	Pretoria ..	Nothing seen.
,,	,, ...	,, May ..	—	Rietfontein, 1428	Waterberg, Nylstroom	Doubtful, smears bad.
,,	Oxen, 2	Inspector Lester ..	J. Breitenbach ..	Vlakfontein ..	Lydenburg ..	Nothing seen.
			Mr. Oliver ..	Kruger's Post ..	,, ..	,, ,,
,,	Ox	D.V.S. Turnbull ..	Unknown, found	wandering by Police	Machadodorp	Strong African Coast Fever.

,,	Oxen, 2	,, Webb ..	Lotasha's Kraal..	Outbreak, 38, Moodie's Concession.	Barberton ..	,, ,, ,, ,,
,,	Ox, 1	,, ,, ..	Ginde's Kraal ..	Continia Block, 1159, 1161	,, ..	,, ,, ,, ,,

Numbers examined for October, 1903, 82.

NOVEMBER, 1903.

2-11-03	Oxen, 4, B1, B2, B3, and A1	D.V.S. Elder. ..	A. McCorkindale	Inidgen	Horo, Embabaan.	Nothing seen in any.
,,	2 Sick Cows ..	,, Webb ..	Ingoni's Kraal ..	Fig Tree Creek, South of Sheba Battery	Barberton ..	Strong African Coast Fever.
3-11-03	Dog	,, Cochrane ..	—	—	Middelburg..	Nothing seen.
,,	Ox	,, May ..	—	Rietfontein, 1428	Nylstroom ..	African Coast Fever.
4-11-03	,, ...	,, Sturge ..	V. d. Westhuizen	Arnotstad, 544 ..	Rustenburg	Nothing seen.
,,	Oxen, 4	A. Holm	Experimental Farm	—	Potchefstroom	,, ,,
,,	Ox, Heart, Kidney, Liver and Spleen	D.V.S. Dale ..	Newington ..	Buffelskloof, 24..	,,	,, ,,
,,	Ox	Inspector Lester ..	F. McLaghlan ..	—	Lydenburg ..	,, ,,
,,	,, ...	D.V.S. Garraway ..	Mr. Carsters ..	Parktown, Wonderboom	Pretoria ..	Blood hæmolytic.
,,	Bull 1, Cow, Heart, Kidney, & Spleen	,, ,, ..	A. A. Struben ..	136, V. d. Walt Street	,, ..	1, Bull, nothing seen; 2, Cow, Ordinary Redwater.
7-11-03	Ox	Inspector Lester ..	Mr. Olivier ..	Kruger's Post, 1193	Lydenburg ..	Nothing seen.
9-11-03	Sick Cow.. ..	D.V.S. Dunphy ..	R. Thompson ..	One Tree Farm..	Krugersdorp.	,, ,,
,,	Pipettes, Pus, 3, 4,5	,, Turnbull ..	Repatriation ..	—	Machadodorp	All Saccharomy. Farcimi.
10-11-03	Ox	,, Edgar ..	A. H. du Preez ..	Verzamen Hoek..	Pietersburg..	Strong African Coast Fever.
,,	Piece of Kidney..	,, Dunphy ..	—	—	Krugersdorp.	Nothing seen.
11-11-03	Ox	,, Bush ..	H. Rabe	Swartwater, 41 ..	Piet Retief ..	African Coast Fever.
,,	,, ...	,, May ..	Mr. Walker ..	Springbok Flats..	Nylstroom ..	Nothing seen.
12-11-03	Calves, 2.. ..	,, Dale ..	Experimental Farm	—	Potchefstroom	,, ,,
,,	Sick Cow.. ..	,, Webb ..	S. Wilson ..	Gullus River ..	Barberton. ..	African Coast Fever.
,,	Cow	,, Garraway ..	Mrs. Parker ..	188, Struben Street	Pretoria ..	Doubtful.
,,	Ox	,, Edgar ..	Mr. Pretorius ..	Zand River, Marabastad	Pietersburg..	Strong African Coast Fever.

NOVEMBER, 1903.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
13-11-03	Ox	D.V S. Turnbull ..	—	—	Machadodorp	Nothing seen.
,,	,, ...	,, Edgar ..	S.A.C.	—	Pietersburg..	,, ,,
15-11-03	Scraping of Pony	,, Dunphy ..	Broude Luff and Smith	—	Krugersdorp.	,, ,,
16-11-03	1, Sick Calf; 2, Pipettes, Pus	,, Webb ..	Barrett's Berlin G.M. Co., Mr. Davidson	Barrett's Berlin G.M. Co.	Barberton ..	1, African Coast Fever; 2, nothing seen.
,,	Smears, Mare	,, Conacher ..	Government ..	Horse Farm ..	Lake Chrissie	Nothing seen.
,,	Ox	,, Turnbull ..	A. Coetzee ..	Hartebeestespruit, 352	Machadodorp	,, ,,
,,	,, ...	,, May ..	Mr. Daly.. ..	Waterberg ..	Nylstroom ..	Strong African Coast Fever.
17-11-03	,, ...	P.V.S.	Mr. Carstens ..	Parktown ..	Pretoria ..	African Coast Fever.
18-11-03	,, ...	D.V.S. Edgar ..	P. Vogel.. ..	Vernamens Hoek	Pietersburg..	Hæmolytic infection.
,,	,, ...	Capt. Christian ..	Repatriation ..	Finks Zyn Drift..	Pretoria ..	Nothing seen.
19-11-03	,, ...	D.V.S. May ..	Mr. Johnston ..	Belvedere ..	Nylstroom ..	,, ,,
21-11-03	Kidney	—	Mr. Malan ..	Hartebeeste Hoek, Horn's Neck	—	African Coast Fever.
23-11-03	Smears, 9.. ..	D.V.S. Elder ..	S.A.C.	—	Embabaan ..	Nothing seen.
,,	A1, A2	,, ,, ..	A. McCorkindale	—	,, ..	,, ,,
,,	Ox	,, Lindsay ..	S.A.C.	Hamman's Kraal	Pretoria ..	,, ,,
,,	Madagascar Ox..	—	—	—	Machadodorp	Strong African Coast Fever.
25-11-03	Oxen, 11 ..	A. Holm	Government ..	Experimental ..	Potchefstroom	Reported elsewhere.
26-11-03	Ox, Dead, ..	D.V.S. Turnbull ..	C. Potgieter ..	Gelok, 29.. ..	Machadodorp	African Coast Fever.
	Mule, ,,					
27-11-03	Smears, 12 ..	,, Garraway ..	Government ..	Experimental ..	Potchefstroom	Reported elsewhere.
,,	Ox	,, Lindsay ..	A. G. Hermann..	Krelings Post, 111	Pretoria ..	Nothing seen.
,,	Smears, Pus, Matter, etc., 1, 2, 3, 4, 5	Capt. Blackburn ..	Government ..	Horse Farm ..	Standerton ..	No. 5, Saccharomy. Farcimi; 1, 2, 3, and 4, nothing seen.
28-11-03	Oxen, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10	D.V.S. Cochrane ..	Land Settlement	Stoffberg's ..	Middelburg..	1, 9, 10, nothing seen; 2, 3, 4, 5, 6, 7, 8, African Coast Fever.

,,	Oxen, 2, 7, 12, 17, and 19	,, Garraway ..	Government ..	Experimental ..	Potchefstroom	Reported elsewhere.
28-11-03	Oxen, 3	Inspector Lester ..	Swaartbooi, Native	Vraainitricht, 1213	Lydenburg ..	Smears broken and useless.
30-11-03	Ox	D.V.S. Sturge ..	Capt. Johnston ..	Zuurplate, 822 ..	Rustenburg..	Nothing seen.
,,	,, ...	,, Conacher ..	Korain, Native ..	London, 1220 ..	Lydenburg (North of Pilgrim'sRust)	Strong African Coast Fever.

Number examined for November, 1903, 110.

DECEMBER, 1903.

1-12-03	Pus, Mule ..	D.V.S. Walker ..	Repatriation ..	—	Carolina ..	Strong Saccharomy. Farcimi.
,,	Blood Smear, Ox	,, Lindsay ..	Mrs. McRae ..	Mountain View..	Pretoria ..	Nothing seen.
2-12-03	Ox	,, Conacher ..	—	Wiunars Poort ..	Machadodorp	African Coast Fever.
,,	,, Smears, 34 ..	,, Garraway ..	Government ..	Experimental ..	Potchefstroom	Notes elsewhere.
3-12-03	Oxen, 1, 2, 3, 4 ..	,, Conacher ..	Land Settlement	Near Wonderf'n.	Middelburg..	Nothing seen.
,,	Ox	,, Conacher ..	—	Wiunars Poort, 1403	Machadodorp	Strong African Coast Fever.
5-12-03	Oxen, unmarked, 3	,, Edgar ..	Van Smallen ..	—	Pietersburg..	All slight African Coast Fever.
6-12-03	Ox	,, Lindsay ..	M. N. Rickert ..	Schoeman's Rust, 498	Pretoria ..	Nothing seen.
,,	,, ...	,, May ..	Native	Cypherfontein,2156	Nylstroom ..	,, ,,
7-12-03	Lungs and Stomach of Horse	C. H. Blackburn ..	Government ..	Horse Station ..	Standerton ..	Emphysema in lung; bot ulcers in stomach.
8-12-03	Ox	D.V.S. May ..	Dr. Hicky ..	Elandspoort ..	Nylstroom ..	Strong African Coast Fever.
9-12-03	Sick Cow.. ..	,, Lindsay ..	Lucas (Native) ..	Roodekoppies, 132	Pretoria ..	Nothing seen.
10-12-03	Ox	Inspector Dunkley	—	Driefontein, Kookfontein	Boetsop, Cape Colony	Nothing seen.
12-12-03	,, ...	,, Lester ..	P. H. Ferreira ..	Magnet Height C.M. Co.	Lydenburg ..	,, ,,
12-12-03	Ox, Spleen & Heart	D.V.S. Chalmers ..	Kahn (Native) ..	Klipportje, 228 ..	Heidelberg ..	Too decomposed to diagnose.
,,	Oxen. 1, 2, 3 ..	,, Turnbull ..	Mazaba, Umbeila, Umgoona (Natives)	Hilversum, 998 ..	Lydenburg ..	1 and 3, nothing seen; 2, African Coast Fever.
14-12-03	Cow	,, Conacher ..	—	Schoengezicht, 38	Machadodorp	Nothing seen.
,,	Ox, Kidney and Heart	,, Elder ..	Commissioner for Swaziland	—	Embabaan ..	Smears useless; no corpuscles seen.

DECEMBER, 1903.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
17–12–03	Horse, Spleen ..	V.S. S. Jackson ..	Repatriation ..	—	Pretoria ..	Biliary Fever.
,,	Ox	D.V.S. Sturge ..	Mr. Peacock ..	Burfontein, 432..	Rustenburg	Nothing seen.
18–12–03	Horse, Lung ..	,, Walker ..	—	—	Ermelo ..	No glanders.
,,	Ox (suspect Quarter Evil)	,, Edgar ..	—	—	Pietersburg..	Nothing seen.
,,	Cow	,, Lindsay ..	H. C. Penzhorn..	Corner Walker and Mears Streets, Sunnyside	Pretoria ..	,, ,,
19–12–03	Ox	,, Garraway ..	P. Smidt.. ..	Elandsfontein, 374	,, ..	,, ,,
,,	Sick Cow.. ..	,, May ..	Christian (Native)	Platannesfontein, 1171	Nylstroom ..	,, ,,
20–12–03	Ox (suspect Gall-sickness)	,, Cochrane ..	Repatriation ..	—	Middelburg..	1, nothing seen; 2, African Coast Fever and Ordinary Redwater.
	2 Oxen (suspect Redwater)		2. —			
21–12–03	Sick Ox	,, May ..	Mr. Montgomery	Groot Vlei ..	Waterberg, Nylstroom	Doubtful, smears not good.
,,	Oxen, 2	,, Cochrane ..	Land Settlement	Quarantine ..	Middelburg..	Very strong double infection.
,,	Scraping, Sheep..	,, Chalmers ..	Major Morley ..	Beerlaagte, 256 ..	Heidelberg ..	Nothing seen.
22–12–03	Sick Ox	,, May ..	Mr. Benardi ..	Bosch Hoek, 349	Nylstroom ..	Trypanosoma.
23–12–03	Smear, Mule ..	,, Conacher ..	—	Pilgrims Rest ..	Machadodorp	Suspected Biliary Fever.
24–12–03	Heart and Spleen of Ox	Inspector O'Connor	Stury Berg ..	Hackplaats ..	Pretoria ..	Too decomposed to diagnose.
,,	1, Ox	D.V.S. May ..	P. Potgieter ..	—	—	1, nothing seen;
	2, Dead Ox ..	—	Repatriation ..	Roodepoort, 2148	Nylstroom ..	2, ,, ,,
28–12–03	Ox	,, Garraway ..	Mr. Friedman ..	118, Minnaar St.	Pretoria ..	Ordinary Redwater.
,,	,, ...	,, Pollard ..	Louisa (Native)..	Velgearnden, 323	Wakkerstroom	Nothing seen.
,,	1, Ox, 435; 2, Black Ox, 567	,, Walker ..	S.A.C.	Carolina	Ermelo ..	1, Nothing seen; 2, ,, ,,
,,	Pus, Bay Gelding	,, ,, ..	,, ...	,, ..	,, ..	Saccharomy. Farcimi.
30–12–03	Oxen, 13, 7, 17, 19, 12, 19, and 17	General Manager ..	Government ..	Experimental ..	Potchefstroom	Notes elsewhere.

Number examined for December, 1903, 91.

2-1-04	Oxen, 3	D.V.S. Webb ..	1, P. Vosloo .. 2, April (Native)	1, Doornhoek, 1029 2, Grobersvilde, 1115	Lydenburg .. —	1, African Coast Fever ; 2, basic and nucl. cells.
3-1-04	Ox	Inspector O'Connor	Mr. Behrens ..	Daspoort	Pretoria ..	African Coast Fever.
4-1-04	Oxen, 7	Manager .	Government ..	Experimental ..	Potchefstroom	Reported elsewhere.
6-1-04	Ox	D.V.S. Garraway ..	J. Steinberg ..	Hoekplaats ..	Pretoria ..	Nothing seen.
,,	Dog	P.V.S.	—	—	,, ..	,, ,, (decomposed).
,,	Ox	D.V.S. Walker ..	—	—	Ermelo ..	,, ,, (smears bad).
,,	Horse	Manager	Government ..	Horse Farm ..	Standerton ..	,, ,,
7-1-04	Oxen, 2	D.V.S. May ..	Repatriation ..	Rhenosterpoort ..	Waterberg ..	,, ,, (smears bad.)
9-1-04	Ox	,, Turnbull ..	Somdite (Native)	Morsy, 1055 ..	Barberton ..	African Coast Fever.
,,	Oxen, 3	Inspector O'Connor	Repatriation ..	Finks Zyn Drift ..	Pretoria ..	Nothing seen.
11-1-04	Ox	,, Lester ..	J. Viljoen ..	Zwagershoek, 1448	Lydenburg ..	African Coast Fever.
12-1-04	Cow	,, O'Connor	Mrs. Schweizer ..	Sunnyside ..	Pretoria ..	Ordinary Redwater and basic cells.
,,	Bull, Dead ..	,, ,,	H. Cavanagh ..	Daspoort Quarries	,, ..	African Coast Fever.
13-1-04	Oxen, 4	A. H. Hilliard ..	Repatriation ..	Rhenosterhoek ..	Nylstroom ..	Nothing seen.
,,	Ox	D.V.S. Bush ..	M'Zeme (Native)	Zandhoek, 100 ..	Piet Retief ..	,, ,,
,,	Blood of Pig ..	,, Dunphy ..	—	—	Krugersdorp.	,, ,,
,,	Ox, Heart, Kidney, Liver, & Spleen	Inspector O'Connor	E. Meintjes ..	Ouderstepoort ..	Pretoria ..	African Coast Fever.
,,	Calf	,, Lester ..	Mr. McGee ..	Sterkspruit, 159 ..	Lydenburg ..	Nothing seen, smears decomposed.
14-1-04	,, Dead ..	,, O'Connor	Mr. Sarel Eloff ..	Daspoort	Pretoria ..	African Coast Fever and Ordinary Redwater.
,,	Oxen, 3	V.S. S. Jackson ..	Repatriation ..	Rhenosterhoek ..	—	1 and 2, nothing seen ; 3, basic cells.
,,	Lung from Lymphangitic Horse	P.V.O. Smith ..	Veterinary Hospital	—	Standerton ..	Doubtful.
15-1-04	Heifer	D.V.S. Dale ..	S.A.C.	Fredericstad ..	Potchefstroom	Nothing seen.
,,	Cattle, 4 ; Ox, 3 Cows	Inspector Lester ..	1, Mr. Oliver .. 2, Mrs. Vosloo .. 3, Mr. Lombard .. 4, Mr. Van Achterberg	Kruger's Post, 1193 Klipfontein .. Rooidraai, 1242 .. Town Lands ..	Lydenburg .. ,, .. ,, .. ,, ..	,, ,, ,, ,, African Coast Fever. ,, ,, ,,

JANUARY, 1904.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
15-1-04	Cow, Heart, Kidneys, Liver, and Spleen	Inspector O'Connor	G. A. Roth ..	Proes Street ..	Pretoria ..	Ordinary Redwater.
„	Ox, Heart, Kidneys, Liver, and Spleen	„ „	F. Esterhuizen ..	Brick Yard, Artillery Barracks	„ ..	Nothing seen, decomposed.
16-1-04	Ox, Liver and Kidneys	D.V.S. Cochrane ..	—	Town Lands ..	Middelburg..	Nothing seen.
18-1-04	Sick Cow, Dead Ox	„ Webb ..	Jacob (Native) ..	Bushbuch Bridge	Lydenburg ..	Both African Coast Fever.
19-1-04	Ox	„ Cochrane ..	—	Doornboom, 480	Middelburg..	Nothing seen.
„	„	„ Turnbull ..	Jonas (Native) ..	Winter Farm, Kaapsche Hoop	Barberton ..	Basic cells numerous.
„	„	„ Garraway ..	David (Native) ..	Swaartkoppies, 269	Pretoria ..	No corpuscles, decomposed.
20-1-04	„	„ Edgar ..	Mr. Vine	Entselbosch ..	Pietersburg..	Nothing seen.
„	„	Inspector Lester ..	Mr. H. Coster ..	Sterkspruit, 159..	Lydenburg ..	African Coast Fever.
21-1-04	„ Blood ..	J. F. Scott.. ..	—	39, Kerk Street..	Johannesburg	Ordinary Redwater.
„	Mule	—	Municipal Stables	—	Pretoria ..	Saccharomy. Farcimi.
22-1-04	Calf	V.S. Verney ..	—	—	Natal ..	Basic cells numerous.
„	Oxen, 2	D.V.S. Garraway ..	Mrs. Grobelar ..	Christoffel Street, 1490	Pretoria ..	1, bull, African Coast Fever; 2, ox, African Coast Fever.
23-1-04	Ox	„ Edgar ..	Mr. Jansen ..	Chumiespoort East	Pietersburg..	African Coast Fever,
„	Horse, Pus ..	„ Webb ..	S.A.C.	Pilgrims Rest ..	Lydenburg ..	Saccharomy. Farcimi.
23-1-04	Cow	Inspector Lester ..	August (Native)..	Ohrigstad ..	„ ..	African Coast Fever.
25-1-04	Cow	V.S. Robinson ..	—	—	Pretoria ..	Nothing seen.
26-1-04	Oxen, 3	Inspector O'Connor	Mouson's Mission Station	Koedoespoort ..	„ ..	1, Nothing seen.
			„	„ ..	„ ..	2, „ „
			Romijn	Sunnyside ..	„ ..	3, Ordinary Redwater.
„	Cow	D.V.S. Sturge ..	Jonas (Native) ..	Kaffir Location..	Rustenburg	Nothing seen.
27-1-04	Ox	„ Cochrane ..	Mr. Bosman ..	—	Middelburg..	„ „
28-1-04	Oxen, 4; Horse, 1	„ Walker ..	—	—	Ermelo ..	Oxen all showed African Coast Fever; horse, Saccharomy. Farcimi.

,,	Horse, Smears ..	Land Department..	—	—	Pretoria ..	Nothing seen.
,,	Ox	D.V.S. Sturge ..	P. Brink.. ..	—	Rustenburg	African Coast Fever.
30–1–04	,, ...	Inspector O'Connor	Kasson (Native)..	Koedoespoort, Keleton	Pretoria ..	Nothing seen.

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FEBRUARY, 1904.

1–2–04	Oxen, 2	Inspector Lester ..	A. Coetser ..	Kleinplaats ..	Lydenburg ..	Both African Coast Fever.
,,	Heifer	D.V.S. Turnbull ..	Dinga Ding(Native)	Simali Valley ..	Barberton ..	Nothing seen.
2–2–04	Ox	,, Sturge ..	Mr. Cox	Waterval, 544 ..	Rustenburg	African Coast Fever.
,,	,, ...	,, Garraway ..	Mr. Ryder ..	—	Pretoria N...	Nothing seen.
3–2–04	Oxen, 3	,, May ..	Repatriation ..	Rietspruit, 305 ..	Nylstroom ..	,, ,,
,,	,, 5	,, Cochrane ..	—	Botshabilo ..	Middelburg..	1, strong African Coast Fever ; 2, African Coast Fever ; 3, 4 and 5, basic cells.
,,	,, 2	,, ,, ..	1, Mr. Herbst ..	Blaauwbank, 35..	,, ..	Nothing seen.
			2, Mr. Strydom ..	Dunkerkrantz, 104	—	,, ,,
4–2–04	Cow	Inspector O'Connor	H. Marais ..	Meintjes' Kop ..	Pretoria ..	,, ,,
6–2–04	Ox (bottle of Blood)	,, ,,	—	—	,, ..	,, ,,(blood hæmolytic)
8–2–04	Oxen, 4	D.V.S. Edgar ..	1 — ..	Maschappa, 865.	Pietersburg..	All four African Coast Fever.
			2, Mrs. Scheepers	Verzaman Hoek,		
			3 — ..	Elim,		
			4, Mrs. Coolssley	Vleifontein, 338		
,,	,, 3	,, Garraway ..	1, A. J. van Gass	Ouderstepoort ..	Pretoria ..	1, nothing seen, decomposed.
			2, J. J. Horn ..	Sheilfontein ..	,, ..	2, ,, ,,
			3, Mrs. Jamison..	Ouderstepoort ..	,, ..	3, African Coast Fever.
,,	Bull	,, Turnbull ..	Umgoona (Native)	Hill Top	Barberton ..	African Coast Fever.
,,	Cow	,, Sturge ..	Petrus (Native)..	Turffontein, 394..	Rustenburg..	,, ,, ,,
,,	Ox	Inspector Lester ..	S. v. d. Merwe ..	Spitzkop, 802 ..	Lydenburg ..	,, ,, ,,
,,	Oxen, 3	D.V.S. Edgar ..	1, Jim (Native)..	Elim Mission,	Pietersburg..	All smears decomposed.
			2, Mrs. Sheil ..	Rossbach,		
			3, Mr. Grey ..	Machappies		
9–2–04	Cow	,, Sturge ..	W. J. Dane ..	—	Rustenburg	Nothing seen.
10–2–04	Oxen, 3	,, ,, ..	1, C. Kleyne ..	—	,, ..	1, ,, ,,
			2, J. Kruger ..	Waterkloof ..	,, ..	2, ,, ,,
			3, B. Cornelius ..	—	,, ..	3, ,, ,, decomposed.

FEBRUARY, 1904.—*(continued*

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
10-2-04	Oxen, 5	D.V.S. Cochrane ..	Land Settlement	Avantuur, 135 ..	Middelburg..	Nothing seen.
,,	Calf, Sick ..	,, Webb ..	Swatkop Kraal ..	Vrainitzicht, 1213	Lydenburg ..	,, ,,
	Cow, ,, ..	,,	J. Byrne.. ..	Boerboom Kraal	,, ..	,, ,,
,,	Sheep, Scraping..	,, Chalmers ..	A. Doig	Koppies Kraal ..	Heidelberg ..	,, ,,
11-2-04	Oxen, 5	,, Cochrane ..	—	Botshabilo ..	Middelburg..	Nothing seen in any.
,,	Cow	,, Edgar ..	A.R.M.	—	Pietersburg..	African Coast Fever.
,,	Pipette Pus, Mule	,, Dunphy ..	R. Morkel & Co...	—	Krugersdorp.	Saccharomy. Farcimi.
12-2-04	1, Cow ; 2, Ox	Station	Mr. Reid.. ..	Daspoort.. ..	Pretoria ..	1, nothing seen ; 2, African Coast Fever.
13-2-04	Ox, Dead ..	Inspector Paton ..	J. Bellnin ..	Geluk	Machadodorp	Ordinary Redwater.
15-2-04	,, ...	D.V.S. Walker ..	M'Gebo (Native)	Buffelspruit, 35..	Carolina ..	Nothing seen (decomposed).
16-2-04	Cow	,, Elder ..	Natives	—	Embabaan ..	Nothing seen.
,,	Ox, Kidney ..	,, Webb ..	—	—	Lydenburg ..	Pyæmic Melaslasis of Kidney.
17-2-04	Pig, Smears ..	,, Chalmers ..	—	—	Heidelberg ..	Nothing seen.
18-2-04	Oxen, 2	,, Edgar ..	1, A. Kruger ..	Myngenkg, 623 ..	Pietersburg..	Both strong combined African Coast Fever and Ordinary Redwater.
			2, T. Venter ..	Onverwaacht, 352		
,,	Ox	,, May ..	—	Rietspruit ..	Nylstroom ..	African Coast Fever.
,,	,, Dead ..	,, Sturge ..	Land Board ..	Town Lands ..	Rustenburg	,, ,, ,,
,,	Heifer	,, Cochrane ..	—	Rooikop, 134 ..	Middelburg..	Nothing seen.
19-2-04	Ox, Heart, Kidney, and Spleen	,, Garraway ..	E. Sussens ..	Riveria	Pertoria ..	African Coast Fever.
20-2-04	,, ,,	Inspector Paton ..	C. Potgieter ..	Geluk	Machadodorp	Nothing seen.
,,	Oxen, 2	D.V.S. Pollard ..	Repatriation ..	Middelspruit, 33..	Wakkerstroom	Nothing seen in either.
22-2-04	Cow	Inspector O'Connor	Mr. Snyman ..	Burgher-rights ..	Pretoria ..	African Coast Fever.
,,	Pipette Pus ..	D.V.S. Cannon ..	—	—	Standerton ..	Nothing seen.
23-2-04	Horse, Lung ..	Major Richardson..	Veterinary Hospital No. 5	—	Bloemfontein	,, ,,
,,	Ox	Inspector Paton ..	C. Potgieter ..	Geluk	Machadodorp	African Coast Fever.
,,	,, ...	Assistant P.V.S. ..	—	Onderstepoort ..	Pretoria ..	,, ,, ,,
,,	Bull	D.V.S. Turnbull ..	Spon (Native) ..	Grobler's Farm ..	Nelspruit, Barberton	,, ,, ,,

24-2-04	Cow	„ Webb ..	Mr. Webb ..	National Bank ..	Lydenburg ..	Strong African Coast Fever and Ordinary Redwater.
„	„	„ Garraway ..	Mr. Romijn ..	Erf 176, Sunnyside	Pretoria ..	Doubtful.
„	Ox	Inspector Paton ..	J. Button ..	Doornhoek, 199..	Machadodorp	Nothing seen.
25-2-04	Oxen, 3	D.V.S. Elder ..	—	—	Embabaan ..	„ „
„	„ 2	„ Lindsay ..	1, P. Strydom ..	Dinkerkrantz ..	Middelburg..	Both decomposed.
			2, N. Swartz ..	Paardekloof, 99		
26-2-04	Ox, Dead ..	Inspector O'Connor	C. B. Dyason ..	Berea Park ..	Pretoria ..	African Coast Fever.
27-2-04	„	„ Lester ..	Mr. Dunedin ..	Town Lands ..	Pietersburg..	Nothing seen.
29-2-04	Cow	„ O'Connor	Mr. Romijn ..	Sunnyside ..	Pretoria ..	African Coast Fever.

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MARCH, 1904.

1-3-04	Ox	D.V.S. Sturge ..	P. Muller ..	Krondal, 117 ..	Rustenburg	African Coast Fever.
„	„ Lung ..	—	—	—	Johannesburg	Pleuro-pneumonia.
„	Sheep, Scraping..	D.V.S. Chalmers ..	—	—	Heidelberg ..	Nothing seen.
„	Ox, Kidney ..	Inspector O'Connor	United Butchery	—	Pretoria ..	African Coast Fever.
2-3-04	Cows, 2	D.V.S. Garraway ..	A. Hulsenbach ..	103, Schoeman St.	„ ..	1, Nothing seen; 2, African Coast Fever.
3-3-04	Oxen, 7	„ Lindsay ..	Messrs. Ruthven and Compton	Longkloof Onder	Middelburg..	Both African Coast Fever.
		„ „ ..	Swaartboy ..	Paardekloof, 99..	„ ..	Decomposed.
		„ „ ..	A. v. d. List ..	„ ..	„ ..	„
		„ „ ..	Titus (Native) ..	Leuwfontein, 174	„ ..	Nothing seen.
		„ „ ..	Seton „ ..	Kaffirskraal, 62..	„ ..	African Coast Fever.
„	Ox	„ Edgar ..	Native	Ongegond, 2080..	Pietersburg..	Combined infection, African Coast Fever and Ordinary Redwater.
4-3-04	Oxen, 2	„ Garraway ..	S.A.C.	Zandfontein, 26..	Pienaar'sRiver, Pretoria	African Coast Fever.
		„ „ ..	J. Orken.. ..	15, Scheiding St.	Pretoria ..	Nothing seen.
„	Ox	P.V.S.	Blood used by Roux	Onderstepoort ..	„ ..	„ „
„	—	D.V.S. Sturge ..	C. Mohl	Kronval, 117 ..	Rustenburg	„ „
7-3-04	Ox	„ „ ..	W. Fouche ..	Commissie Drift..	„ ..	„ „

MARCH, 1904.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
7-3-04	Horse	D.V.S. Edgar ..	Mr. Zeederberg ..	Haenertsburg ..	Pietersburg..	Nothing seen.
,,	Cow, Dead ..	R. H. Wilson, per Captain Stubbs, S.A.C.	—	—	—	,, ,, (decomposed).
,,	Ox, Dead ..	D.V.S. Garraway ..	P. Erasmus ..	Wonderboom, 311	Pretoria ..	,, ,, ,,
8-3-04	,, ...	,, Chalmers ..	Messrs. Oschri and Koseff	Vlakfontein, 101	Heidelberg ..	,, ,,
,,	Cow, Smear of Spleen	,, Dale ..	Government ..	Experimental ..	Potchefstroom	,, ,,
,,	Cow, Heart, Kidney, Liver, and Spleen	Inspector O'Connor	Mr. Struben ..	V. d. Walt Street	Pretoria ..	Ordinary Redwater.
,,	Cow	D.V.S. Garraway ..	Mr. Nel	5, Vermeulen St.	,, ..	,, ,,
,,	Bull	,, ,, ..	Agricultural Department	Dynamite Factory, Modderfontein	,, ..	Basic cells and Ordinary Redwater.
9-3-04	Ox, Dead ..	,, May ..	Mr. De Vos ..	Bospoort, 2151 ..	Nylstroom ..	Nothing seen.
10-3-04	Oxen	,, Sturge ..	Mr. Nortje ..	Bestershoek, 370	Rustenburg	African Coast Fever.
,,	,, ...	,, ,, ..	P.W.D.	—	,, ..	,, ,, ,,
,,	Bay Mare ..	,, Cannon ..	Government ..	Stud Farm ..	Standerton ..	Decomposed.
11-3-04	Tumours from Scrotum of Horse	,, Edgar ..	Native Affairs Department	—	Pietersburg..	Nothing seen.
,,	Cow	,, Garraway ..	Mr. Rowland ..	5, St. Andries St.	Pretoria ..	,, ,,
12-3-04	Ox, Dead ..	,, Lindsay ..	W. Strydom ..	Rondebosch, 178	Middelburg..	African Coast Fever.
,,	Pus from Cow Udder	Inspector Lester ..	Mrs. Smullans ..	Town Lands ..	Pietersburg..	Streptocollus infection.
14-3-04	Ox	D.V.S. Lindsay ..	Silas (Native) ..	Draaihoek, 470 ..	Middelburg..	Nothing seen.
,,	Oxen, 2	,, Sturge ..	E. Denny Smit ..	Paardekraal, 388 —	Rustenburg ,, ..	,, ,, Doubtful, probably African Coast Fever.
15-3-04	Donkeys, 3 ..	,, Turnbull ..	A. Settler ..	Land Settlement	Queen's R., Barberton	1, Doubtful; 2 and 3, piroplasma.
,,	Cow	,, Edgar ..	Mr. Richter ..	TownLands ..	Pietersburg..	Blood contains rings.
16-3-04	Ox	,, Garraway ..	Government ..	Groenkloof, 419..	Pretoria ..	Nothing seen.

,,	Human Smears ..	H. Altenroxel ..	—	Haenertsburg ..	Pietersburg..	1, White man, tropical rings ; 2, Kaffir, nothing seen ; 3, Coolie, tropical rings.
,,	Smears, Cow ..	—	Box 1,080 ..	Norwood ..	Johannesburg	Basic cells numerous.
17-3-04	Ox	Inspector Paton ..	Native	Schoengezicht ..	Machadodorp	Nothing seen.
18-3-04	,, ...	,, Nicholls	E. Patlankky ..	—	Vereeniging	Basic and nucl. cells numerous.
,,	Sick Cow.. ..	,, O'Connor	J. J. Marais ..	Pretorius & Johann Streets	Pretoria ..	Nothing seen.
,,	Ox	D.V.S. May ..	—	Springbok Flats..	Nylstroom ..	,, ,,
21-3-04	,, ...	,, Sturge ..	Mr. Latagan ..	—	Rustenburg	,, ,,
21-3-04	,, ...	,, Garraway ..	Native	Mr. Englehecht's, Rietfontein, 280	Pretoria ..	African Coast Fever.
,,	Clydesdale Mare..	—	Government ..	Horse Farm ..	Standerton ..	Nothing seen.
22-3-04	Bottle Ox Blood..	J. C. Rous.. ..	—	Pyramids ..	Pretoria ..	African Coast Fever.
23-3-04	Heart and Kidney, Horse	Capt. Todd.. ..	Military	Veterinary Hospital, No. 1	Pretoria ..	Gastric and Cardiac Horse sickness.
,,	Lung, Ox ..	Mr. Peddie.. ..	Municipality ..	—	Johannesburg	Nothing seen.
,,	Ox	Assistant P.V.S. ..	A. E. Henry ..	Lt. D.C. XII S.D.	Belfast ..	Smear too thick ; may be Ordinary Redwater.
24-3-04	Horse, Smear ..	D.V.S. Edgar ..	Mr. Zeederberg ..	Haenertsburg ..	Pietersburg..	Nothing seen.
,,	Oxen, 5	,, Lindsay ..	V.S. Crawford ..	Repatriation ..	Middelburg..	1 and 2, nothing seen ;
			S.A.C.	—	,,	3, smear too thick :
			Mr. van Edveld ..	Toeroegat, 89 ..	,,	4, African Coast Fever ;
			Swartboy (Native)	Paardekloof	,,	5, African Coast Fever.
25-3-04	Ox, Dead ..	Inspector O'Connor	G. Kinnear ..	Brickfields ..	Pretoria ..	African Coast Fever.
,,	Calf, Heart and Lung	D.V.S. Chalmers ..	—	—	Heidelberg ..	Nothing seen.
26-3-04	Oxen, 2	,, Lindsay ..	Kleinboy (Native)	Rietfontein, 451, Pokwani	Middelburg..	1, live ox, ring forms ;
					,, ..	2, dead ox, decomposed.
28-3-04	Human Smear ..	Dr Sanders.. ..	—	—	Pretoria ..	Basic cells.
,,	Ox, Smear ..	D.V.S. Dale ..	—	—	Potchefstroom	Anthrax.
,,	,, ,, ..	V.S. Robinson ..	—	—	Pretoria ..	Nothing seen.
,,	,, ...	P.V.S.	M. Jamieson ..	Onderstepoort ..	,, ..	Rings and bacillary forms (not African Coast Fever).
29-3-04	Lung and Pleura, Bay Gelding	D.V.S. Walker ..	C. v. Jeyso ..	—	Ermelo ..	Nothing seen.
,,	Ox	,, ,, ..	M'Anjan (Native)	Glenmore, 222, Mayflower Post	,, ..	,, ,,

Number examined for March, 1904, 74.

APRIL, 1904.

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
2-4-04	Ox, Dead ..	D.V.S. Sturge ..	P. v. Rooyen ..	Modderfontein, 247	—	African Coast Fever.
,,	Cow, Sick ..	,, Lindsay ..	P. Bosman ..	Erf 396	Middelburg..	Ring forms, basic cells, African Coast Fever.
,,	Ox	Inspector Paton ..	C. Mare	Minnaarsp'rt, 1403	—	African Coast Fever.
5-4-04	Bull	D.V.S. Turnbull ..	Mr. Scholtz ..	Nelspruit ..	Barberton ..	,, ,, ,,
,,	Donkeys, 2 ..	,, Garraway ..	Government ..	Zandfontein, 548	Pretoria ..	Biliary Fever.
,,	Cow	,, ,, ..	Mr. Benson ..	Pretoria	,, ..	African Coast Fever.
6-4-04	Ox, Dead ..	,, Lindsay ..	Mr. Swart ..	Paardekloof ..	Middelburg..	Smears broken.
8-4-04	Cow	,, Garraway ..	Dr. v. Wyk ..	Pretoria	Pretoria ..	Nothing seen.
9-4-04	Bull	,, Chalmers ..	Government ..	Experimental ..	Heidleberg ..	,, ,,
10-4-04	Cow	,, Lindsay ..	—	Klipplaats Drift..	Middelburg..	,, ,,
11-4-04	Ox	,, Elder ..	—	Embabaan ..	Swaziland ..	,, ,,
,,	,, ...	,, Garraway ..	Jan (Native) ..	Villieria	Pretoria ..	African Coast Fever.
,,	Cow	,, Sturge ..	Mr. Tacker ..	—	Rustenburg	,, ,, ,,
,,	,, ...	,, ,, ..	Mr. Taillard ..	—	,, ..	,, ,, ,,
13-4-04	Calf	,, Webb ..	A. Erasmus ..	Krugerspost ..	Lydenburg ..	Nothing seen.
,,	,, Dead ..	,, Garraway ..	Mr. T. Lee ..	Baviaanspoort ..	Pretoria ..	Decomposed.
14-4-04	Horse	,, Dunphy ..	Robson & Holtimes	Krugersdorp ..	Krugersdorp.	Micrococcus.
15-4-04	Ox	Inspector O'Connor	Agric. Dept. ..	Groenkloof ..	Pretoria ..	Nothing seen.
,,	Cow	,, ,,	G. W. Malherbe..	,, ..	,, ..	Ordinary Redwater.
16-4-04	Ox	D.V.S. Lee	—	Volksrust ..	Wakkerstroom	Nothing seen.
,,	Indian Ox ..	—	Pretoria Zoo ..	—	Pretoria ..	,, ,,
18-4-04	Heifer	V.S. Dunning ..	Repatriation ..	Vaalbank Stock..	Wakkerstroom	,, ,,
,,	,, ...	Manager	Government ..	Stud Farm ..	Standerton ..	Decomposed.
19-4-04	Ox	D.V.S. Garraway ..	G. Diani	Daspoort.. ..	Pretoria ..	Ordinary Redwater.
,,	,, ...	,, ,, ..	Petrus (Native Chief)	Kroondal, 117 ..	Rustenburg	Decomposed.
,,	Cow, Dead ..	,, Lindsay ..	Mr. Labuschagne	—	Middelburg..	African Coast Fever.
,,	,, ,, ..	,, ,, ..	Mr. Barrett ..	—	,, ..	,, ,, ,,
20-4-04	Heifer, ,, ..	,, Garraway ..	Mr. Malherbe ..	Groenkloof, 419..	Pretoria ..	Nothing seen.
,,	Ox, ,, ..	,, ,, ..	Mr. Turner ..	Riveria	,, ..	African Coast Fever.
21-4-04	Heifer	,, Walker ..	C. Stafelberg ..	Dundonald, 219..	Ermelo ..	Nothing seen.

,,	Ox, Sick, and 2 Animals, Contacts	,, Garraway ..	Mrs. Schweizer ..	Riveria	Pretoria ..	All preparations contain piroplasma of African Coast Fever.
22-4-04	Cows, 5	,, ,, ..	Mr. Struben ..	V. d. Walt Street	,, ..	All slight infection of African Coast Fever.
23-4-04	Ox, Sick.. ..	,, ,, ..	Mr. Jamesom ..	Onderstepoort ..	,, ..	African Coast Fever.
,,	Ox, Dead ..	E. Heron	—	Nelspruit ..	Barberton ..	Decomposed.
,,	Cows, Sick, 5 ..	D.V.S. Garraway ..	Mr. Struben ..	V. d. Walt Street	Pretoria ..	African Coast Fever.
25-4-04	Cow	Capt. Channy ..	—	—	Rustenburg	Decomposed.
26-4-04	Sheep	D.V.S. Chalmers ..	T. H. Delaube ..	Palmietfontein, 162	Heidelberg ..	Nothing seen.
27-4-04	Mule	,, ,, ..	R. Botha.. ..	Blinkpoort, 208..	,, ..	Micrococcus in pus.
29-4-04	Cow	,, Webb ..	Mr. Schoeman ..	—	Lydenburg ..	African Coast Fever.
,,	Oxen, 2 (1 dead, 1 sick)	,, Garraway ..	J. van Zyl ..	Rietfontein, 250..	Pretoria ..	,, ,, ,,
,,	Ox, Dead ..	,, Lindsay ..	Native	Botstabilo ..	Middelburg..	,, ,, ,,
,,	,, ,, ...	,, ,, ..	S.A.C.	—	,, ..	Nothing seen.
30-4-04	Ox	,, ,, ..	,, ...	—	,, ..	,, ,,

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MAY, 1904.

2-5-04	Mule	D.V.S. Chalmers ..	—	—	Heidelberg ..	Nothing seen.
,,	Cow	,, Garraway ..	Mr. v. Alphen ..	—	Pretoria ..	African Coast Fever.
,,	Ox	,, ,, ..	Mr. Stander ..	De Kroon, No. 5, Crocodile River	,, ..	Nothing seen.
3-5-04	Cows, 2, and Calf	,, ,, ..	Mr. Burger ..	Gezina	,, ..	All contained Ordinary Redwater.
4-5-04	Cow	Inspector O'Connor	Mr. Joyce ..	Vermeulen Street	,, ..	Ordinary Redwater and African Coast Fever.
,,	Cows, 2	D.V.S. Garraway ..	H. Steytler ..	Rietfontein, 15 ..	,, ..	Basic and nucl. cells.
,,	Cow	,, ,, ..	R. E. Erasmus ..	119, Kotze Street	,, ..	Combined African Coast Fever and Ordinary Redwater.
5-5-04	Ox	,, Sturge ..	E. Denny ..	Paardekraal, 388	Rustenburg	African Coast Fever.
6-5-04	Oxen, A, B, C ..	,, Bush ..	M'Slabong (Native)	Zandbank ..	Piet Retief ..	All contained African Coast Fever.
,,	Cow	,, Garraway ..	Marhwitshia (Native)	Wonderboom, 311	Pretoria ..	African Coast Fever.

MAY, 1904.—(*continued.*)

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
7–5–04	Oxen, 1, 2, 3, 4..	D.V.S. Lindsay ..	A. J. Viljoen ..	Erf 394	Middelburg..	1, 2, 3, African Coast Fever.
			G. Joubert ..	Klip River, 88 ..	,, ..	4, nothing seen.
,,	Bull and Ox ..	,, Garraway ..	Native Missionary	Koedoespoort, 229	Pretoria ..	Both African Coast Fever.
9–5–04	Oxen, 4	,, Bush ..	W. Prinsloo ..	Sulphur Springs..	Piet Retief ..	All African Coast Fever.
,,	Ox	Mr. Roux	—	—	—	Smears bad, nothing seen.
11–5–04	Oxen, 4	D.V.S. Edgar ..	Natives	—	Pietersburg..	2 African Coast Fever and 2 nothing seen.
,,	Cow	,, Garraway ..	Mr. Townsend ..	Villieria	Pretoria ..	African Coast Fever.
12–5–04	,, ...	,, ,, ..	Mr. Wolmarans..	Sunnyside ..	,, ..	,, ,, ,,
,,	Ox	,, Lindsay ..	—	Pokwani, Rietfontein	Middelburg..	Doubtful, probably African Coast Fever.
,,	Oxen, 2	Inspector Paton ..	Mr. Potgieter ..	—	Machadodorp	Nothing seen
,,	,, 2	D.V.S. Turnbull ..	—	Jinzeala Kraal ..	—	African Coast Fever.
13–5–04	Cow	,, Edgar ..	Mr. J. Duncan ..	Town Lands ..	Pietersburg..	Combined ordinary Redwater and African Coast Fever.
14–5–04	Ox	,, Turnbull ..	—	—	Barberton ..	African Coast Fever.
16–5–04	Cow, Bull, & Calf	,, Walker ..	J. v. Schalkwyk..	Kleinkloof, 259 ..	Ermelo ..	All African Coast Fever.
,,	Ox	Inspector Paton ..	Ballnie Bros. ..	—	Machadodorp	African Coast Fever.
,,	,, ...	D.V.S. Sturge ..	P. Potgieter ..	—	Rustenburg..	,, ,, ,,
,,	,, ...	,, Bush ..	J. Woodman ..	Steinkoppies, 7 ..	Piet Retief ..	,, ,, ,,
,,	Cow, Spleen ..	,, Edgar ..	Mrs. v, Smallen	Town Lands ..	Pietersburg..	,, ,, ,,
,,	Ox	Asst. Commandant	—	Rietfontein, 451..	Middelburg..	,, ,, ,,
17–5–04	Cow, Ox, Calf ..	D.V.S. Garraway ..	Mr. Baerveldt ..	Erf 174, Sunnyside	Pretoria ..	,, ,, ,,
			Mr. Keiser ..	Mission Station, Hebron	,, ..	,, ,, ,,
		,, ,, ..	J. C. Koch ..	Hartebeestepoort	,, ..	Decomposed.
,,	Cow	,, Dunphy ..	Mr. Fourie ..	—	Krugersdorp	Nothing seen.
,,	Ox	District Commandant S.A.C.	S. Schoeman ..	Rietfontein, 280..	Pretoria ..	Decomposed.
,,	Horse, Pus ..	—	Trevenna Brewery	—	,, ..	Lymphangitis.
19–5–04	Ox	D.V.S. Chalmers ..	Mr. Schuurman..	Klipportje, 228 ..	Heidelberg ..	Nothing seen.
,,	,, and Cow ..	,, Sturge ..	Johannes (Native)	—	Rustenburg	African Coast Fever.

,,	Pus	,, Chalmers ..	—	—	Heidelberg ..	Nothing seen.
,,	Oxen, 2	,, Bush ..	Mankon (Native)	Bakenkop ..	Piet Retief ..	African Coast Fever.
			S. de Jager ..	Welverdiend ..	,, ,, ..	,, ,, ,,
20–5–04	Heifer	,, Turnbull ..	Mr. Kincard ..	—	Barberton	Combined Ordinary Redwater and African Coast Fever.
,,	Ox	Inspector O'Connor	F. Secca	Daspoort	Pretoria ..	African Coast Fever.
,,	Cow	,, ,,	C. Stead	Melville Township	,, ..	Nothing seen.
21–5–04	,, ...	D.V.S. Edgar ..	Messrs. Ross & Co.	—	Pietersburg ..	African Coast Fever and Ordinary Redwater.
,,	Pus of Mule ..	,, Garraway ..	P. Berk	Allendale, 215 ..	Pretoria ..	Lymphangitis.
23–5–04	Cow	,, Edgar ..	Mrs. Peterinary ..	Domkraal ..	Pietersburg ..	African Coast Fever.
,,	,, ...	,, ,, ..	Mr. Freland ..	Town Lands ..	,, ..	Nothing seen.
,,	Ox	,, Lindsay ..	S.A.C.	Rietfontein, 451 ..	Middelburg ..	African Coast Fever.
,,	Cow, Dead ..	,, ,, ..	Mr. Wheeler ..	Erf 392	,, ..	,, ,, ,,
24–5–04	,, ...	,, Sturge ..	Rev. Postma ..	—	Rustenburg	,, ,, ,,
,,	Ox	,, ,, ..	P. Trichardt ..	Malut Kloof, 4 ..	,, ..	Nothing seen.
,,	Ox, Dead ..	,, ,, ..	F. Bezuidenhout	131	,, ..	,, ,,
,,	Oxen, 2	J. Beeton	—	—	Pretoria ..	Smears broken, nothing seen.
25–5–04	Ox	D.V.S. Sturge ..	Titus (Native) ..	Nooitgedacht ..	Rustenburg	African Coast Fever.
26–5–04	,, ...	,, Garraway ..	Rooikraal ,, ..	Rietfontein, 280 ..	Pretoria ..	,, ,, ,,
27–5–04	,, ...	,, Edgar ..	Mr. Ross	Dwaars River ..	Pietersburg ..	,, ,, ,,
,,	,, Dead ..	,, Walker ..	Mr. Meyers ..	Bonnie Braes ..	Ermleo ..	Nothing seen.
29–5–04	,, ...	,, Garraway ..	—	Rietfontein ..	Pretoria ..	Combined Ordinary Redwater and African Coast Fever.
30–5–04	,, Dead ..	,, Sturge ..	Kana (Native) ..	Rankanjan ..	Rustenburg	African Coast Fever.
,,	,, P.M. ..	,, ,, ..	Jonas (Native) ..	Burfontein, 923	,, ..	,, ,, ,,
,,	,, ...	P.V.S., per C.S. Cameron	—	Rietfontein, 280 ..	Pretoria ..	Spherical pyrs of African Coast Fever.
31–5–04	Pus, Mules, 2 ..	D.V.S. Chalmers ..	1, G. West ..	—	Heidelberg ..	Lymphangitis.
			2, J. Steyn ..	—	,, ..	,,
,,	Oxen, 2	,, Evans ..	Brett & Beaker ..	Waterkloof ..	Zeerust ..	Nothing seen, smears useless and stuck together.
			Coetze & Smit ..	Koedoesfontein ..	,,	,, ,, ,,

Received.	Animal.	Sent by	Owner.	Farm.	District.	Result.
1–6–04	Ox, Yearling ..	D.V.S. Walker ..	A. Kleynhans ..	Vlakfontein, 29 ..	Ermelo ..	Nothing seen.
,,	,, ...	,, Lee	P. H. Steyn ..	Paardefontein ..	Standerton ..	Basic cells numerous.
2–6–04	Donkeys, 2 ..	,, Garraway ..	Agric. Dept. ..	New Muckleneuk	Pretoria ..	Nothing seen.
,,	Cattle, 4	,, Lindsay ..	S. Grobler ..	Sterkloop, 154 ..	Middelburg..	,, ,,
3–6–04	Donkeys, 3 ..	,, Garraway ..	Agric. Dept. ..	New Muckleneuk	Pretoria ..	,, ,,
,,	Mules (supposed Anthrax), 3	,, Cannon ..	P.W.D.	—	Standerton ..	,, ,,
6–6–04	Ox	,, Elder ..	M'Hlava (Native)	—	Embabaan ..	Ordinary Redwater.
7–6–04	Stud Cattle, 14	Manager ..	Agric. Dept. ..	Experi. Farm.	Potchefstroom	Reported elsewhere.
,,	Oxen, 4	D.V.S. Evans ..	—	Waterkloof ..	Zeerust ..	1, sick, African Coast Fever; 2, sick 4 days, African Coast Fever; 3, sick, African Coast Fever; 4, sick 14 days, African Coast Fever.
8–6–04	Cow	D.V.S. Garraway ..	Mr. Steyn ..	New Muckleneuk	Pretoria ..	Nothing seen.
,,	Horse, Liver and Kidney	,, ,, ..	Agric. Dept. ..	—	,, ..	,, ,,
9–6–04	Horses, 2.. ..	Veterinary Hospital	Military	—	,, ..	,, ,,
,,	Cow, Dead ..	D.V.S. Garraway ..	Unknown ..	Boom Street ..	,, ..	,, ,,
10–6–04	Oxen, 2	,, Lindsay ..	S.A.C.	Mitkek, 52 ..	Middelburg..	African Coast Fever.
			Repatriation ..	Broodsnyersplaats	,, ..	,, ,, ,,
,,	,, 2	,, Walker ..	Mr. Stafelberg ..	Dundonald. 219..	Ermelo ..	Nothing seen.
,,	Heifer	,, Evans ..	Mr. Visser ..	Richert's Dam, 203	Zeerust ..	Ring forms seen.
,,	Stud Cattle, 6 ..	,, Cannon ..	Government ..	Stud Farm ..	Standerton ..	Smears bad, nothing seen.
11–6–04	Ox	,, Webb ..	Mr. S. Hoffman..	Frischgewagd, 82	Lydenburg ..	Nothing seen.
,,	Horse	Veterinary Hospital	Military	—	Pretoria ..	,, ,,
13–6–04	Oxen, 2	D.V.S. Evans ..	—	1, Rietvlei, 77 ..	Zeerust ..	,, ,,
				2, Koedoesfontein	,, ..	African Coast Fever.
13–6–04	Cows, 3	,, Edgar ..	Pabull (Native) ..	Doornboom ..	Pietersburg..	Nothing seen.
			Stephs ,, ..	Bosch Kloof ..	,, ..	,, ,,
			Steph ,, ..	,, ,, ..	,, ..	,, ,,
,,	Ox	Asst. Comdt., S.A.C.	Mr. A. H. Standen	Rietfontein, 280..	Pretoria ..	Smears decomposed.
,,	Cows, 2	D.V.S. Lindsay ..	Natives	Buffelsvlei ..	Middelburg..	Both African Coast Fever.

14-6-04	P.M. Smears, Ox	,, ,, ..	J. C. Scheepers..	Erf 715	,, ..	African Coast Fever.
,,	Ox	,, Sturge ..	S.A.C.	Brakfontein, 278	Rustenburg	,, ,, ,,
15-6-04	Cattle, 2	,, Lindsay ..	— .	Rietfontein, 38 ..	Middelburg..	Nothing seen.
				,, 451..	,, ..	African Coast Fever.
16-6-04	Oxen, 2	,, Evans ..	Mr. v. d. Heever	Schinn's Drift, 283	Zeerust ..	Both African Coast Fever.
,,	Ox	,, Webb ..	Mr. T. Dorey ..	Goetgedacht, 298	Lydenburg ..	African Coast Fever.
,,	,, ...	,, ,, ..	Repatriation ..	Kleinfontein, 1253	,, ..	,, ,, ,,
,,	Sheep and Goat Scrapings	,, Conacher ..	—	—	Machadodorp	Sheep, nothing seen; Goat, Acariosis.
,,	Ox, Dead ..	Asst. Comdt., S.A.C.	Mr. F. Engelbrecht	Rietfontein, 280..	Pretoria ..	Smears bad, unable to diagnose.
,,	P.M. Smears, Ox	D.V.S. Garraway ..	Agric. Dept. ..	New Muckleneuk	,, ..	Nothing seen.
20-6-04	Oxen, 8	Inspector Lester ..	1, Peterinary ..	Fairview ..	Pietersburg	African Coast Fever.
			2, Johannes (Nat.)	Gelelai Valley ..	,, ..	,, ,, ,,
			3, Pepete ,,	Mission Station..	,, ..	,, ,, ,,
			4, Dombrais ,,	Laastegfonden ..	,, ..	,, ,, ,, and Ordinary Redwater.
			5, Andries ,,	Mission Station..	,, ..	African Coast Fever.
			6, Klip ,,	Tcheoma District	,, ..	,, ,, ,,
			7, Ramanesisa ,,	Scheivars ,,	,, ..	,, ,, ,,
			8, S.A.C.	Fort Edward ..	,, ..	,, ,, ,,
25-6-04	Cow	D.V.S. Garraway ..	Mr. Steinberg ..	Lybrant's Kraal..	Pretoria ..	Nothing seen.
,,	Heifer, Sick ..	,, Lindsay ..	J. Sinclair ..	Erf 665	Middelburg..	African Coast Fever.
,,	Portion of Lung..	Inspector Nicholls..	—	—	Vereeniging..	Encapsulated pleuro-pneu-
29-6-04	Ox	D.V.S. Lindsay ..	Jaas (Native) ..	Kaffir Kraal, 62..	Middelburg..;	African Coast Fever. [monia.
,,	Oxen, 3	,, Dunphy ..	Messrs. v. Buuren and P. McQuirk	Geldenhuis Estate, Germiston	Krugersdorp.	Nothing seen.
,,	Ox	Agricultural Dept...	—	Onderstepoort ..	Pretoria ..	African Coast Fever.
,,	,, ...	Asst. Comdt., S.A.C.	Mr. D. van Zyl ..	Rietfontein, 280..	,, ..	,, ,, ,,
30-6-04	,, ...	D.V.S. Garraway ..	A. H. Stander ..	,, ,,..	,, ..	Too decomposed to diagnose.
,,	Bottle Ox Blood..	,, Dunphy ..	Curtis (Butcher)..	Vogelsfontein ..	Krugersdorp.	Nothing seen.

Number examined for June, 1904, 92.

MONTHLY SUMMARY.

July, 1903—	
African Coast Fever	21
African Coast Fever and Ordinary Redwater	4
Saccharomy, farcimi	22
Tuberculosis	1
Measles in pig	1
Piroplasma Equi	2
Nothing seen	48
Total	99
August, 1903—	
African Coast Fever	20
Saccharomy, farcimi	15
Basic cells	3
Piroplasma Equi	1
Anthrax	1
Nothing seen	70
Total	110
September, 1903—	
African Coast Fever	55
African Coast Fever and Ordinary Redwater	1
Ordinary Redwater	6
Saccharomy, farcimi	5
Basic cells	1
Acariosis	1
Piroplasma Equi	1
Nothing seen	72
Total	142
October, 1903—	
African Coast Fever	26
African Coast Fever and Ordinary Redwater	2
Saccharomy, farcimi	5
Basic cells	1
Acariosis	2
Lung putrifactive infection	1
Nothing seen	45
Total	82
November, 1903—	
African Coast Fever	21
Saccharomy, farcimi	4
Ordinary Redwater	1
Potchefstroom cattle (notes elsewhere)	28
Nothing seen	56
Total	110

MONTHLY SUMMARY—(*Continued*).

December, 1903—

African Coast Fever	7
African Coast Fever and Ordinary Redwater	4
Ordinary Redwater	1
Saccharomy, farcimi	2
Piroplasma Equi	1
Emphysema in lung	1
Trypanosoma	1
Potchefstroom cattle (notes elsewhere)	41
Nothing seen	33
Total	91

Jaunary, 1904—

African Coast Fever	19
African Coast Fever and Ordinary Redwater	1
Ordinary Redwater	4
Basic cells	5
Saccharomy, farcimi	3
Nothing seen	42
Total	72

February, 1904—

African Coast Fever	24
African Coast Fever and Ordinary Redwater	3
Ordinary Redwater	1
Saccharomy, farcimi	1
Basic cells	3
Inflammation	1
Nothing seen	49
Total	82

March, 1904—

African Coast Fever	16
African Coast Fever and Ordinary Redwater	1
Ordinary Redwater	3
Basic cells	2
Piroplasma Equi	2
Anthrax	1
Pleuro-pneumonia	1
Streptococcus	1
Ring forms	3
Human basic cells	1
Human Malaria	2
P.M. Organs showing Horse-sickness	1
Nothing seen	40
Total	74

MONTHLY SUMMARY—(*Continued*).

April, 1904—

African Coast Fever	29
Ordinary Redwater	2
Piroplasma Equi	2
Micrococcus	2
Nothing seen	18
Total	53

May, 1904—

African Coast Fever	46
African Coast Fever and Ordinary Redwater	6
Ordinary Redwater	3
Saccharomy, farcimi	4
Basic cells	2
Nothing seen	23
Total	84

June, 1904—

African Coast Fever	27
African Coast Fever and Ordinary Redwater	1
Ordinary Redwater	1
Basic cells	1
Ring forms	1
Acariosis	1
Encapsulated Pleuro-pneumonia	1
Potchefstroom cattle (notes elsewhere)	14
Nothing seen	45
Total	92

DISTRICT SUMMARY.

Barberton—

African Coast Fever	20
African Coast Fever and Ordinary Redwater	1
Saccharomy, farcimi	4
Basic cells	1
Piroplasma Equi	2
Lung putrifactive infection	1
Nothing seen	16
Total	45

Ermelo and Carolina—

African Coast Fever	10
Saccharomy, farcimi	6
Nothing seen	14
Total	30

DISTRICT SUMMARY—(*Continued*).

Heidelberg—

Saccharomy, farcimi	2
Acariosis	3
Ring forms	1
Micrococcus	1
Nothing seen	39
Total	46

Krugersdorp—

Saccharomy, farcimi	1
Micrococcus	1
Nothing seen	13
Total	15

Lydenburg and Machadodorp—

African Coast Fever	54
African Coast Fever and Ordinary Redwater	2
Ordinary Redwater	1
Saccharomy, farcimi	27
Basic cells	3
Acariosis	1
Piroplasma Equi	2
Inflammation	1
Human Smears—basic cells	1
Nothing seen	64
Total	156

Middelburg—

African Coast Fever	38
African Coast Fever and Ordinary Redwater	4
Saccharomy, farcimi	5
Basic cells	3
Nothing seen	63
Total	113

Nylstroom—

African Coast Fever	10
African Coast Fever and Ordinary Redwater	3
Trypanosoma	1
Nothing seen	34
Total	48

DISTRICT SUMMARY—(*Continued*).

Pietersburg—

African Coast Fever	38
African Coast Fever and Ordinary Redwater	7
Ordinary Redwater	1
Basic cells	1
Streptococcus Infection	1
Ring forms	1
Human Smears—Malaria	2
Nothing seen	35
Total	86

Piet Retief—

African Coast Fever	17
Nothing seen	5
Total	22

Potchefstroom—

Ordinary Redwater	4
Anthrax	1
Special report, noted elsewhere	83
Nothing seen	12
Total	100

Pretoria—

African Coast Fever	83
African Coast Fever and Ordinary Redwater	6
Ordinary Redwater	13
Saccharomy, farcimi	11
Basic cells	6
Piroplasma Equi	4
Anthrax	1
Post-mortem Organs showing Horse-sickness	1
Ring forms	1
Measles in pigs	1
Nothing seen	154
Total	281

Rustenburg—

African Coast Fever	26
Ordinary Redwater	1
Piroplasma Equi	1
Nothing seen	21
Total	49

DISTRICT SUMMARY—(*Continued*).

Standerton—		
Saccharomy, farcimi		3
Basic cells		1
Emphysema in lung		1
Nothing seen		24
Total		29
Swaziland—		
African Coast Fever		8
Ordinary Redwater		1
Nothing seen		29
Total		38
Wakkerstroom—		
Nothing seen		5
Zeerust—		
African Coast Fever		7
Ring forms		1
Nothing seen		4
Total		12
Johannesburg—		
Ordinary Redwater		1
Saccharomy, farcimi		2
Basic cells		1
Pleuro-pneumonia		1
Nothing seen		1
Total		6
Vereeniging—		
Basic cells		1
Encapsulated Pleuro-pneumonia		1
Total		2
Natal—		
Basic cells		1
Nothing seen		5
Total		6
Orange River Colony—		
Nothing seen		2
Cape Colony—		
Nothing seen		1
Lourenco Marques—		
Tuberculosis		1

SUMMARY OF DISEASES.

African Coast Fever	311
African Coast Fever and Ordinary Redwater	23
Ordinary Redwater	22
Saccharomy, farcimi	61
Basic cells	18
Piroplasma Equi	9
Ring forms	4
Acariosis	4
Inflammation	1
Emphysema in lung	1
Trypanosoma	1
Lung putrifactive infection	1
Micrococcus	2
Streptococcus infection	1
Anthrax	2
P.M. Organs showing Horse-sickness	1
Measles in Pig	1
Pleuro-pneumonia	1
Encapsulated Pleuro-pneumonia	1
Tuberculosis	1
Human Smears—Basic cells	1
Human Smears—Malaria	2
Nothing seen	541
Total	1,093

THE PIROPLASMA BIGEMINUM OF THE IMMUNE OX.

I.

The disease of cattle caused by piroplasma bigeminum was first described as Texas Fever. Since the discovery of Killborne and Smith, this piroplasma has been traced in several parts of the world, and the disease caused by it has been described in various countries under different names. In South Africa it is known as "redwater." At the suggestion of Lignieres, the term piroplasmosis was proposed as a suitable term for all diseases due to a piroplasma. Accordingly Texas Fever was called the bovine piroplasmosis. Up to that time only one disease of cattle, due to a piroplasma, was known, the one caused by piroplasma bigeminum. During the last few years, however, a new malady has been traced on the East Coast of Africa, and has recently been introduced to South Africa. It was then called "Rhodesian Redwater," later as "Rhodesian Tick Fever," and to-day is known as "East Coast Fever." The Russian investigators, Dschunkowsky and Luhs, also noticed it in Transcaucasia, and designated it as "tropical piroplasmosis." In 1897 Dr. Koch, who had remarked the disease in East Africa, believed it to be identical with Texas Fever, and he thought at that time that the small and characteristic piroplasma found in the blood were first stages of piroplasma bigeminum.

After the disease had been introduced into Rhodesia and the Transvaal, it was repeatedly observed that the blood of the sick animals contains the piroplasma bigeminum along with the piroplasma of tropical piroplasmosis. And the opinion of Dr. Koch, which it may be said was also shared by Laveran, of the identity of these two parasites, was for some time maintained. When however, the study of the disease was undertaken in a systematic manner, it was found that there were really two distinct diseases, the one due to the small piroplasma, the other to the piroplasma bigeminum. The examination

of many hundreds of smears taken from sick and dead cattle revealed the fact that in the great majority of cases the small piroplasma was exclusively present. It was then also observed that this disease had a somewhat different course, accompanied by peculiar morbid lesions, and that cattle which are immune against Texas Fever were not immune against the new malady. The main feature was, however, that the tropical piroplasmosis could not be inoculated into susceptible cattle, even with large quantities of blood containing the small piroplasma in great numbers. This fact clearly marks the two diseases, since Texas fever is easily inoculable into susceptible cattle.

Thus, as the new piroplasma must be considered to be a species of its own, I propose to call it by the name of "piroplasma parvum" (*n. spec.*). This piroplasma is smaller than any of the already known species. It appears either in the form of a small sphere or as a rod (bacillary form). When stained with any of Romanowsky's methods (as, for instance, Laveran, Azur II., or Makonkey's), the sphere takes the shape of a ring, the karyosoma being visible on one side of the sphere, the margin slightly blue, and the centre usually colourless or only slightly tinged. The rod is inflated at one end, and herein lies the karyosoma. The rod-shaped parasites may be straight or curved. The rings are round, oval-shaped, or oblong, and one can also find forms which indicate intermediate stages between these rings and rods. After the death of the animal the piroplasma parvum takes the ring form.

These parasites are found towards the end of the disease in enormous numbers, investing from 30 to 90 per cent. of all red corpuscles, and several may be counted in the same blood cell.

The presence of piroplasma bigeminum together with piroplasma parvum is probably due to the breaking down of immunity against redwater through the influence of the piroplasma parvum. It is a well known fact that any animal which has recovered from an attack of Texas Fever, must contain the piroplasma bigeminum in its blood. This fact can be easily demonstrated by injecting such blood into a susceptible animal. Cattle known to be immune against redwater are often observed to suffer a second time from this disease when weakened through adverse circumstances. This is often the case when the animal has contracted some febrile disease, as, for instance, rinderpest. I have purposely dwelt on the description of the piroplasma parvum of tropical piroplasmosis, since its size, form, and shape are so typical that their presence permits of the diagnosis of the disease.

II.

The piroplasma bigeminum is so well known that any description is merely superfluous. It is found in all cases of redwater during the fever stage, and disappears afterwards. It also occurs in the blood of a recovered animal, although its form and shape have not hitherto been described. The strongest proof that the piroplasma is present in the blood of immune oxen is to inject such blood into young calves, when the typical piroplasma reappears, causing a reaction. This experiment has been made use of as a method of preventive inoculation, since it has been noticed that the reaction does not, as a rule, cause death, but produces immunity. For some time it has been known that after the first reaction, in some animals at least, a second occurs which may even prove fatal, and that during this reaction the piroplasma bigeminum reappears. Afterwards immunity is established. In an immune ox the usual form of piroplasma bigeminum may occasionally be found. If, however, the typical piroplasma, in its usual pear shape, is thought to be present in every immune ox, then organism must be so rare as to escape microscopic examination. It is also believed that the parasite may change to some other state in which it remains dormant.

III.

When the examination of the blood of a calf which has been injected with blood from an ox immune against redwater is made and is continued after the second reaction has taken place, then a peculiar phenomenon is observed, namely, the occurrence of endoglobular parasites, which correspond to the description of piroplasma parvum. Sometimes they infest a considerable number of red corpuscles, but never as many as tropical piroplasmosis of some duration. Nevertheless, the microscope reveals a picture resembling that found in East Coast Fever at the early stage of the disease, so that an error, unfortunately, is more than likely. We have observed rings and rods (bacillary forms), as described above and similar in size and shape. In some instances these organisms are already present during the second reaction after the inoculation of redwater immune blood. Usually, however, they appear after the second reaction is over, and sometimes along with piroplasma bigeminum in its typical shape. Increasing for the first few days, they may in exceptional cases infest about 10 per cent. of red corpuscles, decreasing gradually. Some calves show them in varying numbers; for the first few months one or two in each microscopical field, but the decrease is maintained finally, so that after several months they have become so rare that it needs much patient searching and successful staining to find them. These parasites are still found in the blood of animals even after the lapse of a year.

In several cases the appearance of these organisms is preceded or accompanied by poikylocytosis of the red corpuscles by the formation of basophile granulations and nucleated cells. The basophile granula are either uniformly distributed in the blood cells, and may be of different size, or appear only as a single dot, nearly always on the margin of the blood corpuscle. These granula cannot be mistaken with the above-mentioned organisms, since they are of a uniform dark blue when stained with methyline blue, and contain neither a chromatic body nor an achromatic zone. What I have described as endoglobular organisms are undoubtedly of a protozoic nature, inasmuch as they take the distinctive stain of piroplasma.

IV.

I consider these organisms as the type of piroplasma bigeminum in the immune ox, which when injected into fresh susceptible animals reappear in the form of the pear-shaped parasites. This immune form has nothing to do with piroplasma parvum, with which it is often easily mistaken. The following experiments will support this view:—

Experiment 1.—Ox VI., about two years old, born in Aliwal North, Cape Colony, from where it was imported, a district where no redwater is known. It was tied up in our station and never left the premises. On July 30th, 1903, it was injected with 10 c.c. defibrinated blood of Ox No. 347, which had contracted ordinary redwater towards the beginning of November, 1902. Already on the fourth day after injection the temperature began to rise in Ox VI.; reached, on the fifth day, 106·2 F. On the next day the typical piroplasma bigeminum was present in rather large numbers. On the 9th August, 1903, red urine was voided for the first time; on August 13th, piroplasma bigeminum was still present, but the urine cleared up. On August 14th the first basophile granulated red corpuscles appeared; they increased during the next few days, and marked poikylocytosis was visible. On August 16th the piroplasma bigeminum was rare; the temperature was then 104·2 F. in the evening. A week later the basic granulations were still noticed. On September 9th, 1903, a second reaction set in, which lasted about eight days, and the evening temperature rose as high as 105·8 F. On September 12th, 1903, the blood was

examined, and small endoglobular organisms were found in the red corpuscles. The basic cells reappeared the following day. The parasites increased during the subsequent days; poikylocytosis and nucleated corpuscles also appeared. On September 15th the parasites were in such numbers that the presence of tropical piroplasmosis was suspected, and the count revealed an infection of 10 per cent. of all corpuscles. On the 17th the number began to decrease, the basic cells being still abundant. Finally they almost entirely vanished; but whenever, at any later date, the blood was examined, they were found. During the second reaction nothing appeared amiss with the ox. There was, nevertheless, some doubt in mind that the ox had contracted the tropical piroplasmosis, although I am unable to explain where the infection could have come from. It should be stated here that an animal which has recovered from East Coast Fever is immune. I have tested such animals on badly infected ground for over a year. This demonstrates their complete immunity. We may therefore conclude that when we are able to give Ox VI. the East Coast Fever in the natural way—that is, by tick infection—that the reaction in which the small piroplasma was found had nothing to do with East Coast Fever. On February 15th, 1904, the blood of this ox was examined, and the typical rings were still found present in small numbers. On this date the ox was infested with 20 males and females of the tick *Rhipicephalus appendiculatus* (Neumann). These ticks had been feeding on nymphæ on an ox suffering from East Coast Fever. The same brood of ticks had already produced the disease in three other oxen. On February 23rd, 1904, the temperature of Ox VI. began to rise, and kept up during the next thirteen days, and on March 7th the Ox VI. died of tropical piroplasmosis. The piroplasma parvum was present in large numbers.

Thus I conclude that the two reactions in which the small endoglobular parasites were seen had nothing to do with one another; and that piroplasma parvum, although resembling in form and size the endoglobular parasite found in the second reaction of Ox VI., is altogether a different species.

V.

If what I describe as the immune form of piroplasma bigeminum is really only found after redwater reaction, then it follows it will not be found in animals which are susceptible to this disease. This point can easily be settled by examining blood of freshly imported cattle from England and of calves which are born in stables in the Transvaal. Such cattle, when injected with blood from an immune ox, should naturally show the appearance of rings and bacillary piroplasma.

Experiment 2.—Calves 240 and 241 were both born on the premises of the Laboratory; they were at the time in question still sucking, and there was no chance that they had previously contracted tick infection. Their blood was repeatedly examined and found to be free of any endoglobular parasites. On the 13th February, 1904, the blood of the redwater immune Ox 347 was examined and the typical rings were found to be present in small numbers. The ox was bled, and the defibrinated blood was injected into the calves.

Calf 240 was injected intraperitoneally, and Calf 241 subcutaneously with 10 c.c. def. blood.

Calf 240.—The blood which, since the injection was daily examined, showed piroplasma bigeminum for the first time on the 24th February. There was no rise of temperature. The next day the parasites were still present; also on the 26th, the 27th, and 28th. On the last date a slight elevation of the morning temperature began, but there was never any distinct reaction. On the 1st

March the blood was found to be free of parasites. From the 11th March a reaction began, during which the temperature rose considerably, and reaching 106° F. on the 17th March. From the 13th March onwards the marginal basic points were now noticed in the red corpuscles; poikylocytosis and basophile red corpuscles appeared, and were present on the 23rd March, when piroplasma bigeminum was again visible. From that date (23rd March, 1904) onwards a few rings and bacillary forms were noticed; they were present the next day, and on the 28th had considerably increased. The piroplasma bigeminum was still found in preparations dated the 26th and 27th March. The following days the rings still increased, and on the 7th April they were so numerous that the microscopic field resembled an infection due to piroplasma parvum at the beginning of East Coast Fever. There were sometimes two and three rings and rods in one corpuscle. A decrease took place from the 13th April, 1904. A daily examination was continued up to 1st June, 1904, when rings and bacillary forms were constantly found.

Calf 241.—The temperature of this calf began to rise on the 7th March. On the 13th March there was a distinct poikylocytosis present, a few basophile red corpuscles, and also the basic marginal points; and these were daily noticed up to the 26th March. On the 21st March piroplasma bigeminum appeared, and the first bacillary forms and rings were noticed. Since the 26th the rings were somewhat frequent during the days following. On April 13th basophile cells were still present. The decrease of rings began about the 13th April. The blood of the animal was also daily examined during the next months, and up to the present time the rings are still present in small numbers.

VI.

When Calf 240 came into a second reaction I deemed it advisable to test whether the blood of this reaction would produce the same effect as the immune blood. Accordingly, two calves, Nos. 242 and 243, were injected. These calves were also born on the premises. They served as controls for the numbers 240 and 241, and during the time of examination never showed any endoglobular organisms. Both calves were injected with 10 c.c. defibrinated blood under the skin on March 17th, 1904.

Experiment 3.—Calf 242.—On March 29th, 1904, the first piroplasma bigeminum was noticed. There was no distinct temperature reaction. The parasites were seen again on April 3rd, from which date an irregular course of temperature began. On the 17th April the piroplasma bigeminum was again found. On the 18th a few basophile marginal points were noticed in red corpuscles, and on the 19th the ring forms were already present. On April 21st rings and bacillary forms were seen in fair numbers. There were basophile granulations in red corpuscles. On April 25th basophile cells were still present, so were also the piroplasma bigeminum and the rings and bacillary forms. The piroplasma bigeminum disappeared again, the number of red cells with basophile granulations became reduced, and the rings remained in moderate numbers on the date of writing (1st June, 1904).

Calf 243.—From the sixth day after the injection of the blood a distinct reaction set in, lasting about seven days. The piroplasma bigeminum was observed on March 26th, when the temperature reached 106·4° F. On March 25th, 1904, poikylocytosis made its appearance; the piroplasma bigeminum was present during the next five days, and then gradually disappeared. The poikylocytosis was noticeable during that time. On the 19th April the poikylocytosis was again noticed, as so were the marginal points and the basophile

granulations. The piroplasma bigeminum appeared on the 20th April for the second time. The rings and bacillary forms appeared on the 26th, on which date a few basic cells were still noticed. After that date the bacillary forms and rings were continually found in the blood, but in gradually decreasing numbers.

VII.

The above experiments were done for the purpose of ascertaining as to whether, after an inoculation of immune redwater blood, the appearance of rings and bacillary forms is a necessary result. From various casual experiments made at a previous date when the true nature of these rings was not yet apparent, I remarked endoglobular parasites identical with those just described, although in the latter case the examination of blood was not carried out from the very beginning. The oxen used in this experiment all came from Aliwal North, a country free of redwater. The idea of the experiment was to ascertain whether the blood of an immune redwater ox, mixed with serum of a hyperimmunised ox simultaneously injected, or serum injected before virus, or virus injected before serum, modifies in any way the reaction due to the immune redwater blood.

Experiments.

3/12/03.

Injection of Blood—	
Sub-cut., Ox 229 Intrajug., Ox 230	100 c.c. serum. 5 c.c. blood VI., simultaneously.
Blood—	
Sub-cut., Ox 233 Intrajug., Ox 228	5 c.c. blood VI. and 24 hours later 100 c.c. serum.
Blood—	
Sub-cut., Ox 227 Intrajug., Ox 226	100 c.c. serum and 24 hours later 5 c.c. blood.
Sub-cut., Ox 211 Intrajug., Ox 225	Mixture of 100 c.c. serum and 5 c.c. blood 24 hours old.
Blood—	
Sub-cut., Ox 232 Intrajug., Ox 231	Controls 5 c.c. blood.

The result was that the serum had no influence on piroplasma bigeminum, inasmuch as the controls behaved exactly as the serum treated animals. In some of the animals a disturbance of the temperature was noticed, similar to that in ordinary redwater, whilst others showed no reaction.

Result of Inoculation and Microscopical Examination.

No. 229.—A slight primary and distinct secondary reaction was present. The blood was examined on the 25th February, 1904, and small rings were present.

No. 230.—No distinct reaction took place. The animal was used on the 23rd January, 1904, for a heartwater experiment, and died on February 14th. Nothing could be found in the blood of the dead animal.

No. 233.—This ox had a primary reaction, with high evening temperature. A secondary reaction was not so distinct, the morning temperature being somewhat higher. The animal was also used for a heartwater experiment and inoculated on the 12th January, 1904. It died on January 20th from heartwater. An examination of the blood during the reaction revealed the presence of poikylocytosis, basophile granulations in the red corpuscles, marginal basic dots; bacillary and rings were seen in rather large numbers, sometimes two and three endoparasites in the same corpuscle.

No. 228.—No distinct primary reaction was noticeable, but a secondary reaction lasted for some time. On 23rd February, 1904, the blood was examined, when numerous small rings and also bacillary forms of endoglobular parasites were noticed.

No. 227.—A primary reaction was noticeable. There was no distinct secondary reaction. Small parasites were seen on 23rd February, 1904.

No. 226.—The inoculation of blood was followed by an irregular course of temperature. No distinct primary or secondary reaction was noticeable. The blood examined on the 25th February, 1904, showed a few rings present.

No. 211.—No reaction took place. There was no examination.

No. 225.—A very irregular reaction took place, which could be differentiated into a primary and secondary one. Rings were also found on the 25th February, 1904.

No. 232.—No disturbance of temperature took place. Rings were noticed on the 25th February, 1904.

No. 231.—A slightly primary reaction and a distinct secondary reaction. On 17th February, 1904, the examination of blood gave negative results.

The result of the inoculation was the appearance of rings and bacillary forms in the majority of the animals. We are justified in concluding from the above experiments that the appearance of rings was due to the injection of immune redwater blood, the serum had no effect whatever, the blood giving rise to reactions such as are usually observed in redwater inoculations.

The examination of the blood was in some animals negative, which is not to say that the parasites were entirely absent, but merely that they escaped observation.

VIII.

In addition to the evidence brought forward under the foregoing paragraphs, I am able to quote more additional and similar experiments. Eighteen imported English thoroughbred heifers, belonging to the Hereford, Shorthorn, Jersey, Lincoln, Polled Angus and Aberdeen Angus breeds, were inoculated against redwater; two on September 10th, four on October 24th, ten on November 14th. With the exception of two, all had reactions; most of them had primary reactions, during which time, the piroplasma bigeminum was noticed; while in the majority of the heifers, poikylocytosis and basophile granulations of the red corpuscles were observed. Four of the animals died, two from the primary reaction and two from the secondary reaction. Some weeks after the reaction, the blood of the animals was again examined and in all of them rings were found. At the same time, the blood of ten imported animals of the same breeds which were never injected, but had served as controls, was examined and gave negative results.

IX.

Finally I was able to trace the rings, and also bacilliary forms in cattle which, at one time or another; had been injected with blood from other animals.

Calf 239 was born on the place. It was injected on December 28th, 1903, with the blood of an animal (Ox 491), which some weeks previously had the Trypanosoma Theileri in its blood. The object was to ascertain whether the trypanosoma was still present in the ox.

The Ox 491 was immune against South African Redwater. The result was that the calf showed a primary reaction; the secondary reaction was indicated by an irregular course of temperature. Since February, 1904, the blood was daily examined and rings and bacilliary forms were found in every preparation up to the present date.

Calf 198 (Africander heifer).—Was injected on May 13th, 1903, with 50 c.c. blood taken from a Redwater immune Queensland heifer, suffering from some unknown disease. There was some disturbance of the temperature in the injected calf, but the diagnosis was not satisfactory. It was later immunised against rinderpest by the simultaneous method. On August 21st, 1903, it was again injected with blood from an animal which had Trypanosoma Theileri in its blood. Again a disturbance ensued, but no trypanosomes were seen. On the 25th February, 1904, a careful examination of the blood was made, when rings could be found.

Calf 212.—On the 5th October, 1903, injected with blood of Ox 490, which contained Trypanosoma Theileri. The calf proved to be immune against the trypanasoma, but nevertheless, a well marked primary and also a distinct secondary reaction were noticeable. On the 22nd October, 1903, a very marked poikylocytosis, basophile granulations and marginal basic points were registered. which were visible for some time. No further notice was taken of the animal until February 17th, 1904, when the ring forms of piroplasma bigeminum were found.

Since it was evident that the rings and bacillary forms were a necessary result from the previous inoculation with redwater immune blood, it was further concluded that these rings would be found in most oxen, which at one time or another had been injected with the blood of immune cattle. At the Rinderpest Serum Station, which was under my charge, there were about two hundred oxen, all of which were hyper-immunised against rinderpest. They all had received at least 4,000 c.c. rinderpest virulent blood. This blood was taken from sick cattle, and as most of the cattle had been born and bred in redwater areas, it naturally followed that through the injection of rinderpest blood, the immunised cattle also became highly immunised against redwater. Indeed, the piroplasma bigeminum was repeatedly noticed in inoculated cattle which went through a rinderpest reaction. A microscopical investigation of twenty head was made at repeated intervals, when, in almost every instance. rings and bacillary forms were met with, at one time or another.

X.

I have observed that the rings and bacillary forms multiply in an immune animal when it is placed under adverse circumstances, as, for instance, when it is attacked by some other disease.

Calf 200 (an Africander heifer).—This animal was immunised against rinderpest on June 26th, 1903. It went through the same usual rinderpest reaction. On the 24th July, it was used for a heartwater experiment and injected with 10 c.c. blood of Goat 62, suffering from heartwater. The heifer reacted on August 10th. On August 13th, the blood ws examined, when ring shaped and rod shaped endoglobular parasites were noticed, resembling in every respect the piroplasma parvum. They decreased within the next few days, and when the heartwater reaction was over, on the 16th August, these parasites had become very rare. That this reaction was nothing else than pure heartwater was proved, for the fact of the inoculation of blood into sheep and goats, which died from typical heartwater and, further, that the endoglobular parasites were missing in these animals. The micro-organism of heartwater is not known. When a search for ring forms in Calf 200 was made at a later date (December 1903), they were always found and in comparatively large numbers.

Calf 187 (Somali).—This animal was injected with serum on April 11th, 1903. There was no virus used, but a disturbance of temperature was noticed afterwards and, during the reaction, basophile red corpuscles were observed. In my opinion, the cause of this must have been due to a natural infection with redwater, from which the animal had suffered before it came under my care. Later, June 26th, 1903, it was immunised against rinderpest by the simultaneous method, when a rinderpest reaction was seen. On July 27th, an injection with heartwater blood of Goat 60 was made. A slight reaction followed, during which the ring and rod shaped endoglobular parasites were seen. On February 25th, 1904, the blood was examined and the rings were still found to be present in fair numbers.

Ox 484.—This was an ox hyper-immunised against rinderpest. It was running on the pasture behind Magaliesberg. On the 12th December, 1903, it was noticed to be ill, not feeding and losing condition. Its temperature was high. An examination of the blood was made and numerous small piroplasmata were present, so that the diagnosis was rather doubtful. An inoculation of blood was immediately made into other cattle and also into sheep, when it was found that the disease was heartwater. The small parasites soon disappeared and the ox recovered. A repeated examination was made of its blood, and up to the present time the rings are present in fair numbers. Also in this case, the sheep did not show any parasites.

XI.

The foregoing notes indicate that the presence of ring shaped and rod shaped endoglobular parasites is due to a previous infection of ordinary redwater. Seeing that cattle suffering from tropical piroplasmosis only recover to the extent of 5 per cent., the thought that East Coast Fever was complicating the foregoing experiments must be excluded. In addition to this, the case of the first mentioned Ox (No. VI.) has clearly demonstrated that when small rings and bacillary forms have once been present in rather large numbers (10 per cent.), such an animal, when exposed to tick infection, will succumb, since it has not yet acquired any immunity against tropical piroplasmosis; neither does the presence of rings in the blood of animals indicate that immunity must be expected against East Coast Fever.

The ring shaped and bacillary shaped piroplasmata have been overlooked hitherto, in connection with ordinary redwater. That may be attributed to the fact that the examination of blood of animals suffering from ordinary redwater has not been continued long enough, but also to the fact that the parasites stained with ordinary aniline dyes are not easily recognised as such, and, finally, because in the majority of cases, they are so rare that they are easily overlooked. As I have previously pointed out the typical stains for protozoa, such as Romanowsky's modification, are necessary, in order to recognise the chromatic nucleus, which removes all doubt as to the true nature of the endoglobular parasite.

XII.

The ring shaped and bacillary shaped piroplasmata in immune cattle are considered by Professor Koch to be identical with what I have termed piroplasama parvum of tropical piroplasmosis. He has also observed that the inoculation of blood taken from a sick animal suffering from tropical piroplasmosis, does not produce the disease in the new animal. When, however, the inoculation is repeated, the appearance of the usual small parasites in the blood is noticed in some of the animals. And he concludes that, under

certain conditions, the organisms of East Coast Fever may multiply and undergo reproduction in the blood of the injected animal, notwithstanding that there is no reaction. Furthermore, he states :—

> " Other experiments have also shown that inoculations with the blood of recovered animals, which only contains an inconsiderable number of single parasites, will induce similar modified attacks of African Coast Fever, and, while these experiments have not been numerous, they intend to indicate that recovered animals are even more suitable for inoculation purposes than those which are actually sick."

At different intervals I have injected animals immune against South African Redwater, with virulent blood of tropical piroplasmosis and also with blood of animals immune against this disease, in quantities up to 2,000 c.c. at a time. After intervals of varying duration, these animals were exposed to natural infection, when they all, with only one exception, contracted the disease and died. The injection of blood from an animal suffering from tropical piroplasmosis, into another one may, indeed, cause the rise of the temperature, and very often after the reaction, basic points and basophile red corpuscles are noticed, together with the appearance of rings and bacillary piroplasma. Of five Texas calves, which were injected on November 24th, 1903, with quantities of 10, 50, 100, 500, and 1,000 c.c. virulent blood (East Coast Fever blood), all showed reaction due to the injection ; the inoculated blood was derived from another Texas calf whose blood was swarming with the piroplasma parvum. The animals were later examined for rings, when it was found that the bacillary and ring shaped piroplasma were present, and in the animals alive to-day they are still found. The calf which was injected with 1,000 c.c. virulent blood, was infected on February 8th, 1904, with only two pathogenic ticks (one male ; and one female ; Rhipicephalus appendiculatus). It contracted the tropical piroplasmosis, and, after an illness of 13 days, died from the disease. In this case it is uncertain whether the rings were not already present before the injection ; no previous examination having been made of the blood. The appearance may be attributed to the injection of virulent blood. But whatever the cause of these rings, I am forced to the conclusion that their presence does not indicate the immunity of the animal against piroplasma parvum.

When we analyse the experiments made by injection with tropical piroplasmosis blood, or blood of any immune animal, it becomes evident that with the piroplasma parvum, we also inject the organisms of ordinary redwater in its immune form. All animals which have hitherto recovered from tropical piroplasmosis are immune against redwater ; East Coast Fever has only so far been observed in redwater countries, the increase of rings and bacillary forms of piroplasma bigeminum may be a result of such injections, even when an animal is already immune against redwater.

XIII.

Since 19th January, 1904, fourteen oxen are undergoing treatment according to Professor Koch's method of inoculation. They have already been injected thirteen times with blood of animals recovered from tropical piroplasmosis, two of the immune oxen showed the rings in their blood, one had none. The East Coast Fever immune animals were tested as to their immunity by exposing them in infected areas for a long period. The fourteen oxen were previously used for the reproduction of rinderpest serum, and had been injected to the extent of 4,000 c.c. virulent rinderpest blood. They were injected the first time on 19th January. The second injection was made a week later, and on

this occasion their blood was examined, and in all animals, except two, the rings were noticed. The inoculation of blood brought no reaction on; the animals through their repeated rinderpest hyper-immunisation, also hyper-immunised against redwater, and, accordingly, the subsequent inoculation of small quantities of blood (10 c.c.) had no effect. Their blood was examined weekly for ring shaped and bacillary piroplasmata, which were found, without exception, in all animals during the period of five months, but always in very small numbers, and at no time was a distinct increase observed. It must be concluded that in these animals, the ring shaped and bacillary piroplasmata were due to the rinderpest blood injections and not to the inoculation with recovered blood of tropical piroplasmosis.

It is intended to expose the cattle to natural infection, treated according to Koch's method, and which, after five months' treatment, are expected to prove immune against East Coast Fever. The question of immunity due to inoculation, has, however, since been settled in Rhodesia, where about 5,000 head of cattle have been inoculated. Mr. Gray, Principal Veterinary Surgeon, at the last Congress of Veterinary Surgeons held on the 25th May, 1904, in Capetown, made the statement that cattle inoculated, according to Koch, died of East Coast Fever, just as cattle did which were never inoculated. The Congress passed, therefore, the following resolution :—

> "That this Conference, after considering the reports of the experts who have had practical experience of the effects of inoculation, as proposed by Dr. Koch, is reluctantly compelled to the conclusion that it will be vain to trust to inoculation to arrest the spread of African Coast Fever."

Conclusions.

1. The injection of blood into a susceptible animal, taken from an ox immune against ordinary redwater or Texas Fever, usually gives rise to a primary and a secondary reaction, during which period the typical pear-shaped piroplasma bigeminum makes its appearance.

2. Either with the second reaction or shortly afterwards, endoglobular parasites appear in the shape of rings and rods, which resemble the piroplasma parvum of East Coast Fever.

3. These rings and rods are seen in the majority of all cattle living in a redwater infected area and which, at one time or another, have been injected with blood of cattle immune against redwater.

4. The presence of the rings and rods in the blood of an ox, proves immunity against redwater.

5. When rings are observed in cattle injected with blood of cattle suffering from or immune against East Coast Fever, then they are due to the simultaneous injection of immune blood of ordinary redwater. At the present time, East Coast Fever exists only in redwater infected areas, where the cattle are immune against redwater.

6. The presence of rings does not indicate that an animal is immune against East Coast Fever.

7. The rings and rods may, under certain conditions, multiply and increase in numbers.

8. The rings and rods represent a phase in the life history of piroplasma bigeminum in the immune ox.

TEMPERATURES OF CALF 243.

March 11, 1904.		12		13		14		15		16		17		18		19		20		21	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102^{6}	103^{6}	102	104^{8}	101^{2}	102^{4}	103^{2}	102^{8}	103^{6}	103^{8}	102^{4}	104	102^{8}	104^{6}	102^{2}	104^{6}	102^{6}	104^{2}	102^{8}	103^{4}	101^{4}	104^{4}
22		23		24		25		26		27		28		29		30		31			
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E		
101^{4}	104^{8}	101^{8}	104	102^{2}	105^{2}	102^{4}	105^{6}	102^{4}	106^{4}	104^{4}	105^{2}	102^{6}	104	102^{8}	105	102^{4}	104^{2}	102^{6}	104^{6}		
April 1. 1904.		2		3		4		5		6		7		8		9		10		11	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102^{2}	104^{8}	103	104	103	104^{4}	102^{4}	105^{4}	102	104^{2}	102^{6}	105^{2}	102^{6}	105	103^{4}	105^{6}	103^{6}	105^{2}	102^{8}	104^{4}	103^{4}	104
12		13		14		15		16		17		18		19		20		21			
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E		
102	103^{8}	101^{4}	104^{4}	102^{4}	104^{2}	102	103^{6}	102^{2}	103^{6}	101^{4}	104^{2}	102	103^{4}	101^{2}	104^{4}	101^{6}	103^{6}	101^{6}	103^{6}		

TEMPERATURES OF OX VI.

July 30, 1903.		31		August 1, 1903.		2		3		4		5		6		7		8		9	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100²	103	99²	103⁴	99⁶	102⁸	98	103²	101	101⁸	100⁸	106²	100⁶	105⁶	101	103	99⁴	102⁸	99⁴	103⁶	100⁸	104⁶

10		11		12		13		14		15		16		17		18		19		20	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99⁶	103⁴	101⁶	105	101⁸	105²	102⁴	104²	103²	106	103	104⁶	102⁶	104²	100⁶	103⁶	98⁶	104	98⁴	102	100⁴	101⁸

21		22		23		24		25		26		27		28	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102⁶	103²	101	105⁶	101	104²	101²	104²	100⁸	103⁸	101	104⁸	99²	103	100⁶	104²

SECOND RE-ACTION TEMPERATURES OF OX VI.

Sept. 7, 1903.		8		9		10		11		12		13		14		15		16		17	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99^{2}	102	99^{6}	104	100^{4}	133^{4}	99^{8}	$\bullet 104^{6}$	100^{8}	105^{6}	102^{4}	105^{4}	102	104	102	105^{6}	103	105^{6}	102	104^{2}	101	105

18		19		20		21		22		23		24		25		26		27			
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E		
100	103^{2}	99^{8}	103^{2}	100^{8}	103^{4}	99^{8}	103^{4}	100	103^{2}	99^{8}	104^{6}	100^{6}	103^{6}	100	104	100^{6}	103	—	—		

TEMPERATURES OF CALF 240.

February 16, 1904.		17		18		19		20		21		22		23		24		25		26	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
	102.8	102	102.8	101.4	103	103.4	103	101.4	104	102	102.8	101.8	103	99.8	102.6	102	103.8	104	103.4	101.2	102.4

27		28		29		March 1, 1904.		2		3		4		5		6		7		8	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102	104.6	103	103.6	102	103.6	102.6	103.6	101.6	103.4	102.2	102.6	101.4	103.4	102	103.2	102.4	102.8	102.8	104.8	102.4	104.4

9		10		11		12		13		14		15		16		17		18		19	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101.8	104.8	102	105.2	103.2	104	103.8	104.4	103.8	105.2	104.6	104.8	105.4	104.8	105.2	105.4	104.8	106	104.6	105.4	104.2	104.8

20		21		22		23		24		25		26		27	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102.8	104.2	103.6	105.2	103.4	105.2	104	104.6	102.8	104.6	102.4	104.2	103.2	104.6	103.4	104

TEMPERATURES OF CALF 241.

February 16, 1904.		17		18		19		20		21		22		23		24		25		26	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	104	103	103⅝	103	104	—	103⅝	101½	104½	102	103½	101⅝	102¼	101¼	103¼	101⅛	103½	102½	102⅝	100¼	101⅝

27		28		29		March 1, 1904.		2		3		4		5		6		7		8	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101	103	101⅛	103⅛	101⅝	103	102	103¼	100¼	103	101¼	102½	101¼	103	102	103¼	101¼	104	103	105⅝	102⅝	105½

9		10		11		12		13		14		15		16		17		18		19	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102¼	104⅛	102⅝	105⅝	103⅛	105½	104	105⅝	104¼	105½	102⅛	104⅝	104⅝	104	106	105	104½	105	103½	104⅝	103¼	104½

20		21		22		23		24		25		26		27		28		29		30	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
103½	104⅛	102⅛	104	102⅝	104	102¼	104¼	102½	103⅛	102⅝	104½	103	104⅛	102⅝	104⅝	102½	104⅝	103⅛	104⅛	102½	105⅝

31		April 1, 1904.		2		3		4		5		6	
M	E	M	E	M	E	M	E	M	E	M	E	M	E
102½	104	102½	104	103	103⅛	103⅝	103½	101⅝	104½	102¼	105½	102⅝	104¼

TEMPERATURES OF CALF 242.

March 11, 1904		12		13		14		15		16		17		18		19		20		21	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102¼	103⅔	102⅔	103⅔	101½	102⅔	101⅜	103¼	102¼	102⅝	101⅜	104	102⅝	104¼	102	104¼	101⅔	104	102¼	104	102¼	104½

22		23		24		25		26		27		28		29		30		31		April 1, 1904.	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101¼	103⅔	101⅔	103⅔	101¼	104⅔	100⅔	103¼	101¼	104½	101⅜	103⅔	101½	103⅜	103	104	102½	103⅔	101¼	104¼	101⅔	103⅔

2		3		4		5		6		7		8		9		10		11		12	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101¼	102¼	102⅔	103¼	101¼	104⅜	100⅔	103	102¼	104⅜	102½	104	101⅜	104½	101⅜	103⅜	102⅜	104½	102¼	104½	102	103¼

13		14		15		16		17		18		19		20		21		22		23	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101¼	103¼	101⅔	103¼	101¼	103½	100⅜	102⅜	101½	103½	101½	102⅜	101	102⅔	101¼	102¼	101	102⅔	100¼	103½	100¼	103⅔

East Coast Fever.

(Results of Former Experiments.)

This disease was first heard of in the Transvaal in May, 1902, when a heavy mortality amongst cattle was observed in the Elands River Valley, especially in Nelspruit and Komati Poort. Its nature was, however, not recognised, and it was generally mistaken for rinderpest.

The spread of the disease and the heavy mortality caused a thorough investigation, the result of which was laid down by me in an article, "The Rhodesian Tick Fever," in No. 4 of the *Transvaal Agricultural Journal*, 1903. The malady was described under different names as Rhodesian Redwater, Rhodesian Tick Fever, and lastly Professor Koch proposed the name of East Coast Fever. It was generally accepted that the disease started from the East Coast of Africa—more accurately, from German East Africa—where it was first observed by Professor Koch in 1897, and who then described it under the name of Texas Fever, he being then under the impression that the peculiar small piroplasmata which are found in the blood corpuscles were young forms of the piroplasma bigeminum, the cause of Texas Fever or redwater. When the disease made its appearance in Rhodesia, it was still believed to be identical with redwater; further, it was thought that the ordinary redwater had acquired a very high virulency, and therefore altered in type. This theory was based on the fact that the disease first appeared in a herd of freshly imported Australian cattle (about a thousand), which were not immune against ordinary redwater or Texas fever, and which finally completely succumbed. At the same time it was noticed that a certain number of cattle showed, during life, the symptoms of haemoglobinuria, and, on post-mortem, lesions corresponding with those of Texas Fever, and the examination of the blood proved the presence of the typical piroplasma bigeminum in addition to the bacillary piroplasma as already described. The same observations were also made in the Transvaal. The fact, however, that the disease carried off the immune low veldt cattle in the same way as the high veldt cattle, suggested the supposition that although the disease resembled Texas Fever very much, yet it might be another and a new disease. The experiments undertaken for this purpose gave the following results:—

1. Cattle which are immune against redwater or Texas Fever, such as low veldt cattle, Madagascar cattle, Texas cattle, and Queensland cattle, contracted the disease and died.

2. Transvaal cattle, which had been hyper-immunised against rinderpest in the first instance, and then against ordinary redwater to the extent of 8,000 c.c. virulent redwater blood, contracted the disease and died.

3. Cattle which were known to be immune against redwater showed, on post-mortem, lesions of redwater and, in their blood, the piroplasma bigeminum, together with the bacillary piroplasma.

4. A certain amount of cattle did not show piroplasma bigeminum in their blood.

5. The average incubation period after exposure to natural infection was about 12 days; the average course of the temperature reaction was 13 days. The shortest period of incubation was 10 days; the longest was 20 days. The shortest course of the disease was five days; the longest period was 20 days.

6. The majority of the animals which had piroplasma bigeminum in their blood showed the symptoms of haemoglobinuria, and the majority of animals which only had the bacillary form of piroplasma present did not show this symptom.

7. The count of red corpuscles during the fever reaction showed the remarkable fact that, in the majority of cases, there was no decrease of red corpuscles, or only a very slight one.

8. The susceptibility of cattle to the disease was 100 per cent., and the mortality averaged 95 per cent.

9. The disease proved not to be contagious; healthy cattle placed in stables alongside sick cattle did not contract the disease.

10. The *post-mortem* lesions in cases of infection with bacillary piroplasma differed from the ordinary redwater in the majority of cases.

11. The inoculation of blood, into susceptible cattle, taken from sick cattle, which contained the bacillary piroplasma in great numbers, did, in no instance in a series of thirty experiments, produce the disease. The sick blood was injected in quantities of 5 c.c. to 2,000 c.c.

12. Cattle which had thus been treated acquired no immunity; they contracted the disease after they were exposed to natural infection.

13. Cattle which were injected with blood of immune oxen did not show any reaction due to the injection of such blood, neither did they acquire any immunity therefrom.

14. Cattle which were injected with serum of immune oxen did not acquire any passive immunity when exposed to natural infection. They died as rapidly as did the controls.

15. Animals which have recovered from East Coast Fever have acquired a permanent immunity.

Conclusions.

The disease has nothing to do with Texas Fever or redwater; it is a new disease, due to a piroplasma, different to the one found in Texas Fever. The observation that both piroplasmata are found together in some animals is explained by the fact that almost all animals which hitherto contracted the new disease were born in a country where redwater also existed, and they naturally were immune against redwater. Immunity in redwater differs from immunity acquired by recovering from a disease caused by bacteria, inasmuch as in the former case the blood remains infective to susceptible animals. The piroplasma must, therefore, still be present in the blood (*vide* article, "The Piroplasma of the Immune Ox"). Now, when such an animal comes under adverse conditions, as, for instance, in contracting a virulent fever, its immunity breaks down, and the piroplasma reappears under its normal shape and complicates the disease. Such observations were made in connection with rinderpest especially. Even exposure to fatigue may cause the reappearance of piroplasma bigeminum in the immune animal. The complication of East Coast Fever with Texas Fever can only be explained in this way. The observation that piroplasma bigeminum generally appears only towards the end of the fever reaction in East Coast Fever supports this view. The main difference between Texas Fever and East Coast Fever is the fact that the former one is inoculable into susceptible cattle, and the latter one is not; and again, that the blood of redwater immune cattle is still infective, whilst immune East Coast Fever blood is not infective.

Other diseases due to piroplasma, such as piroplasmosis of the dog (malignant jaundice), piroplasmosis of the horse (biliary fever), behave similarly to Texas Fever. Both the sick and recovered blood produce the disease when injected into susceptible animals. We can, therefore, distinguish two groups of piroplasmoses—one which is inoculable and to which belongs the Texas Fever of cattle, the biliary fever of the equine species, and of the dog. All these diseases are caused by a piroplasma, which is much larger than that found in the uninoculable Coast Fever. The Coast Fever piroplasma, commonly called the bacillary-shaped one, represents, therefore, quite a different species, and I propose to call it by the name of *Piroplasma Parvum* (n. spec.).

The Transmission of East Coast Fever by Ticks.

I.

From analogy to other diseases of stock, due to the presence of piroplasmata in the blood, we concluded from the outset that East Coast Fever must be propagated by ticks. All evidence pointed to this fact. Thus it was demonstrated that the disease, which is not of a contagious character, since it attacks cattle only in certain localities where previously infected cattle have sickened or died of this disease. Consequently it was common knowledge that these regions became more or less infected. When apparently healthy cattle, which had only been a short period on infected ground, were removed out of such areas, then they begin, after a lapse of some time (usually about twenty days later), to sicken and die. Although these sick cattle, which were running on a clean farm, mingled with healthy cattle, no immediate outbreaks of disease occurred amongst the healthy stock. Some weeks later, however, the same healthy cattle, which had been grazing over the same farm, began to sicken and die of the same disease. A direct infection had accordingly to be excluded; the infection had to pass through the pasture. A similar observation was already known in connection with Texas Fever, where a certain species of tick (rhipicephalus annulatus [boophilus bovis]) was found to be the intermediate host of the infection. Accordingly our task was to find out the right species of ticks which would act in the same manner. It was, in the first instance, our object to ascertain what species of ticks infected such parts of South Africa where East Coast Fever was rampant. In the different samples of ticks sent by District Veterinary Surgeons, and in those collected by myself in various outbreaks, but more especially in those of the low veld, I found the following species in varying numbers :—

1. Rhipicephalus decoloratus (Koch), the common blue tick of South Africa.

2. Rhipicephalus appendiculatus (Neumann), now commonly known by the name of the brown tick.

3. Rhipicephalus simus (Koch), designated by Lounsbury as the black-pitted tick.

4. Rhipicephalus evertsi (Neumann), the red tick.

5. Amblyomma hebræum (Koch), the tortoiseshell or variegated tick, especially known as the bont tick.

6. Hyalomma ægyptium (L.), the striped-leg tick, known by the name of the bont-leg tick.

II.

The ticks enumerated above belong to three different genera of the family of ixodidæ, viz., rhipicephalus, amblyomma, and hyalomma. The life cycle of every species is now well known, thanks mainly to the investigations of Lounsbury. The life cycle of a tick embraces the following stages :—(1) Egg ; (2) larva ; (3) nympha ; (4) imago, adult or sexual tick.

It is advisable to begin with the description of the habits of the sexual forms. Males and females meet on a host. After feeding, they seek each other for copulation. In some species the males seek the females, which remain attached to the animal ; in others the reverse takes place. Males and females are usually the same size, but after fertilisation the latter begins to engorge enormously. This is the tick commonly spoken of by farmers. The male may be found alongside, and compared to the fully engorged female is very small. This led to the popular South African opinion that these tiny males were young ticks. The female repletes herself very quickly ; she detaches and drops to the ground. Usually she hides in grass or in soft ground. After a lapse of a certain period, which varies according to the different species, the female begins to lay eggs. This process also varies in length of time according to the species of the female. After the lapse of some weeks or months the eggs begin to hatch and young larvæ appear. These larvæ soon crawl to the next grasses and bushes, from where they attach themselves to the first suitable host which may pass.

So far all the above-mentioned species of ticks behave similarly. Some difference now enters into their habits, according to which we can divide the ticks into three biological groups.

1. *Ticks with one host.*—The tick which, in the form of a larva, attached itself to the host, undergoes on the same animal its complete life cycle, and accordingly leaves the host as an imago ; that is a sexual tick.

2. *Ticks with two hosts.*—In this case the tick remains on the same host during the first two stages in its life cycle. It comes on as a larva and leaves as a nympha. The nympha moults on the ground, and the resulting adult tick has to find a new host. (Rhipicephalus evertsi and Hyalomma ægyptium.)

3. *Ticks with three hosts.*—Here the tick leaves the host in each stage, and for the completion of the life cycle three hosts are required. (Rhipicephalus appendiculatus, Rhipicephalus simus, and Amblyomma hebræum.)

Regarding the details in the life cycle of the different ticks, the following notes may give some information. A zoological description of the several species mentioned does not enter into the spirit of this paper.

(1) *Rhipicephalus decoloratus.*—The blue tick is perhaps the most common South African tick, and is found everywhere. It is principally found on the larger domesticated animals ; more rarely on sheep and goats. It is met with under various climatical and tellurical conditions. Certainly it is less frequent on the high veld than in the middle veld and low veld. It is a very small tick ; nevertheless the female may reach a good size when engorged, and has then a rather bluish colour ; hence the name, blue tick. The female begins to lay eggs usually five days after she has left the host ; this applies only to the summer season. In the winter several weeks may elapse. The eggs hatch in the warmer season, usually after three to six weeks, and on an average after thirty-six days. In the winter time many weeks may pass. The young larvæ are endowed with longevity, which is a quality common to all species of ticks. They may live in the bottle and glass vessels for many months. They show much activity as soon as they are disturbed, probably because they hope to find a host. Once

on the host they begin to feed, and after about seven days the first moulting process takes place. Now appears the nympha, which settles close to the same place and begins to suck blood. After a lapse of time, which averages about seven days, the second moulting process takes place, and the sexual tick appears. A few days later copulation takes place, and the female begins to engorge rapidly. She soon drops. Thus the life cycle of the blue tick on a host is finished in about three weeks, and the complete life history requires, under the most favourable conditions, little more than two months

2. *Rhipicephalus evertsi.*—The replete female is bigger than that of the former species, and naturally lays a larger number of eggs, the reddish colour of which is peculiar to this tick. The hatching period of these eggs lasts, in summer, about thirty-one days. The young larvæ creep into the inner ear of their host, where they undergo their first moult. The nympha remains there until repletion, and drops sometimes as early as the tenth day, but usually, however, after sixteen days. The second moulting process takes place on the ground, generally after an average period of twenty-four days. The adult ticks select principally the hairless parts of the host, around the anus and the vulva. Thus the whole life cycle of this tick lasts, under the most favourable conditions, about ten weeks. The red tick is found on almost all domesticated animals, but rarely on the dog. It is distributed over all South Africa, and is also found in high altitude.

3. *Hyalomma ægyptium.*—This tick may be considered to be the largest of South Africa. The engorged female reaches an enormous size. The number of eggs she lays is calculated to amount to from 15,000 to 20,000. The hatching period averages thirty-two days. Until recently the complete life cycle of this tick was not known. The larvæ would not feed on any mammalia. Finally Mr. Lounsbury succeeded in feeding the larvæ on fowls, on which they also undergo their first moult, and drop as replete nymphæ after about ten days I have repeated the experiment with a similar result. The second moulting process takes place on the ground. The adult bont-leg ticks are, perhaps with the exception of the dog, found on almost all domesticated mammaliæ. The peculiarity of this tick lies in the changing of the species of the host during the intermediate stages ; that is from a mammal to a bird. This tick is also a very common South African parasite, and is found under the same conditions as the red tick, and often in company with it. It merits close attention, since it very often causes a festering wound on the spot of attachment, and even large pieces of skin may become necrotic and drop out.

4. *Rhipicephalus appendiculatus.*—The brown tick is about the same size as the red one. Sometimes brown males, which vary in size, are present on a host, and some have a caudal appendix, which is missing in others. Thus the idea arose that we had to do with more than one species. From observations on adult males which had moulted in the laboratory, it was seen that they shed their nymphal skin without a visible tail, which, however, develops a few days after the tick had a feed. The sexes mate usually from the third day after they are placed on a host, and the female repletes herself rapidly, and may drop engorged from the fourth day. Laying eggs usually begins after six days. The hatching period averages, in the warm season, twenty-eight days. The shortest period in my records is thirteen days. In the winter time the hatching takes several months. The eggs seem to suffer greatly from cold, and the rearing of young larvæ has often proved a failure during that time. The young larvæ readily attach themselves on cattle. They also engorge rapidly, and leave the host in as brief a time as three days. They have a rather bluish colour after feeding on blood, but are red and even white when they derive their

nourishment from an œdematous ear. In summer time the first mould takes place after an average period of twenty-one days. The shortest recorded period was sixteen days. The casting of the nymphal skin is preceded by a discolouration, which begins at the head and gradually reaches the opposite end. Finally the capsula becomes white, breaks on both lateral margins, and a colourless nympha creeps out. After a few days this nympha acquires its normal colour, becoming strong and active. These nymphæ may live in covered vessels over three months. Lounsbury records an instance where they lived up to seven months. Shortly after moulting these creatures do not seem to be very hungry, and do not bite readily. A few weeks later, however, they eagerly seek attachment when placed on cattle. Three days later some of them are already found replete and then drop. The outward changes of the moulting process of the nymphæ are the same as observed in the larvæ. The adult ticks appear in the summer time, after an average period of eighteen days. They are like the larvæ and nympha, almost colourless and very weak. A few days later, however, they have taken on their characteristic colour and are also much more vigorous. The adult brown ticks live in the bottle up to three months, and it is probable, under natural conditions, a much longer period. Once they are hungry they bite readily, and soon begin to mate, as already mentioned. The length of the whole life cycle of this tick lasts 73 days under the most favourable conditions. Under unfavourable conditions this time may last over six months. The Rhipicephalus appendiculatus is principally a summer tick, during which time it is found on various domesticated animals, chiefly in the ear or scattered all over the body. They usually prefer the hairy margin of the ear, where they are met with in scores. Larvæ and nymphæ are met with over the whole surface of the body, principally, however, on the legs. The brown tick prefers, as its habitation, warm stretches of the country. It is abundant in the low veld, less frequent in the middle veld, and is very rare, often entirely absent, on the high veld, where, on account of the cold winters, it cannot live.

5. *Rhipicephalus simus.*—This tick is similar in size to the former one. The life cycle is the same. The hatching period of the eggs averages 30 days. The first moult usually takes place after 20 days, and the second moult after 25 days. Adult black-pitted ticks are found on all domesticated animals. The dog seems to be a favourite host. I have observed that larvæ and nymphæ do not readily bite on oxen. It is quite probable that under natural conditions larvæ and nymphæ select other animals than cattle as their hosts. This tick is likewise principally found in the warmer parts of the country. It is not found on the high veldt.

6. *Amblyomma hebræum.*—The bont tick attains the same size as the bont-leg tick. The engorged female is sometimes even larger, and may weigh four grammes. This tick takes a long time for its complete evolution. The female begins the laying of eggs in summer time, about a fortnight after dropping, but three months may pass in winter time. The shortest hatching period lasts about 10 weeks, sometimes as many months, for it averages from four to six months. The young larvæ when hungry bite readily, and replete themselves in from four to five days. The majority drop between the fifth and seventh day. The first moulting takes place after one month, but sometimes three months may pass. The nympha repletes itself on a new host in from three to four days. The last moulting takes place in about three weeks. The adult ticks mate in about five days after they get on a host. The females drop about seven days after copulation, having engorged enormously. The males may remain many months on the same host. This is not a peculiarity of the bont

tick; the males of almost every species may do so. Once they are removed, however, from the host they soon die. This tick also produces festering sores, especially on the udders of heifers, and may even cause the constriction and sloughing off of teats. The whole life cycle of this tick is, under favourable conditions about nine months; under adverse conditions over a year is required, or perhaps even two. The bont tick is an inhabitant of the low bush veld; it does not appear in the middle veld. The principle hosts of this species are the ruminants, on which all life stages are met with. The adults attach themselves on the ventral parts of the body and on the legs. They select such places by preference where the skin is soft, as under the elbows and behind the hind legs, on the groins, and on the udder. Larvæ and nymphæ are frequently found on goats and sheep.

III.

The knowledge of the life cycle of a tick indicates the direction in which experiments must be pursued in order to ascertain whether such ticks act as transmitters of the disease.

1. The transmission is effected by means of young larvæ, whose mothers have been sucking on sick animals. This is probable for all species. The piroplasma bigeminum of Texas Fever, as Smith and Killbourne have first shown, is transmitted in this way by Boophilus bovis (Rhipicephalus annulatus), a tick closely allied to our South African blue tick. Koch has demonstrated the same mode of transmission in redwater in German East Africa. Lately Kossel, Schutz, Weber, and Miessner have produced the hæmoglobinuria of cattle, that is, redwater, in Germany by the progeny of Ixodes reduvius. In these instances the infection is transmitted through the egg.

2. The infection does not go through the egg. It is contracted either in the larval state and transmitted as nympha or adult, or it is acquired in nymphal stage and communicated in the adult state. The life cycle of the blue tick excludes such a possibility. The bont-leg tick has also to be excluded, since its intermediate stages live on the bird. The red tick might act as a transmitter since it passes the larval stage on an ox, and leaves the host as a nympha. It might communicate the disease as an adult. The other three species, the brown, the black-pitted, and the bont tick must be chiefly considered. Indeed, it is a well established fact that heartwater in sheep, goats, and cattle is transmitted by the nymphal and adult bont ticks, which have been feeding on sick animals as larvæ and nymphæ. Lounsbury, to whom the observation is due, has also shown that the adult tick occasionally transmits the disease when it has passed its second feeding stage on another non-susceptible host, after it has infected itself as larva on a sick animal.

3. The transmission is effected by the adult tick which acquired the infection in its previous sexual form, the infection having passed from the mother tick into the eggs, remaining latent in the larva, in the nympha, and appearing in the adult. Such, at least, takes place in the case of biliary fever of the dog, as Lounsbury has shown. Hæmophysalis leachi (Audouin), the South African dog tick, leaves the host as larva and nympha, and produces the fever only after it has obtained its sexual stage. None of the intermediate stages have a pathogenic effect.

4. *Direct Transmission.*—It is a popular opinion that the ticks in their adult stages crawl from one animal to the other. This is possible, but it is not general; and it needs a very intimate contact, as, for instance, when one animal

rubs the ticks against another one. It is also generally believed that adult ticks, after they drop off, seek another host, and especially those ticks which leave a dead animal. Brown ticks do indeed leave the dead host. When such ticks—more often males, but also females which have not started repleting themselves—are placed on fresh cattle, most of them will bite. The same ticks, when kept a few days without feeding, will soon die, whereas the brown adult tick which emerges from its nymphal skin, lives for many months. We may safely suppose that, under natural conditions, only a few of the adult ticks will reach a new host after they have left a dead animal.

A direct transmission of a disease has been observed to take place in the Brazilian fowl, spirillosis. Marchoux and Salimbeni have demonstrated that an Argas (fowl tick), feeding on sick birds, will communicate the disease to healthy birds. Fowl spirillosis is, however, an inoculable malady, and Argas is a tick which leaves the host after feeding, to return again. Thus a direct transmission is possible. Our ticks, however, leave their host solely to undergo further development before they seek a new host. In addition to this, East Coast Fever is not an inoculable disease, and a direct mechanical transmission, which would amount to nothing else but an ordinary inoculation, is impossible. Nevertheless, some development might be expected to take place in the body of the adult tick in like manner to the plasmodium of malarial fever in the body of a mosquito.

IV.

With the exception of Amblyomma hebræum, my own experiments were made with the above-mentioned ticks and in the different stages of their life cycle. For this purpose adult ticks, larvæ, and nymphæ were placed on the ears of experimental animals where, as a rule, they bite readily. A bag was then placed on the ears of the animals in order to collect the engorged ticks. The replete ticks were placed in Petri dishes, and were kept there to undergo their moulting process. The necessary moisture was applied by means of blotting paper placed under the cover of the Petri dish, which was wetted periodically. The ticks may be equally well fed on the leg of a quiet ox, and in order to collect them a kind of stocking was drawn over the leg. The blue tick larvæ were simply scattered over the skin. They attach themselves readily after having been confined for some time. As a rule it is advisable to starve the ticks for a short period before placing them on the animals, as they will bite the more readily when hungry. The engorged females were collected by simply picking them off the animals or by placing a cap over the ears, into which they dropped. This is decidedly the best way to collect brown ticks and red nymphæ. A number of animals were exposed to infection in Nelspruit, a country very badly infected with ticks and East Coast Fever. The temperature of the exposed animals was taken daily and as soon as a reaction set in, and the parasites were in sufficient quantities in the blood; the caps were put over the ears, and a daily collection of engorged ticks was made. In addition to this, all these ticks were bred in the Laboratory, and were fed on healthy and sick animals. It was in this way that we were able to study the life cycle of the different species. The skins of dead animals were carefully searched, and everything was collected. In this way we noted more especially the engorged intermediate stages of the brown tick. The details of the experiments will be given in the next paragraphs. It is impossible to give an enumeration of all the experiments in chronological order, inasmuch as the collection and feeding of the ticks depended very much upon what opportunity offered and the season which prevailed.

V.

Experiments with Larvæ, the Progeny of Ticks which were Collected from Sick Cattle.

(Rhipicephalus Decoloratus.)

1. Ox 212.—On the 13th January, 1904, this animal was infested with several thousand larvæ whose mothers were collected on the 5th November, 1903, from cattle exposed at Nelspruit. The larvæ had hatched after the 1st January, 1904. The cattle at Nelspruit were, at the time of the collection of the ticks, suffering from East Coast Fever. A second lot of the same brood of larvæ was placed on this ox on the 21st January, 1904. The engorged females dropped from Ox 212 after the 6th February, 1904, and were collected in large numbers. This animal remained healthy.

2. Ox 218.—On the 14th January, 1904, this animal was infested with numerous larvæ, the progeny of blue mother ticks collected on the 22nd December, 1903, from three Nelspruit Cattle, Nos. 5, 11, and 21, which were suffering on this date from East Coast Fever. The larvæ began to hatch on the 4th January, 1904. Engorged females dropped from Ox 218 on the 6th February, 1904, in large numbers. This animal remained healthy

3. Ox 221.—On the 15th January, 1904, this animal was infested with numerous larvæ, the progeny of blue ticks collected on the 16th November, 1903, from the Nelspruit Heifer, No. 16, which died on that date from East Coast Fever. A second lot of the same brood of larvæ was placed on Ox 221 on the 21st January, 1904. The engorged females dropped from the ox after the 7th February, 1904. This animal remained healthy.

4. Ox 222.—On the 16th January, 1904, this animal was infested with several thousand larvæ collected from the 15th November to the 17th November, 1903, from the Nelspruit Cattle, Nos. 13, 14, and 15, which were suffering on these dates from East Coast Fever. The engorged females dropped from Ox 222 after the 8th February, 1904. This animal remained healthy

5. Ox 168.—On the 21st January, 1904, this animal was infested with larvæ, the progeny of blue ticks collected from Nelspruit animal, No. 11, during the last four days of its illness, viz., from the 24th to the 28th November, 1903. These larvæ hatched from the 7th to the 11th January, 1904. The engorged females began to drop from Ox 168 after the 17th February, 1904. This animal remained healthy.

6. Ox 191.—On the 13th November, 1903, this animal was infested with larvæ, the progeny of ticks all collected from cattle at that time sick or dead at Nelspruit. This animal remained healthy.

Rhipicephalus Evertsi.

1. Ox 194.—On the 23rd November, 1903, this animal was infested with numerous larvæ of the red tick, whose mothers were collected from an Indian ox which died at Nelspruit from East Coast Fever (Ox 194 died on the 16th December, 1903, from pleuro-pneumonia).

2. Ox 232.—On the 8th February, 1904, this animal was infested with red larvæ, whose mothers were taken from the Nelspruit animal, No. 36, on the 15th December, 1903, which was at that date suffering from East Coast Fever and from Nelspruit animal, No. 8, which died on that date. The eggs began to hatch on the 5th February, 1904. This animal remained healthy.

Rhipicephalus Appendiculatus.

1. Ox 168.—On the 11th March, 1904, this animal was infested with brown larvæ, the progeny of females collected on the 18th January, 1904, from Nelspruit cattle, Nos. 29 and 31, being at that date at the end of their reaction. On the 14th March, 1904, Ox 168 was re-infested with the larvæ whose mothers were taken from Nelspruit animal, No. 39, which was suffering from East Coast Fever on the 20th January, 1904. These larvæ began to hatch on the 7th March, 1904. This animal remained healthy.

2. Ox 226.—On the 11th March, 1904, this animal was infested with the larval brood of some females collected on the 14th January, 1904, from the sick animals, Nos. 29 and 31, and from the dead animal, No. 32. The engorged nymphæ began to drop from Ox 226 on the 13th March, 1904. This animal was re-infested on the 11th March, 1904, with large numbers of larvæ whose mothers were collected on the 19th January, 1904, in Nelspruit, from animal, No. 37 the day after death; from animal, No. 39, then being in the middle of the disease, and from animal, No. 43, then at the beginning of the disease. The larvæ began to hatch on the 3rd March, 1904. The engorged larvæ began to drop from Ox 226 on the 16th March, 1904. This animal remained healthy.

3. Ox 709.—On the 14th March, 1904, this animal was infested with the larval progeny of the brown female ticks collected from the Nelspruit animal, No. 39, on the day of its death, and of all the other animals which were sick at that time. The engorged larvæ began to drop from Ox 709 on the 19th March, 1904. This animal remained healthy.

4. Ox 184.—On the 23rd November, 1903, this animal was infested with the larval brood of a collection of females which were taken from dead cattle of a fresh spontaneous outbreak. The microscopic diagnosis proved the presence of East Coast Fever. The larvæ which were placed on Ox 184 were ten days old. This animal remained healthy.

Rhipicephalus Simus.

1. Ox 226.—On the 8th February, 1904, this animal was infested with the progeny of black-pitted females, which were collected on the 21st November, 1903, in Nelspruit, from animal, No. 30, which died at that date. The larvæ began to hatch on the 27th January, 1904. The engorged larvæ began to drop from animal, No. 226, on the 15th February, 1904. On the 15th February, 1904, Ox, No. 226, was re-infested with the second lot of the above discribed larval brood. This animal remained healthy.

2. Ox 262.—On the 14th March, 1904, this animal was infested with the larval brood of black-pitted females collected on the 16th January, 1904, from the Nelspruit animals, Nos. 29 and 32, which were then about in the middle of the disease. On the 24th March, 1904, Ox 262 was reinfested with the larval progeny of females taken from the Nelspruit animal, No. 29, on the 22nd January, 1904, that is, two days before death; and from animal, No. 22, which was then at the beginning of the disease. The engorged larvæ began to drop from Ox 262 on the 30th March, 1904. This animal remained healthy.

Result.—All experiments had negative results. We are, therefore, entitled to conclude that tropical piroplasmosis is not transmitted by larvæ whose mothers have been sucking blood from sick cattle.

Experiments to see whether Nymphæ which have been feeding as Larvæ on Sick Cattle will transmit the Disease.

Rhipicephalus Appendiculatus.

1. Ox 203.—On the 3rd June, 1903, this animal was infested with a small number of brown nymphæ, which had been feeding as larvæ on the sick Nelspruit Calf, No. 164. The engorged nymphæ were collected on Ox 203 on the 11th June, 1903. The mothers of these nymphæ were taken from an immune ox, which in August, 1902, had recovered from East Coast Fever. The larvæ had been feeding on Calf 164 from the 29th April, 1903, and from the 1st May, 1903, up to the 5th May, 1903, on which date Calf 164 died from East Coast Fever. Ox 203 remained healthy.

2. Ox 184.—On the 8th June, 1903, this animal was infested with a second lot of the above nymphæ, which had been feeding on the sick Calf 164. The moulting had only taken place a few days previously. The engorged nymphæ dropped off Ox 184 from the 11th June, 1903. This animal remained healthy.

3. Ox 188.—On the 9th June, 1903, this animal was infested with about fifty nymphæ of the above brood, which, as larvæ, had been feeding on the sick Calf 164. These nymphæ moulted on the 7th June, 1903. The engorged nymphæ began to drop off Ox 188 on the 16th June, 1903. This animal came into reaction on the 22nd June, 1903. The typical piroplasma of East Coast Fever was present. The animal died on the 5th July, 1903. The incubation period lasted twelve days, the disease fourteen days.

Post-mortem.—Rigor mortis present; condition poor. The beef was pale; the blood was not completely coagulated. There was some liquid in the pericardial cavity. The liver was dotted with white and red infarcts. The gall-bladder was small and contained yellow bile. The spleen was normal. The kidneys were pale, dotted with numerous small, white infarcts and a few red ones. The urine was red. The mucous membrane of the fourth stomach was slightly reddened and very much swollen; and so also the mucosa of the small intestines, which was dotted with numerous hæmorrhages. In the colon were a few congested patches. The lungs were slightly œdematous. The endocard of the left ventricle was dotted with hæmorrhages.

4. Ox 207.—On the 30th June, 1903, this animal was infested with twenty-five nymphæ of the above brood, which had been feeding, as larvæ, on Calf 164. The engorged nymphæ dropped from Ox 207 from the 10th until the 24th July, 1903. This animal remained healthy.

5. Ox 184. On the 22nd December, 1903, this animal was infested with numerous nymphæ, which had yeen feeding, as larvæ, on the Nelspruit Calves 5 and 6. Calf 5 was sick from the 7th November, 1903, and died on the 29th November, 1903. Calf 6 was sick from the 15th November, 1903, and died on the 29th November, 1903. The larvæ were feeding on this calf from the 23rd November, 1903, until the 27th November, 1903. The mothers of these larvæ originated from healthy animals, and were bred in the laboratory through one generation. Calf 184 sickened on the 5th January, 1904, and died on the 18th January, 1904. The period of incubation lasted twelve days and the disease thirteen days. The piroplasma of the East Coast Fever appeared on the 8th January, 1904, and increased daily until death.

Post-mortem.—Made about half an hour after death. There was no rigor mortis. The animal was in good condition; the blood was not yet coagulated. The lungs were slightly œdematous. The mediastinum was infiltrated with yellow liquid; there was also liquid in the pericard. The heart was flabby;

ment, taking particular care to utilise only fresh imported stock preferably of young age. For this purpose, cattle and goats were obtained in the Cape Colony from regions where neither horse sickness or heartwater are known to exist. The sheep were all young animals born on the stud farm at Standerton, where both diseases are absent.

Experiment No. 1 with Angora Goats.

"A." The following goats were all injected on the 14th November, 1905, subcutaneously with 10 c.c. fresh virus horse 382.

Goat 371.—A slight reaction was noticeable, starting on the 5th day —19/11/05—and lasting about 15 days.
Bled on the 10th day after injection—November 24th—and on the 24th day after injection—December 8th.

Goat 372.—A slight temperature reaction indicated by a higher elevation of the morning temperature, the evening temperature remaining normal.
Bled on the 14th day after injection—November 28th—and on the 24th day after injection—December 8th.

Goat 373.—Temperature reaction began on the 9th day after injection. This reaction, however, was slight and principally indicated by a higher elevation of the morning temperature. There was a drop to normal about the 18th day.
Bled on the 14th day after injection—28th November—and on the 24th day after injection—8th December.

"B." The following goats were both injected on the 14th November, 1905, subcutaneously with 20 c.c. fresh virus of horse 382.

Goat 374.—A reaction began about the ninth day and lasted for nine days—December 2nd, 1905. The temperature during this reaction was somewhat irregular.
Bled on the 10th—14th day after injection—24th and 28th November.

Goat 375.—The temperature rose on the 5th day after injection. Reaction lasted until the 14th day and was well pronounced.
Bled on the 8th day after injection—November 22nd—the 10th day after injection—November 24th—and on the 24th day after injection—December 8th.

"C." The following goats were injected on the 14th November, 1905, intrajugularly with 10 c.c. fresh virus horse 382.

Goat 376.—A rise of temperature started on the 8th day—November 22nd—and returned to normal on the 17th day—December 1st. The reaction was somewhat irregular.
Bled on the 14th day after injection—November 28th—and on the 24th day after injection—December 8th.

Goat 378.—A distinct reaction started about the 7th day and lasted until the 17th day after injection—December 1st. Both morning and evening temperatures remained high on the 14th, 15th and 16th days between 105° and 106° F.

Bled on the 8th day after injection—November 22nd—and on the 24th day after injection—December 12th.

Goat 379.—No indication of a temperature reaction.

Bled on the 14th day after injection—November 28th—and on the 24th day after injection—December 8th.

"D." The following goats were injected on the 14th November, 1905, intrajugularly with 18 c.c. fresh virus horse 382.

Goat 380.—Irregular temperature observed.

Bled on the 10th day after injection—24th November— and on the 24th day after injection—December 8th.

Goat 381.—Irregular reaction; the temperature rose on the 7th day after injection—November 21st.

Bled on the 8th day after injection—November 22nd—and on the 24th day after injection—December 8th.

Remarks.—It has to be stated that none of the goats shewed symptoms of any disease. They were all feeding well during the observation time, and nothing was noticed amiss in any of them.

Conclusions to be Drawn from the Results of Experiment No. 1, "A," "B," "C," "D."

The foregoing experiments demonstrate that the blood of a horse suffering from horse sickness injected into young Angora goats bred in a country free from horse sickness produces a fever reaction in some goats. This reaction appears after an incubation time and lasts for some days.

It now remains to be seen whether the blood taken from these goats during the fever reaction is virulent for horses and mules.

Experiment No. 2.

To shew whether the blood of Angora goats injected with horse sickness virus and tapped during the reaction is virulent.

For the above purpose the blood of goats Nos. 375, 378 and 381, tapped on the 22/11/05—8 days after injection—was utilised. These three goats shewed the most distinct fever reaction.

Horse 495.—Injected intrajugularly on the 22nd November, 1905, with 5 c.c. of a mixture of defibrinated blood of the above goats.

The temperature rose on the second day after injection and reached 106.6° F. on the 4th day—November 26th.

3. Ox 207.—On the 23rd December, 1903, this animal was infested with six brown adult ticks, male and female, taken from an animal which had died from East Coast Fever in Nelspruit. The animal began to sicken on the 11th January, 1904, that is, after an incubation time of eight days, and it died on the 26th January, 1904, or after an illness of fifteen days. The piroplasmas were found the first time on the 13th January, 1904, and were present on the last day, infesting about 50 per cent. of corpuscles.

Post-mortem was made soon after death. The cadaver was in a fair condition; there was a discharge of mucous foam from the nose and mouth. The beef was somewhat pale and the fat was slightly yellow and of a gelatinous character. There was about 400 c.c. yellow liquid in the pleural cavity. The lungs were very œdematous, with numerous hæmorrhagic infarcts. In the heart bag was about 20 c.c. yellow liquid; the endocard of the left ventricle contained pronounced hæmorrhagic infiltrations; there was a gelatinous infiltration at the base of the heart and in the mediastinum. The liver had, on section, a glossy appearance; the bile was thick and of a brown colour. The spleen was normal. The kidneys were embedded in tissue, which was infiltrated with liquid; only one infarct was found in the kidneys. The urine was clear. The mucous membrane of the fourth stomach was slighlty reddened, and there were several hæmorrhagic ulcers, attaining the size of a threepenny piece. The mucous membrane of the duodenum showed red patches and was swollen. There were also a few hæmorrhagic ulcers in the small intestines about as big as a pea; the mucous membrane was swollen throughout the whole length of the intestines; and there were hæmorrhagic patches in the colon and the cœcum; some of the patches were of a slightly slate colour.

4. Ox 190.—On the 15th December, 1903, this animal was infested with five female and three male adult brown ticks, which were taken as nymphæ from cattle which had died at Nelspruit. The animal began to sicken on the 29th December, 1903, that is, after an incubation time of thirteen days, and it died on the 11th January, 1904, of East Coast Fever, after an illness of fourteen days.

Post-mortem was made directly after death. Rigor mortis was not yet present. The blood was not coagulated. Condition fair. The beef was pale. There was some liquid in the pleural cavity; there were some petechiae on the base of the heart-bag, which contained a little liquid. The epicard was spotted with hæmorrhages, which were also present in both ventricles. The lungs were œdematous. There were red infarcts under the pleura about the size of a pea. The liver was jaundiced. The gall-bladder was thickened and contained brown bile. The spleen was normal. There were a few white infarcts in the kidneys. The urine was normal. The fourth stomach was very much reddened, and there were numerous small hæmorrhages on the surface of the mucosa. There were red streaks and patches in the duodenum, the colon, and the cœcum. About 60 per cent. of all red corpuscles were infested with the piroplasmas of East Coast Fever.

5. Ox 224.—On the 8th February, 1904, this animal was infested with one male and one female brown tick which had been feeding, as nymphæ, on Calf 190, during the time it was suffering from East Coast Fever. These nymphæ moulted the 25th January, 1904. The larvæ came from mother ticks taken from a healthy ox. They had been feeding on a healthy ox. Ox 224 began to sicken on the 19th February, 1904, that is, after an incubation time of ten days, and it died on the 2nd March, 1904, the disease lasting thirteen days.

Post-mortem was made two hours after death. The condition of the cadaver was poor. The rigor mortis was not yet complete. The beef was rather pale. There were red infarcts in the lungs of various size. There were a few hæmorrhagic spots on the epicard and also a few in the left ventricle. The liver was hard and jaundiced. The gall-bladder was thickened and contained yellow liquid bile. The spleen was normal. There were a few infarcts in the kidneys. The urine was normal. The mucous membrane of the fourth stomach was very much swollen and uniformly reddened. The mucous membrane of the small intestines was also swollen. That of the colon was swollen and slate coloured, and a few ulcers were present, varying in size from a threepenny piece to a penny, and covered with necrotic matter. There were a few red patches in the rectum. The piroplasmas were pretty numerous on the date of its death.

6. On the 23rd February, 1904, this animal and the following one served as an ocular demonstration to convince some non-believers that East Coast Fever was carried by ticks. They were chosen by Mr. D. G. E. Erasmus, a well-known Transvaal cattle breeder, who carefully selected oxen which were immune to the common diseases of the country.

*E.*I was infested on the 23rd February, 1904, with eleven male and female brown ticks of the same brood, which had been feeding on Ox 190 during its illness. The animal began to sicken on the 5th March, 1904, that is, after an incubation time of ten days. It died on the 17th March, 1904, that is, after an illness of twelve days.

The *post-mortem* was made some hours after death. There were several large spots infiltrated with jelly-like matter in the region of the shoulder, the abdomen and the groin. There were hæmorrhagic infarcts in the lungs, which also showed a fibrous patch on the pleura. The liver was enlarged and dotted with numerous small white spots. The bile was thick and viscid. The kidneys were also spotted with numerous white infarcts. The fourth stomach and all the intestines were in a state of acute gastritis and enteritis. The urine was brown in colour.

7. *E.*II. was infested on the 23rd February, 1904, with nine females and males of the same brood of ticks, which had been feeding, as nymphæ, on Ox 190. This ox also began to react on the 5th March, 1904, after an incubation time of ten days, and it was killed on the 22nd March, 1904, being then in a state of collapse.

Post-mortem was not made by myself, but the examination of smears during the illness and taken the day previous to death, showed the characteristic piroplasmas of East Coast Fever.

8. Ox VI.—On the 15th February, 1904, this animal was infested with twenty male and female ticks of the brood feeding, as nymphæ, on Ox 190. The history of Ox VI. has already been related. This animal, after an injection of ordinary immune redwater blood, had a reaction, during which the small piroplasmas were seen in the blood. This experiment was made to test whether these small piroplasmas were those of East Coast Fever. Accordingly, if the ox now contracted East Coast Fever, it would prove that the first reaction had nothing to do with the disease. Ox VI. began to sicken after an incubation time of ten days, and it died on the 7th March, 1904, or after an illness of eleven days.

Post-mortem was made directly after death. The cadaver was in a poor condition. The lymphatic glands of the intermaxillary space were swollen. There were signs of a yellow diarrhœa. The blood was not properly coagulated. The beef had a peculiar brown colour. There was a yellow watery infiltration

of the different serous membranes. The lungs were pale, with a few red infarcts. There was much yellow liquid in the heart-bag, and there were a few petechiæ on the surface of the heart. The spleen was of normal size, its mesentery was infiltrated with copious hæmorrhages. The liver was enlarged, it was granular on section, and had a glossy appearance. The bile-bladder was distended with green thick bile. The kidneys were congested and contained numerous white infarcts. The urine was normal. The mucous membrane of the fourth stomach was strongly congested. There were numerous small hæmorrhages on the folds of the stomach. In the fundus of the stomach were two hæmorrhagic ulcers, reaching the size of a sixpence. The mucous membrane of the whole length of the entrails was swollen, reddened and contracted into folds. The outside aspect of all the intestines had a swollen appearance. The parasites of East Coast Fever were present in large numbers.

9. Ox 200.—On the 22nd February, 1904, this animal was infested with one male and one female adult brown tick of the brood which had been feeding, as nymphæ, on Ox 190. Both ticks bit readily. The animal remained healthy.

Result.—Out of nine oxen which were infected with pathogenic adult brown ticks, eight died from East Coast Fever. The experiments prove that two adults are able to produce the disease. But the experiment, Ox No. 200, proves that two ticks are not always able to produce the disease. The Experiments, No. 224 *E*I., *E*II., and No. VI., prove that the adult ticks have contracted infection as nymphæ. Their mothers were taken from healthy cattle, and the larvæ had also been feeding on healthy cattle.

Experiments with Adult Ticks, which as Nymphæ produced the Disease.

1. Ox 234.—On the 2nd February, 1904, this animal was infested with seventeen male and female brown adults, which, as nymphæ, were feeding on Ox 184, and which gave this animal the disease. The moulting into adults had taken place on the 17th January, 1904. The engorged females began to drop off from Ox 234 on the 10th February, 1904. This animal remained healthy.

2. Ox 205.—On the 30th July, 1903, this animal was infested with adult brown ticks which had been feeding, as nymphæ, on Ox 188, and so communicated East Coast Fever to this animal. The nymphæ began to moult in the laboratory on the 7th and 8th June, 1903. This animal remained healthy.

3. Ox 212.—On the 24th February, 1904, this animal was infested with ten adult brown ticks which had been feeding, as larvæ, on the sick Nelspruit animal, No. II. The nymphæ were feeding on Ox 184, during the incubation time. The last nymphæ dropped engorged on the same day, when the fever reaction started. The engorged females began to drop from Ox 212 on the 30th February, 1904. This animal remained healthy.

4. Ox 176.—On the 27th July, 1903, this animal was infested with seven adult brown ticks which had been feeding as nymphæ on Ox 188 and produced disease in this animal. The engorged females began to drop from Ox 176 on the 11th August, 1903. This animal remained healthy.

Result.—Brown ticks, which, as nymphæ, were pathogenic, did not produce the disease as adults. The case, No. 212, proves also that a nympha sucking blood during the incubation time does not take the infection, which *a priori*, had to be surmised.

Experiments with Adult Ticks which were taken from Sick and Dead Cattle and placed on Healthy Ones.

Rhipicephalus Evertsi.

Ox 216.—On the 19th November, 1903, this animal was infested with numerous adult red ticks removed on the 18th November, 1903, from the dead Nelspruit animal, No. 4. This ox remained healthy.

Hyalomma Ægyptium.

Ox 215.—On the 19th November, 1903, this animal was infested with numerous bont-leg ticks, which were removed on the 18th November, 1903, from the dead Nelspruit animal, No. 4. The male adults were still found on Ox 215 on the 19th December, 1903, and on the 6th January, 1904. This animal remained healthy.

Rhipicephalus Appendiculatus.

1. Ox 187.—On the 24th April, 1903, this animal was infested with numerous adult brown ticks, which had been taken from the dead Nelspruit animal, No. 167, on the 24th April, 1903. On the 7th May, 1903, seven adult males were still found fast in the ear of Ox 187. This animal remained healthy.

2. Ox 181.—On the 1st May, 1903, this animal was infested with twenty-two brown adult ticks, which were found in the ear cap of the Nelspruit animal, No. 168, after its death. On the 9th May, 1903, the replete females dropped from Ox 181. This animal remained healthy.

3. Ox 171.—On the 6th May, 1903, this animal was infested with adult brown male ticks removed from the Nelspruit animal, No. 164, shortly after death. On the 7th May, 1903, thirty-one adult male ticks were found attached to Ox 171. This animal remained healthy.

3. Ox 172.—On the 6th May, 1903, this animal was infested with the second lot of the adult male brown ticks removed from the Nelspruit animal, No. 164. On the 7th May, 1903, nineteen adults were found fast on Ox 172. This animal remained healthy.

5. Ox 238.—On the 12th December, 1903, this animal was infested with thirty-seven adult brown ticks, including two females. These ticks were removed from a dead cow in Machadodorp, which, on microscopical examination of the blood, proved to have been suffering from East Coast Fever. This animal remained healthy.

6. Ox 448.—On the 13th March, 1904, this animal was infested with male adult brown ticks which had produced the disease in the oxen *E.*I., and *E.*II., The ticks bit readily. The animal remained healthy.

7. Ox 223.—On the 8th March, 1904, this animal was infested with five male brown ticks which had produced the disease in Ox VI. The animal remained healthy.

Rhipicephalus Simus.

1. Ox 212.—On the 19th November, 1903, this animal was infested with adult black-pitted ticks, which were removed on the 18th November, 1903, from the dead Nelspruit animal, No. 4. On the 6th January, 1904, the male black-pitted ticks were still found on Ox 212. This animal remained healthy.

2. Ox 142.—On the 9th December, 1903, this animal was infested with nine black-pitted adults removed from the dead Nelspruit animal, No. 2, on the 29th November, 1903, These ticks were still found on the 19th December, 1903. This animal remained healthy.

Results.—Adult ticks which were taken from sick animals did, in no instance, produce the disease.

Brown ticks, which, as nymphæ, had produced the disease, did not produce the disease in their last feeding stage, that is, as adults. This experiment appears to prove that the brown tick is able to produce the disease only once. I have, however, as yet no experiment to show whether it is the male or the female tick that, after feeding, as nymphæ, on sick cattle, produces the disease.

Conclusions.—East Coast Fever is transmitted in South Africa by Rhipicephalus Appendiculatus and by Rhipicephalus Simus. The former tick must be considered as the principal carrier of infection, while the adult brown tick, which has been feeding on sick cattle as nymphæ, must be considered as the main intermediate host. All other ticks may be excluded. We made no experiments with Amblyomma hebræum, but since it is a tick with three hosts, there is a probability that it might also act as a transmitter of the disease. Again we may further conclude that the pathogenic tick produces the disease only once. The same tick which has once produced the disease does not remain long enough on the animal to reinfect itself again. We have to expect East Coast Fever in South Africa in such regions where the intermediate hosts of the piroplasma are found, or where such ticks are present which require three hosts for the development of their life cycle. These hosts live chiefly in the warmer parts of the Transvaal, and it is in such parts that East Coast Fever has become firmly established.

TEMPERATURES OF OX 207.

Dec. 23, 1903.		24		25		26		27		28		29		30		31		Jan. 1, 1904.		2	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
97²	103⁶	97	102	96⁴	103	99⁴	101	98⁴	102	97⁸	104	99⁶	103	98⁴	103⁴	99²	102⁴	100	99⁸	98⁴	102⁸

3		4		5		6		7		8		9		10		11		12		13	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99	102²	98	102²	98⁴	103	98⁴	102	96	101⁶	98⁸	102⁸	97⁸	101⁶	99	102⁸	100⁴	104	102⁸	104²	101⁸	105

14		15		16		17		18		19		20		21		22		23		24	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
103⁶	106	104⁶	106²	104⁶	106	102⁸	105⁸	103⁸	105	102	104⁸	104	105⁶	104⁴	106⁴	105	106⁴	104⁸	106⁶	105	106⁶

25		26	
M	E	M	E
106	106⁴	106	Died

TEMPERATURES OF OX 190.

Dec. 15, 1903.		16		17		18		19		20		21		22		23		24		25	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	—	—	—	—	103²	101	101	99	102	100⁴	101⁴	101	102²	99²	101⁶	99²	103	99	101⁸	96⁸	103

26		27		28		29		30		31		Jan. 1, 1904.		2		3		4		5	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
104	101	96	104	100	102⁸	102⁸	104⁶	103	106²	104⁶	106⁸	105⁸	103⁴	105	106⁶	104⁸	107	103²	105⁴	103	106²

6		7		8		9		10		11	
M	E	M	E	M	E	M	E	M	E	M	E
102	104	99⁶	105	105⁶	106⁴	106	107⁴	108	107⁸	103⁶	Died

TEMPERATURES OF OX 224.

Feb. 8, 1904.		9		10		11		12		13		14		15		16		17		18	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99²	102⁴	100⁴	102⁴	100⁶	102⁸	100⁸	102⁶	100	102⁶	100	103	99	102⁸	100⁸	104	100⁶	103	99⁸	102⁶	100²	102⁶

19		20		21		22		23		24		25		26		27		28		29	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102⁶	105²	104⁴	105⁶	104⁸	106²	105⁶	106⁸	106	106⁴	105⁴	105⁶	104²	105⁴	105	106	105²	106⁶	105⁶	107²	105⁸	107²

March 1, 1904.		2		3
M	E	M	E	
107²	107⁶	106⁸	108⁴	Died.

TEMPERATURES OF OX 188.

June 9, 1903.		10		11		12		13		14		15		16		17		18		19	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99⁸	102⁶	102	103	100⁴	102⁶	100⁸	103	100⁶	102	100²	102⁶	100⁸	103	101	103⁸	100	103⁸	99⁸	103	102	102⁴
20		21		22		23		24		25		26		27		28		29		30	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100⁶	103⁴	102	103⁴	103⁶	105⁶	105⁶	106⁴	103⁶	105⁶	101	105	104⁴	105⁸	105⁶	107	102	107	104²	106	103⁶	105⁴
July 1, 1903.		2		3		4		5		6											
M	E	M	E	M	E	M	E	M	E	M	E										
104⁸	106⁸	104⁶	106	105⁴	106⁴	105⁸	107⁴	104²	105⁸	Died.											

TEMPERATURES OF OX 184.

Dec. 23, 1903.		24		25		26		27		28		29		30		31		Jan. 1, 1904.		2	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99	103¼	99	102	99⅝	101⅜	100	101½	97	103	98¼	103	100	102¼	98	102¼	98	102	100	101¼	99⅝	102⅝

3		4		5		6		7		8		9		10		11		12		13	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99	102¼	97½	102⅜	102¼	107	104⅜	106	103½	105⅝	106⅝	108⅜	107	107½	105¼	105	105	105	104⅝	105¼	104⅜	105¼

14		15		16		17		18	
M	E	M	E	M	E	M	E	M	E
105	106¼	106¼	107½	105¼	107	106	107¼	106¼	Died.

TEMPERATURES OF OX 114.

Feb. 8, 1904.		9		10		11		12		13		14		15		16		17		18	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99²	102⁶	99	101²	99	102⁴	100	102⁶	99⁸	101⁶	99⁶	102	97⁶	102	97⁴	103⁶	100²	103²	99	101²	99⁶	102

19		20		21		22		23		24		25		26		27		28		29	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99⁴	102²	101	103⁶	99⁶	101⁴	98⁶	101⁶	98⁸	102⁶	98⁴	102⁸	100	—	—	105²	100	105⁴	103⁸	106⁴	105⁴	107²

March 1, 1904.		2		3		4		5		6		7		8	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
106⁴	107⁴	105⁸	107⁴	105⁶	106⁸	105⁶	106⁶	105⁴	106²	104⁶	104⁶	102⁴	103²	101⁴	Died.

EXPERIMENTS WITH SERUM AGAINST EAST COAST FEVER.

I.

In the second report by Professor Koch on the above subject, he mentioned that he was conducting experiments with a view of obtaining a curative serum. For the purpose of obtaining an anti-toxic serum which would tend to neutralise the toxic products of the organism, the animal whose serum it is proposed to use is gradually inoculated with increased doses of virulent blood.

Since it was discovered that healthy animals are also able to resist large doses of virulent blood, animals were prepared by inoculation with successive large doses of virulent blood for the purpose of obtaining a cytolytic serum. Such a serum possesses the property of directly attacking the specific parasite instead of neutralising its products as an anti-toxic serum would.

For the production of anti-toxic serum and cytolytic serum, only immune animals can be used, as susceptible animals tend to break down under repeated doses of virulent blood.

In this way immune animals were injected with increasing doses of blood taken from sick animals, starting with doses of 5 c.c., and concluding with a maximum of 2,000 c.c., while others received a succession of doses of 2,000 c.c. each of sick blood injected subcutaneously, or of 1,000 c.c. injected intravenously. Care was taken to utilise sick blood which contained a large number of organisms for fortifying purposes. After three or four large injections given at intervals of from two to three weeks, the serum of these animals was found to possess very remarkable properties, which may be summarised as follows:—

Injection of such serum into healthy animals in doses up to 150 c.c. resulted in no systematic disturbance. Injection of the same serum into sick animals caused a striking change in the African coast fever parasites circulating in their blood. The parasites became smaller, their outline was lost; sometimes they were scarcely visible, and, in the course of a few days they disappeared. Unfortunately, however, this specific serum also possessed an undesirable property in a very high degree which exerted a solvent action upon the blood cells of sick animals, whilst healthy animals remained unaffected.

Injection of 50 c.c. of hæmolytic serum into sick animals has nearly always been fatal, death being primarily due to its solvent action upon the blood cells. In sick animals treated in this way, there

of the dead animal did not show any trace of the parasites of East Coast Fever. The remaining nine animals were kept inside the fence during the months of December, 1903, January, February, March, April, and up to 30th May, 1904, when they were removed by a route, over which no cattle had been for more than eighteen months, to another fenced-in area. On the latter ground 28 head of cattle had died from Rhodesian Tick Fever in January and February, 1903.

During the same time, Texas cattle from the same shipment were exposed on another farm at Nelspruit, in the neighbourhood of the experimental ground previously referred to. Since May, 1903, this second farm had remained infected by constant re-introduction of fresh cattle. Here the cattle died as described in the inoculation experiment mentioned in former chapter. Some of them died during the months of December and January.

A final lot of ten Texas cattle were exposed on January 7th, 1904, on the same infected farm, and they died during the months following. The result of the latter experiment serves as a control, and proves that the infection did not die out of those places where, since the first outbreak (May, 1902), cattle had been repeatedly re-introduced.

From the foregoing observations we may conclude that an area, which at one time was badly infected with East Coast Fever, does purify itself again after a reasonably short period. This may be considered for the present to average fifteen months, but it is possible that our further experiments now in progress may show that a shorter period will suffice for purification. At the same time we must remark that our general observations on this disease have supplied us with a strong evidence that pathogenic ticks may remain active on an infected farm for six months after cattle have been removed thereof, and we have also a certain amount of evidence to show that infection may last for eight months. The importance of this knowledge is evident, and we are inclined to believe that the necessary period for purification will ultimately turn out to average between twelve and fifteen months.

Similar observations by Mr. Gray, regarding the time in which ground purifies itself, have since been made in Rhodesia, and they all formed the basis of our recommendations regarding the stamping out policy, which was endorsed by the South African Conference of Veterinary Surgeons, in Capetown, where the following resolution was unanimously carried :—

> "That this Conference is of opinion that the only effective method of eradicating African Coast Fever is to kill off all cattle in infected areas, and to leave such areas free of cattle for a period of not less than eighteen months."

II.

Inoculation Experiments According to the Methods of Professor Koch.

In a former article on East Coast Fever it was stated,* by one of us, that the injection into susceptible cattle of defibrinated blood of oxen, which have recovered from the disease, did not produce any reaction, and that the same cattle, when exposed to natural infection, contracted the disease and died. The oxen were, however, only subjected to one inoculation with immune blood. Since then Professor Koch has published some experiments in which he repeated the inoculation of immune blood every eighth day, and in this way produced what appeared to him to be modified attacks of African Coast Fever. He said, "while these experiments have not been numerous, they tend to indicate that

* Theiler, *Transvaal Agricultural Journal.*

recovered animals are even more suitable for inoculation purposes than those which are actually sick." After stating that single small or single large injections of blood from a sick, or recovered animal, did not afford any protection, he goes on to say that repeated injections were more satisfactory, and appeared to confer an undoubted immunity. He further stated his belief that the repeated injection of small doses of blood would confer an immunity which would be heightened in direct proportion to the number of injections to which the animal was subjected, and that his experiments showed that it was not necessary to employ the blood of sick animals for this purpose, as blood taken from recovered animals had a similarly satisfactory effect, and in some cases even a better one. He then recommended that defibrinated blood from recovered animals be infected subcutaneously four times, with an interval of seven days between each injection. After this 10 c.c. doses of defibrinated blood were to be given for some time every two weeks, after which one dose a month would suffice

We decided to follow the recommendations of Professor Koch and carry out the inoculation as an experiment under such conditions as would likely be met within an infected area when the disease has begun to make its appearance. We submitted ten animals to four inoculations with 10 c.c. of defibrinated blood at intervals of seven days between each injection. We then exposed them to natural infection on the veld, and continued the inoculations during the time of exposure.

The cattle employed in these experiments came from oversea to the laboratory, and had never been on Transvaal pastures.

Nine Texas calves, Nos. 21 to 25 and 27 to 30, were inoculated as follows:

1. Inoculation on October 28th, 1903.
2. Inoculation on November 4th, 1903.
3. Inoculation on November 11th, 1903.
4. Inoculation on November 17th, 1903.

On November 18th all nine animals were sent by rail to Nelspruit to be exposed on infected pastures. They arrived at Nelspruit on November 20th. The results were as follows :—

Calf, No. 21.—The fifth inoculation was made on December 1st. On December 2nd, or after 12 days' exposure, this animal's temperature began to rise. The fever continued for 12 days, and death ensued on December 13th. Blood smears showed a pure infection of East Coast Fever. Typical lesions of East Coast Fever were present in the organs.

Calf, No. 22.—The fifth inoculation was made on December 1st, and a sixth on December 12th. This animal started to sicken on January 19th, *i.e.*, after 60 days' exposure. Death took place on 29th January, after an illness of 10 days. The piroplasma bigeminum, ordinary redwater and that of East Coast Fever, were both found in the blood smears. The infection, then, was mixed in this case, but the chief point is that the animal was attacked with East Coast Fever and typical lesions of the disease were present after death.

Calf, No. 23.—The fifth injection was made on December 1st. Fever was noted on 7th December, *i.e.*, after an exposure of 17 days. Death resulted 16 days later on 17th December. Blood smears showed a strong double infection with the piroplasma of East Coast Fever and that of ordinary redwater. The lesions were typical of East Coast Fever.

Calf, No. 24.—The fifth and last inoculation was made on 12th December. The animal was still alive after six months' exposure. Several examinations of blood smears were made during febrile reactions, but the piroplasma could never be found.

Calf, No. 25.—The fifth inoculation was made on 1st December, and a sixth on the 12th December. Fever set in on 21st December, *i.e.*, after 31 days' exposure. The animal died after 12 days' illness; both piroplasmata were present in the blood. Typical lesions of East Coast Fever were present.

Calf, No. 27.—The fifth injection was made on 1st December, and a sixth on the 12th December. Fever started on the 18th December, *i.e.*, after 28 days' exposure. The animal died after an illness of 11 days. Both piroplasmata were found in the blood smears. Typical lesions of East Coast Fever were present.

Calf, No. 28.—The fifth injection was made on the 1st December, and a sixth on the 12th December. Fever began on 15th December, *i.e.*, after 25 days' exposure. The animal died after an illness of 14 days. The piroplasma of East Coast Fever only was present in the blood smears. The lesions were typical.

Calf, No. 29.—The fifth and sixth inoculations were respectively made on the 1st and 12th December, 1903. Fever started on the 10th January, 1904, *i.e.*, after 15 days' exposure. The disease lasted 13 days, and ended in death. Both piroplasmata were found in the blood smears. Lesions were typical of East Coast Fever.

Calf, No. 30.—The fifth inoculation was made on the 8th December, *i.e.*, after an exposure of 18 days. Death took place on 20th December, or after an illness of 12 days. Both piroplasmata were present in the blood smears. Lesions typical of East Coast Fever.

Control Experiments.—Along with the nine inoculated calves, five non-inoculated Texas calves of the same shipment were exposed on the 20th November. The results were as follows:—

Texas Calf 31.—Fever began on the 10th January, *i.e.*, after 51 days' exposure. The animal died after an illness of 11 days. Both piroplasmata were present in the blood smears.

Texas Calf 32.—Fever began on the 28th December, *i.e.*, after 28 days' exposure. The disease ended fatally after 15 days. There was a strong double infection with both piroplasmata in blood smears.

Texas Calf 33.—Fever began on 6th December, 16 days after exposure. Death occurred on 21st December, after 15 days' illness.

Texas Calf 34.—This animal was killed on 25th December, on account of poverty after 35 days' exposure. It had never shown any reaction, and on *post-mortem* proved to be free from any disease.

Texas Calf 35.—This animal showed fever on 30th December, after 40 days' exposure, and died 13 days later from typical East Coast Fever.

Texas Calf 36.—Fever set in on the 3rd December, after 13 days' exposure. The animal died on the 16th December, after 13 days' illness.

In all the above cases, with the exception of 34, the typical lesions of East Coast Fever were present.

Conclusions.—It will be seen that, of the inoculated animals, eight died of East Coast Fever; one survived. The average time of exposure before they contracted the disease was 30 days. The non-inoculated animals (the one which was killed after 35 days' exposure, and which was found to be healthy, may be taken into account) also sickened after an average exposure of 30 days. We must conclude, therefore, that the inoculation, which was repeated six times in the majority of cases, had not produced any immunity. The long time which elapsed before the herd was wiped out must be explained in another way than

by accounting for it by the protective effects of inoculation. We have repeatedly observed the same thing in herds upon infected pastures where no inoculation had been tried. There is more than one possible explanation for this apparent irregularity, but at present we are without exact knowledge. It may be due to a paucity of infected ticks, or, what amounts to the same thing, the parasites for some unknown reason may not always develop freely inside the tick host. We know for certain that nymphæ infected as lurvæ do not so surely infect cattle as adults infected as nymphal. We also know that at certain seasons the ticks do not show the same tendency to bite. Ticks travel but a short distance on their own legs; the infection of pastures, therefore, occurs in patches, and one can easily imagine that an animal may graze for months without coming in contact with an infected patch. The same thing is observed in anthrax and quarter-evil. This matter, however, is further dealt with in Article IV.

We consider the survival of Calf 24 as a simple coincidence, considering that the mortality in this disease is about 95 per cent., and we have seen exactly the same thing take place repeatedly where inoculation did not occur as a factor to be considered. We admit that in the face of Dr. Koch's latest statement, "that immunity can only be expected after five months, during which the inoculation has to be kept up" the foregoing experiments prove nothing against his latest assertion. The experiments show, however, that on infected ground the inoculation would be of little value, since the animals under treatment died off as rapidly as did the controls, and so it would be with animals under treatment on infected pastures.

The experiment of repeatedly inoculating cattle, according to Dr. Koch's latest pronouncement during five months, was undertaken with 16 animals. During this period they were injected 13 times, and were kept on non-infected ground.

The exposure took place only at the end of June, 1904, so that results cannot yet be expected.

The question of immunity, however, after that time has, we think, been settled by the trials in Rhodesia, the results of which were communicated to the South African Conference of Veterinary Surgeons, at Capetown, by Mr. Gray. The Conference recorded its opinion in the resolution already alluded to, and by another, as follows:—

> "That this Conference, after considering the reports of the scientists who have had practical experience of the effects of inoculation as proposed by Dr. Koch, is reluctantly compelled to the conclusion that it will be vain to trust to inoculation to arrest the spread of African Coast Fever."

III.

Dipping Experiments.

Although at the time when East Coast Fever first appeared in the Transvaal the particular species of ticks which could communicate the disease were not known, yet everything pointed to the fact that the disease must be carried by ticks. Accordingly, it was thought that by destroying ticks the spread of the disease might be arrested. From the very first appearance of the disease advice was given to dip or wash the cattle with anti-parasitic fluids, and then remove them to fresh, non-infected areas. In this way many herds were freed from the disease, but this was probably due more to changing the pastures than to dipping as such. It was, indeed, observed that changing the pastures

without any dipping was followed by similar success, whereas, dipping without change of pasture did not seem to stop the outbreaks in infected areas. There was much controversy about the effects of dipping in infected areas, and also regarding the effectiveness of the dips used. As no experimental evidence was available regarding the efficiency of dipping as a preventive for East Coast Fever, we decided to carry out some experiments with different dips in a badly infected area, in order to settle the points in dispute. The Anglo-American Stock Trading Company kindly handed over to us a number of imported Texas cattle, derived from parts of that country known to be badly infected with Texas Fever. Their object was to ascertain whether their animals would prove to be immune against East Coast Fever. The cattle to be experimented upon were divided into lots of 10; one lot was to be dipped regularly, and the other was to serve as a control—that is, they were not to be dipped at all. The experiment started in October 22nd, 1903, at Nelspruit, on a farm which we knew, from previous experience, to be badly infected. All the cattle had up to that date been kept in a clean kraal in Machadodorp, and had been fed on imported lucerne only. They were brought to Nelspruit by rail, and directly after off-loading from the trucks they were sprayed by means of a hand spray. In order to obtain thorough soaking of the skin the animals were made to lie down. The spraying was repeated every eight days. The materials used for dipping were (*a*) paraffin, (*b*) arsenical solutions, (*c*) izal and arsenice.

1. Paraffin finely divided in water.—The mixture was obtained by a spray pump, called the "Success" spray pump, and the proportion of paraffin to water was one to four, 25 per cent. paraffin. Two animals were treated in this way, Nos. 7 and 10.

Result.—No. 7 came into fever on the 5th November, viz., after 15 days' exposure; it had been sprayed twice during that time. The fever lasted up to the 18th November—13 days—when the animal died of typical East Coast Fever. The infection with piroplasma was a very strong one.

No. 10 started to react after the same period of exposure, and died on the 15th November, *i.e.*, after an illness of 10 days' duration. This animal had also been dipped twice during the exposure. On the day of the death about 50 per cent. of all red corpuscles were infested with piroplasma.

Conclusions.—Taking into consideration that on the date of the first dipping no pathogenic ticks could have been on the cattle, and that the incubation time of the disease averages 12 days, we came to the conclusion that the dipping did not materially retard the infection. A careful examination was made every day after dipping. It was found that only a few ticks were present on the upper parts of the body, and that most of them were dead. On such parts, however, as the tip of the tail, the heels, and in between the hoofs, living ticks could constantly be found. The brown tick was at that particular period of the year conspicuous by its scarcity.

2. Arsenical Dips.—Demuth's modification of the Queensland dip was used. This modification allows the material to be used in solution with cold water, whereas the original formula had to be dissolved in warm water. The solution was made at 1 to 25 of water, it being considered by the maker that in a solution of 1 to 30 the efficacy would be the same as that of the original formula. The dip was applied by means of the Douglas Spray Pump. Six animals were thoroughly sprayed in the way before indicated.

Result.—Texas Calf, No. 3.—Fever began on the 5th November, after 15 days' exposure to infection. During this time the spraying was carried out twice. The animal died on the 19th November, after an illness of 14 days. The infection with piroplasma of Coast Fever was a very strong one.

Texas Calf, No. 4.—This animal also became sick after 15 days' exposure, and it died after an illness of 14 days. Piroplasmata were very numerous on the day of death.

Texas Calf, No. 5.—This animal sickened after an exposure of 25 days, and it died of East Coast Fever after an illness of 14 days.

Texas Calf, No. 6.—The reaction started on the 15th day after exposure, and death resulted 14 days later, on the 19th November. There was a strong infection with piroplasmata.

Texas Calf, No. 8.—This calf started to show fever 47 days after exposure. The disease lasted 14 days. The animal died on December 19th with a strong infection of the red corpuscles. The calf was sprayed four times, when spraying had to be discontinued on account of the blistered condition of the skin.

Texas Calf, No. 9.—The fever began on the 14th day. Death resulted on the 15th November, 1903, after an illness of 11 days.

Conclusions. — Out of six animals which were sprayed with arsenical solution, two contracted the disease after 14 days, two after 15 days, one after 25 days, and one after 47 days. In four animals the dipping had no marked effect; it did not retard the infection. It apparently did so to a slight extent in one case, and distinctly in the case of No. 8.—But it must be remembered, as we have already pointed out, that some animals which are not dipped escape infection for a much longer period than others, and, therefore, their survival may be due to other causes than dipping.

3. Experiments with arsenical dip and an addition 10 per cent. raw Izal. The mixture was also applied with a Douglas Spray Pump. Two animals were treated, Nos. 1 and 2.

Result.—Texas Calf, No. 1, sickened after an exposure of 14 days, and died after an illness of 9 days with a strong infection of Rhodesian Tick Fever piroplasmata.

Texas Calf, No. 2.—Died from East Coast Fever after an exposure of 24 days and an illness of 15 days.

Spraying every eight days with arsenical solution in the strength used so blistered the skins of the animals that it could not have been kept up indefinitely.

Conclusion.—The additiou of Izal did in no way increase the effectiveness of the arsenical dips.

IV.

RESULT WITH THE (NON-SPRAYED) CONTROL ANIMALS EXPOSED AT THE SAME DATE.

	Time of Exposure.	*Disease Lasted.*
No. 11.	25 days	15 days.
No. 12.	12 days	16 days.
No. 13.	12 days	16 days.
No. 14.	14 days	11 days.
No. 15.	14 days	12 days.
No. 16.	14 days	12 days.
No. 17.	Died 21 days later from exhaustion.	
No. 18.	10 days	10 days.
No. 19.	24 days	14 days.
No. 20.	28 days	17 days.

A comparison of the average incubation time in the sprayed cattle with the average incubation time in non-sprayed cattle should give some indication whether the dipping can be considered to have warded off infection. It averaged in the first instance 16 days (out of 10 cases), and in the second instance 17 days (out of nine cases). Thus it appears that dipping in a badly infected area has no effect whatever if the animals continue to be exposed to infection; the non-dipped animals have even a longer average incubation time than those dipped. Although the average incubation time does not demonstrate that the dipping exerted any influence, it might be argued that if the shortest periods of incubation are taken into consideration, then a slight inhibitory influence is noticeable in some instances. It is, however, slight enough to be negligible, as at the utmost it can only be calculated as a difference of one day.

There is also an apparent slight variation in the duration of the disease in dipped and non-dipped cattle. The period of sickness amounts to an average of 12·8 days in the former, and to 13·3 days in the latter. The difference, however, we consider too small to justify the conclusion that the dip had any influence on the duration of the disease.

For practical purposes we consider that we have demonstrated that the application of dips in a badly infected area gives no guarantee against the further spread of the disease, and no hope of extinguishing an outbreak unless the animals be also removed to fresh pastures. The latter operation, however, incurs the serious risk of infecting the route over which the animals are moved, not to speak of the new pastures.

In coming to these conclusions we do not wish it to be understood that we are foes to the general idea of dipping cattle. We agree with those who hold that systematic dipping will reduce the number of ticks on a farm, but it is principally the blue ticks, and those with a similar life history which are affected. The blue tick remains on an animal for the best part of a month, whereas the intermediate stages of the brown tick may drop off in three days.

The dipping materials at our disposal do not, in our experience, keep ticks off for any appreciable time, and it is not practical to dip cattle say every fourth or fifth day in an irritating and poisonous bath.

We are also far from advising private individuals against erecting a dip on their farms for the exclusive use of their own cattle. The brown tick, on account of its short sojourn, is difficult to catch by the periodical dipping possible. It is only, however, reasonable to suppose that a few will be caught and destroyed at each dipping, and that years of the dipping regime will reduce the number of brown ticks on a farm, but we doubt if the reduction will be a very material one, and we certainly do not think that anything approaching annihilation will be accomplished, for be it remembered that many other animals than cattle act as hosts for the development of the tick in question, although they do not infect it with disease.

We are of opinion that those who are inclined to trust to dipping to keep tick fever off their farms are leaning on a broken reed. If a man fences a clean farm, keeps his cattle on the place, and only brings in fresh animals after they have undergone a period of quarantine for six weeks in a shed or special paddock, we do not think he need greatly fear tick fever even in an infected district. If, however, he insists on doing transport with his oxen on dangerous roads, his animals will sooner or later pick up the disease and bring it on to his farm in spite of dipping.

In conclusion we wish to express the opinion that the erection of a few dipping tanks in a district for common use is a dangerous policy. The pastures around these dips are very likely to become badly infected, and cattle coming

to the tanks may pick the infection up. If a farmer has a clean farm the soundest policy he can follow is to rigidly keep his cattle upon it, and if a herd is infected, or suspected of being so, it is in the interests of the community not to move them over other farms for dipping or any other purpose.

V.

Possible Influence of the Different Seasons on the Outbreaks of East Coast Fever.

During the years 1903 and 1904, several lots of cattle were exposed in Nelspruit, which has been repeatedly alluded to as a very badly infected area. The number of animals exposed in the different experiments amounted to 89, of which, so far, only one has recovered. The different lots were exposed during different months of the year. We observed that the shortest average time required to wipe off a lot of 23 cattle was 22 days and 19 hours. This is the shortest period on record, and may serve for comparison with the periods after which other lots died during the various months. We obtain in this way, some indication of the influence the season may have on the development of the disease.

1st Lot.—Exposed on the 14th January, 1903, 23 animals. Time of exposure before last animal died, 22 days and 19 hours.

2nd Lot.—Exposed on the 8th March, 1903, 17 animals. Average time of exposure before death, 24 days 11 hours.

3rd Lot.—Exposed on the 3rd April, 1903, four animals. Average time of exposure before death, 25 days and 18 hours.

4th Lot.—Exposed on the 25th July, 1903, six animals. Average time of exposure before death, 39 days 16 hours.

5th Lot.—Exposed on the 22nd October, 1903, 19 animals. Average time of exposure before death, 31 days 7 hours.

6th Lot.—Exposed on the 20th November, 1903, 13 animals. Average time of exposure before death 44 days 3 hours.

7th Lot.—Exposed on the 7th January, 1904, 8 animals. Average time of exposure before death, 25 days and 10 hours.

Conclusions.—The shortest period required to wipe out an exposed herd of cattle, numbering 23 head, is about 23 days. This happened in the months of January and February. During the following months, March and April, the period varies only by one or two days. The delay begins in June and is noticeable up to the end of the year. The longest period of exposure required was in November and December. It is a rather startling observation that the infection was so long retarded in these two months, during which the temperature is so favourable for the development and moulting of ticks. A careful examination of the cattle, however, during these periods was constantly made, and it revealed the remarkable fact that the brown ticks were the reverse of plentiful. The veld had been burnt during the winter. During the month of October, the grass had hardly sprung up, and it was very short during the following months. As soon as the grass had fully grown, the ticks became more frequent, and were then found to be present in the ears in large numbers. The experience at Nelspruit seem, therefore, to indicate that the rapidity of infection is in direct relation to the growth of the pasture. This might be explained by the observation that the ticks, in order to reach a host, climb to the tops of grasses from which they have every chance of attaching themselves to animals, hence the scarcity of cattle when the grass is short and their frequency when the grass is long. We may further conclude that the greater the number of ticks which infest a beast, the more is the chance of there being pathogenic ones amongst them. Accordingly, the chances of rapid infection

are greatly increased when the grass is long. This probably helps to explain the common observation that grass-burning has a decided influence on those diseases which are carried by ticks, but, of course, many of the carriers must be destroyed by the fire.

This observation, made in the low country, does not exactly coincide with the number of outbreaks which occurred in other parts of the Transvaal in the various months, but it must be borne in mind that grass-burning is not practised at the same time in all parts, nor are the other conditions, which influence tick life, seasonably symetrical.

The Danger of the Simultaneous Immunisation with Serum and Virulent Blood for Rinderpest in Cattle not Immune against Redwater.

I.

The method of inoculation commonly known as the simultaneous one, was introduced by Turner and Kolle, and was, at one time, very much used in South Africa. This method consists in the injection of a standardised serum on one side of the body, and of a small quantity of virus on the other side. The result is that, after an incubation time of, usually, five days, a typical reaction follows, which may be very slight or is accompanied by all the symptoms which characterise rinderpest. The percentage of mortality from rinderpest was small where serum has been used whose immunising properties were previously known. The advantage of this method is the active immunity which is produced through the reaction. This immunity lasts as long as the one acquired by recovery from the disease naturally contracted. The method is advantageous in countries where rinderpest has become endemic.

The method has, however, one great disadvantage, since with the injection of the virus as blood, diseases due to blood parasites may be transmitted. The danger is very great in such countries where blood diseases, such as redwater, trypanosomiasis and heartwater exist. This is the case in the Transvaal and in several other parts of South Africa. During the last rinderpest outbreak in the Transvaal, 1901 to 1903, the danger of the simultaneous injection of serum and blood became apparent at the Rinderpest Station, Pretoria. During this period, the total number of animals hyper-immunised against rinderpest amounted to 452. They were all injected in the way indicated, and, with the exception of 30, were all intended for hyper-immunisation. These cattle may be divided into three lots :—

1. Cattle born and bred in parts of South Africa known to be redwater infected.
2. Cattle born and bred in parts of South Africa, free of redwater.
3. Cattle imported from oversea, from countries where redwater does not exist.

This classification will allow us to draw exact conclusions as to the role redwater plays in connection with blood injections. It is a well-known fact that cattle born and bred in redwater infected areas are immune against the disease, and that the injection of such blood into susceptible cattle causes redwater. The disease may be so slight that hardly any mortality is noticeable, whereas in other instances, a heavy mortality may result.

These facts, borne in mind, make it plain that—when the simultaneous method, in which blood of redwater immune cattle is used as rinderpest virus—redwater must be expected in animals not immune against this disease. Further,

it must be expected that the mortality of cattle thus treated is greater than when blood alone is injected, which does not contain the redwater virus. Such an animal is suffering from two diseases, rinderpest and redwater. The incubation period of the former being shorter, rinderpest lesions have time to fully develop. When the reaction is nearly over, the incubation time of redwater has passed also and, in addition to the former disease, this latter one makes its appearance. And the result is generally death.

The second South African disease, which plays an important role in connection with blood injection, is that due to the trypanosoma Theileri, I have frequently found this parasite in oxen which were inoculated against rinderpest by the simultaneous method, and more often in oxen which were hyperimmunised. It produces a high fever, which may be accompanied by an acute anæmia. The animal refuses to feed, rapidly loses condition and dies. The disease is not, however, fatal, at least in the majority of cases; the anæmia takes a chronic course and the ox becomes very poor. It may then die of poverty, long after the trypanosoma has disappeared out of the blood. There are a good many animals which, apparently, do not suffer from the presence of the trypanosoma and notwithstanding the fever, which is always present. These animals show as a rule, but few trypanosomes, so that they may easily escape microscopical examination. The difficulty to exclude them under all circumstances is, therefore, evident. When rinderpest reaction is accompanied with this trypanosomiasis, the anæmia becomes more accentuated and debility often ensues, even after the animal has, to all outward appearances, passed through a slight attack of rinderpest. The blood of oxen which have recovered from an attack of this trypanosomiasis, is no longer virulent; thus the danger of infecting healthy cattle by means of the simultaneous method is not so great as that in the case of immune redwater blood.

Heartwater is a third disease which may probably be a factor in complicating rinderpest inoculation. It is also inoculable with blood from sick into healthy susceptible cattle. There will be much difficulty in diagnosing this complication, inasmuch as the characteristic lesions are those of acute gastro-enteritis, similar to those of rinderpest. Only when the prominent symptom—that of an increased collection of liquid in the pericardial bag is present—the disease may be suspected. The danger, however, is not so great, and exists only in certain parts of the country which correspond with the distribution of the bont tick, and in cattle which have been born outside those districts and have recently been introduced.

A fourth disease which, at the time of the first rinderpest outbreak, had, in certain regions, to be taken into consideration, was the tsetse fly sickness, caused by the Trypanosoma Brucei. In the blood of oxen which are bitten by the tsetse fly, Glossina morsitans, the trypanosoms may live for many months, during which time the microscopical examination may fail to detect them, and only the injection of such blood into dogs will allow a certain diagnosis. It was in the Portuguese territories that the tsetse disease was observed to have been communicated by the injection of defibrinated blood.

A fifth disease which I have noticed in connection with rinderpest inoculation is the Spirillosis. Spirillum Theileri is, however, not inoculable with blood in which it is found. It is, therefore, somewhat difficult to understand how the parasite came into the blood circulation of the inoculated animal. These cases have, in my opinion, to be considered as pure coincidences.

I am not prepared to state that the above mentioned diseases complete the number of possible complications; I am inclined to believe that there are others which we are yet unaware of.

Return of Outbreaks of Redwater after the Simultaneous Rinderpest Inoculation.

A.—Cattle born and bred in redwater-infected areas.

	Inoculated.		*Redwater Symptoms.*	*Died.*
1.	7	animals	2	2
2.	9	„	0	0
3.	11	„	0	0
4.	6	„	0	0
5.	9	„	0	0
6.	18	„	2	2
7.	11	„	0	0
8.	6	„	0	0
9.	21	„	0	0
10.	9	„	0	0
11.	9	„	0	0
12.	7	„	0	0
13.	16	„	1	0
14.	5	„	0	0
15.	8	„	0	0
16.	15	„	0	0
17.	9	„	0	0
18.	4	„	0	0
19.	5	„	0	0
20.	5	„	0	0
21.	15	„	0	0
24.	9	„	0	0
29.	3	„	0	0
30.	30	„	1	1
32.	7	„	0	0
33.	16	„	0	0
34.	38	„	0	0
35.	5	„	0	0
36.	8	„	0	0
37.	7	„	0	0
	328	animals	6	5

Result.—The percentage of cattle which contracted redwater from the simultaneous injection is 1·8 per cent.; the mortality was 1·5 per cent.

B.—Cattle born and bred in districts of South Africa known to be free of redwater.

	Inoculated.		*Redwater Symptoms.*	*Died.*
Lot 25.	11	animals	3	2
Lot 26.	9	„	2	2
Lot 27.	34	„	5	3
Lot 28.	20	„	20	13
	94	animals.	30	20

Result.—The percentage of cattle which contracted redwater from the simultaneous inoculation was 32·9 per cent., and the morality was 21·2 per cent.

The cattle of the above-mentioned different lots came from Aliwal North, a district where redwater is not known to exist.

C. Cattle imported from Oversea, from a country free of redwater.

Inoculated.		*Redwater Symptoms.*	*Died.*
Lot 22.	8 Calves (Shorthorn) ..	3	3
Lot 23.	22 Heifers (Shorthorn) ..	22	11
	30 animals	25	14

The percentage of animals contracting redwater or, at least, of animals which showed distinct redwater was 83·3 per cent., and of mortality was 46·6 per cent.

Calves are known to have some resistance against redwater; they certainly do contract it, but rarely die from it. In the abovementioned experiments, the calves were not under as close observation as were the heifers, and it is quite possible that all of them had an infection of piroplasma bigeminum, which, however, escaped our observation. The heifers, which were about two years of age, suffered from redwater to the extent of 100 per cent. and the mortality was 50 per cent. The history of these heifers is further interesting. Two samples of blood were used as virus. The first sixteen animals were inoculated with blood from an ox, which, on microscopical examination, proved to be absolutely free from blood parasites. Notwithstanding this, thirteen animals showed the trypanosoma in their blood. The second lot from 17 to 22, were pure redwater infection. Out of 16 animals, nine died (56·2 per cent.), and out of the six remaining, two (33·3 per cent.). It was the double infection both with trypanosoma and piroplasma, which killed the cattle after they had passed through the rinderpest reactions.

The mortality from redwater and trypanosomiasis, calculated on the total of all cattle treated with the simultaneous injection of rinderpest blood, was 8·6 per cent.

The shorthorn heifers were imported from the Argentine Republic, and were born and bred in the province of Buenos Aires, which, to judge from the publications of Lignieres, is free from tristezza, viz., redwater.

These cyphers speak for themselves. The danger of producing redwater by the simultaneous method is not great when cattle are injected which are born and bred in infected countries. It is very great in cattle which come from regions where redwater does not exist.

II.

Symptoms and Pathological Changes observed in Rinderpest complicated with Redwater.

We may divide the cattle suffering from the two diseases into four different lots :—

1. Cattle which died within the period of the typical rinderpest reaction.
2. Cattle which died shortly after the rinderpest reaction had subsided.
3. Cattle which died at some period after the rinderpest reaction.
4. Cattle which recoverd.

Ad. 1.—The reaction of rinderpest usually begins on or about the third day; sometimes the fourth; rarely the fifth; and more exceptionally on the sixth day after inoculation. During the first few days of the reaction, nothing seems to be amiss with the animal. Towards the end of the reaction, which lasts from five to eight days, the symptoms of rinderpest become visible. The animal will not feed for a day or two, there is a slight running from the eyes, nose and mouth, and diarrhœa. The excreta are sometimes mixed with mucous and blood. The diarrhœa usually lasts a day, rarely longer, and the recovery begins withs its disappearance. When redwater is present, the symptoms are aggravated. The animal is then very ill, it rapidly loses condition, there is a diarrhœa of pure arterial blood, which, when discharged is, as a rule, not coagulated; in other cases big clots are noticed. There is sometimes copious bleeding from the nose. The urine becomes dark in colour (Met-hæmoglobinuria). The animal dies. On post-mortem, the lesions found are those of redwater and rinderpest, but the hæmorrhagic inflammation of the intestines is more pronounced than that met with in a grave case of rinderpest. The cœcum is engorged with a big clot of coagulated blood. Long clots are also found in the colon and rectum. The mucous membranes of the intestines are strongly congested; they present a uniform hæmorrhagic aspect throughout their whole length. The spleen is enlarged. The kidneys are black. The lungs are sometimes in a state of acute œdema and contain hæmorrhagic infarcts.

Ad. 2.—Cattle which died shortly after the rinderpest reaction, show a more or less pure picture of redwater; the rinderpest lesions are but little marked. The animal dies of acute anæmia, caused by piroplasma bigeminum.

Ad. 3.—Cattle which died some time after recovery from rinderpest, show the symptoms of chronic redwater. The anæmia is very marked and a general jaundice is present. The liver is yellow and friable; the bile is thick, dark green, brown or yellow. The spleen is still enlarged. The kidneys are pale. The urine clear. The blood is watery; the piroplasma has disappeared in almost every instance, but there exists poikylocytosis, microcytes and macrocytes; many of them with nuclei and typical basophile granulations.

Ad. 4.—The animals which recover show the same microscopical lesions of the red corpuscles as those described above. The oxen become very poor in condition, they waste away almost to a skeleton. Some of them do not recoup, and those which really recover, require many months ere they are again fit for work.

The rinderpest reaction in which trypanosomes were found was, in the majority of cases, also accompanied by the piroplasma bigeminum. In these cases the symptoms of redwater were predominant. In redwater immune oxen, trypanosomiasis became noticeable when the temperature, due to rinderpest, did not drop after the typical reaction was over. It kept on for several days, and the animal lost condition. Only a few deaths, due to the trypanosoma, were recorded. They showed the peculiarity that the rinderpest lesions in the bowels were not healed out, there was still necrosis of the mucous membranes of the small intestines and particularly of the Peyer's patches. Also profuse hæmorrhages in the bowels were noticed. The animal died of anæmia.

Conclusion. The simultaneous method of inoculation against rinderpest is dangerous, when cattle, which are not immune against redwater, are inoculated with blood of immune oxen. It will be difficult in practice to separate a redwater immune ox, since the piroplasma of redwater cannot always be detected by microscopical examination. These points guided South African experts, at the Bloemfontein Conference, in not recommending the simultaneous method;

but in advocating serum alone for such outbreaks where cattle can be quarantined, and repeating the inoculation of pure serum as long as the disease is found in the herd. In certain cases where active immunity is essential, as, for instance, during the late war, when cattle had to be shifted rapidly from place to place, the inoculation with pure bile, as recommended by Professor Koch, was considered to be the safer method.

Notes on Piroplasmosis of the Horse, the Mule and the Donkey.

I.

In former publications on biliary fever of the horse, I have described the micro-organism of this disease, which is known under the name of Piroplasma Equi (Laveran). In those papers I have stated that I was not able to produce the disease by inoculation of blood taken from a sick animal into healthy animals. The explanation of the failure was, that at that particular time, I could only experiment with horses which were born in South Africa, and were, therefore, naturally immune against this disease. This conclusion was arrived at by the analogy with other diseases caused by piroplasma, as, for instance, our ordinary redwater. It is a well known fact that calves are less susceptible to redwater and, when brought up in redwater infected areas, become immune to this disease. The analogy with redwater has led me to a further point, viz., to ascertain whether a horse, which has recovered from an attack of biliary fever and has acquired immunity through that attack, carries the piroplasma in its blood; and whether the injection of such blood would produce the disease in a susceptible healthy animal. I have described the form which piroplasma bigeminum takes in an immune ox. It has not the typical pear-shaped appearance, but appears mainly as rings and rods. I have not been able yet to trace the piroplasma equi in the immune horse, but that it must be present is borne out by the following experiment. Two years ago, a horse was suffering from biliary fever. The presence of the piroplasma equi was at that time demonstrated. The horse was treated and recovered. On April 30th, 1904, this animal was tapped. 10 c.c. of defibrinated blood was injected subcutaneously, and 10 c.c. into the jugular vein of a horse (No. 491). This horse came from Johannesburg; it had been imported some years previously from England and was constantly kept in a stable. There was, accordingly, every reason to believe that it would be susceptible to biliary fever. The morning temperature was, eight days later, a little higher than during the previous days, and the animal was slightly sick. The next day a jaundiced condition of the mucous membrane of the eyes was noticeable, and an examination of the blood proved the presence of the piroplasma equi. A serious turn, however, only took place on the twelfth day after inoculation, the morning of which date, when the temperature rose to 104·6 and alarming symptoms of biliary fever became plain. The typical parasites and a good number of rosettes were constantly present up to the date of death, which occurred during the night from the 15th to the 16th May, 1904.

The *post-mortem* was made on the morning of the 16th May, 1904. General condition of the carcase was fair. The colour of the muscles was normal. The fat was slighly tinged with yellow. The blood was normal. The lungs were œdematous, and the interlobular spaces were infiltrated with yellow liquid. There was a fair amount of liquid in the pericardial sack. Petechiæ were

scattered over the external surface of the auricles and about the auricular-ventricular furrow. The endocardium of the ventricle showed well-marked petechiæ and hæmorrhagic patches. The mucous membrane of the stomacch was highly congested and showed several prominent hæmorrhagic patches. The mucous membrane of the small intestines was thickened, yellow in colour and covered with mucous. The spleen was greatly enlarged and congested; the substance soft, breaking down on section. The liver was slightly enlarged, with fibrous adhesions on the capsule. The kidneys were slightly congested.

Conclusion.—Horse 491 died of typical equine piroplasmosis. The disease was produced through the injection of blood taken from a horse which, two years previously, had recovered from an attack of the same disease. The incubation period lasted seven days.

The piroplasmosis of horses behaves similarly to the ordinary piroplasmosis (redwater) of cattle, and also to the piroplasmosis of the dog (malignant jaundice).

II.

In former communications on this subject, I observed that I had not, up till that time remarked the piroplasmosis in mules and donkeys.

After the Repatriation Department had distributed imported mules and donkeys in the various districts of the Transvaal, several reports reached the station that these animals died in rather large numbers from an unknown disease. Thanks to the co-operation of Mr. Conacher, District Veterinary Surgeon for Lydenburg, and of Mr. Thomas Dale, Veterinary Surgeon to the Repatriation Department, material was obtained for microscopical examination. The latter gentleman has published his experience in the sixth issue of the *Agricultural Journal.* Personally, I had the opportunity of making several *post-mortem* examinations. The preparations made with blood from sick donkeys, and especially those made from the spleen, showed the presence of a typical piroplasma, which was found in all acute cases of the disease. This endoglobular parasite shows itself usually as a round disc, rarely in the pear-shape of the piroplasma bigeminum or piroplasma canis. The specific chromatic stains, such as Laveran's and Mackonkey's modifications of Romanowsky's method, also Azur II., show a clearly defined karyosoma, which is always situated on a marginal segment of the disc. The cytoplasma usually shows a clear zone round the nucleus, and the remainder takes a slightly blueish tinge. The parasite varies in size considerably, from $\frac{1}{8}$ to $\frac{1}{4}$ of a red corpuscle. There are very small discs, with almost no protoplasma, and also large discs. The former are probably young parasites. This may be explained, seeing that the large forms split up into four sections, each section containing a karyosoma. They are then found resembling a rosette. The sections—the young parasites—soon detach themselves and pictures appear, somewhat resembling a shamrock leaf; whilst in a few corpuscles, the young parasites are irregularly distributed.

The same observation is made with the piroplasma of equine malaria. From the morphological appearance of the parasite, I have no hesitation in declaring it to be identical with that of the horse. Laveran of Paris, to whom I sent blood specimens from a sick mule, confirmed my opinon.

This piroplasma is found in donkeys and mules only during the acute stage of the disease. The rosette forms are, as a rule, present in most instances. I never missed them in smears of the spleen. They are also found in the liver, and sometimes very frequently in the kidneys.

The chronic forms of piroplasmosis in donkeys and mules were, in several instances, accompanied by a secondary infection, which I have termed the sequelæ of biliary fever in horses. The serum of several donkeys and mules agglutinated the bacteria, which I then isolated from the enlarged spleen of the horses.

Description of the Disease.

The piroplasmosis of the donkey varies in the clinical aspect from that of the horse. In the horse, a never failing symptom is a general jaundice. This is markedly absent in the donkey, in which the symptoms of an acute anæmia are dominant, characterised by a very pale mucous membrane of the eyes, which are frequently discoloured by blood spots. The fever is, as a rule, high, and may reach as much as 106° F. Notwithstanding this, a good appetite is present and, judging from this symptom, the animal seemed in perfect health, but in several instances, it was found dead the next day. Those were the cases in which the parasites were most frequent.

The *post-mortem* lesions found in cases of the acute disease, resemble those found in equine malaria, save for the absence of jaundice. In most cases, the tumour of the spleen was the prominent lesion. Its pulpa was soft. The lungs were sometimes in a state of œdema. There was also a collection of liquid in the heart-bag. Petechiæ were found on the surface of the heart and on the endocardial lining of the left ventricle. A gelatinous infiltration of the capsula rhenalis was also noticed. The large organs, such as heart, liver and kidneys, were sometimes found in a state of parenchymatous disintegration. Usually, the symptoms of a slight catarrh of the mucous membrane of the intestines were present; sometimes the mucosa of the stomach was in a stage of hæmorrhagic inflammation.

The clinical symptoms in acute piroplasmosis of the mule are intermediate between those of the donkey and the horse. Jaundice is not always present; but the discolouration of the mucous membrane of the eye is a fairly constant sign. Fever is always present. Loss of condition is usually observed, and nervous symptoms are also found. The animal may be found to have a drowsy appearance, and is often weak in the hind quarters. Copious staling is often noticed. Alarming symptoms, indicating heart failure, which is characterised by a quick and feeble pulse, accelerated respiration, often accompanied the lethal issue. In others, death was proceded by coma, in which state the animal was found for some hours before it died.

Post-mortem lesions correspond with those found in the donkey and also in the horse. Jaundice may be present or absent. The lungs may be found œdematous, the intralobular tissues separated by an infiltration of liquid. There may be liquid in the heart-bag, which has often a brownish colour. The surface of the heart and the lining of the left ventricle usually show petechiæ, which are sometimes very frequent. The blood has a pale appearance. The spleen is usually enlarged. The lymphatic glands are also enlarged. The symptoms of a catarrh may be present in the stomach and the intestines. There are also sometimes œdematous infiltrations of the rhenal capsula and of the mesentery.

Chronic piroplasmosis of the doneky is chiefly characterised by an increasing anæmia and poverty. The animals show a peculiar tendency to walk in a straight direction, they are somnolent and do not notice their surroundings. They often walk until they meet a barrier, against which they stand for some time, or they may tumble into sluits, in which they are often found dead.

The tonus of the anus is relaxed. There appears, sometimes, an eczema on the back and on the legs, where the hair will fall out. The animals have, as a rule, a good appetite, notwithstanding which, they lose condition.

The blood, taken in chronic stages, did not contain the piroplasma. The *post-mortem* lesions found in chronic piroplasmosis are those of anæmia and dropsy. There is usually a collection of yellow liquid in the peritoneal cavity. The pericard and the base of the heart are full of the same liquid, the sulci longitudinales and transversales of the heart are infiltrated with liquid. The heart muscle is flabby and friable. The blood coagulates in long fibrinous clots. Tumour of the spleen is also present. The liver has a pale appearance and is often cirrhotic. The capsula of the kidneys are found infiltrated with yellow liquid. The mucous membranes of the stomach and intestines are often found in a state of chronic catarrh, swollen, covered with viscid mucous, and show slate-coloured patches and streaks. The urinary bladder is usually found distended with clear urine.

The *post-mortem* lesions of chronic piroplasmosis of the mule, are, generally speaking, similar to those described in the donkey.

Experimental inoculations with the blood of sick donkeys were made in order to produce the disease in horses. They failed. This is probably because the horses were immune against biliary fever. I consider the piroplasmatic diseases of horses, mules and donkeys are identical. The variation of the symptoms, both during life and on *post-mortem*, is only slight and is not sufficient to declare the diseases as distinct. The conclusive proof, however, viz., that of producing the disease in the horse with virulent mule or donkey blood or *vice versa*, is still wanting.

At the present moment, I have no experiments to show in what way the disease is propagated. But considering the other maladies caused by piroplasmas, we may naturally conclude the tick is the real disseminator of the disease. Further experiments will have to be conducted, in order to discover what species of ticks are the agents in the transmission of the disease.

I.

Notes on the Immunity of the Piroplasmosis of the Dog.

The piroplasmosis of the dog, known by the name of malignant jaundice, or biliary fever, is a common disease of South Africa, due to the presence of an endoglobular parasite, known as piroplasma canis. Thus the disease is related to redwater of cattle, East Coast Fever, biliary fever in horses, mules, and donkeys, all of which are caused by a specific piroplasma.

Malignant jaundice of dogs is transmitted by a species of tick, Hæmophysalis leachi (Audouin), and this fact points to a striking similarity to redwater. A study of the immunity of dogs against this disease will, therefore, throw some light on the immunity of the whole group of the diseases which are caused by the larger piroplasms.

Infectious diseases are caused either by bacteria or by protozoa. The recovery from both diseases is usually accompanied by immunity. There is, however, a difference in the immunity due to a disease caused by bacteria which are vegetable organisms, and the immunity caused by protozoa, which belong to the animal kingdom.

The bacteria which have caused a disease in any particular animal disappear from its system as soon as it has acquired immunity. And bacteria, subsequently injected, are completely destroyed. When blood of an animal immune against a bacterial disease is injected into other susceptible animals, no disease will result. Such is not the case, however, in diseases caused by the genus of the large piroplasma. The blood of an ox which has recovered from redwater, or of a horse which has recovered from biliary fever, remains infectious even after years when injected into susceptible animals of their respective species. This is also the case with the blood of dogs which have recovered from malignant jaundice. The piroplasma canis must be present in the blood of the immune dog under a form which is not yet known. I have demonstrated the form of the piroplasma bigeminum in the immune ox, and probably a similar organism will be found in dogs. The disease which is caused by the injection of blood of immune dogs into susceptible ones is in every respect identical with that due to pathogenic ticks. There is an incubation time of several days, after which the typical piroplasma canis appears in the blood. Nor is there any difference in the virulency. Susceptible dogs injected with immune blood die as rapidly as those contracting it in the natural way. So far I have not come across a dog which has been able to resist the disease after injection with immune blood. On the other hand, any dog which is immune against the disease must have acquired it by passing through the disease. This has been clearly proved by the fact that blood of old dogs injected into young ones produced the disease in every instance. Such is, undoubtedly, the rule in Pretoria, where biliary fever of dogs is prevalent.

The serum of animals which have recovered from a bacterial disease and which have been systematically hyper-immunised with increasing quantities of bacteria of the particular disease, acquires preventive properties against the same bacteria. This can be proved by injecting a susceptible animal, first with serum, and then with bacteria, when no disease will develop. It will be of interest to ascertain whether by hyper-immunising an immune dog with large quantities of virulent dog blood, the serum of such a dog also acquires immunising properties. It must be remembered that blood of immune dogs is virulent. For this purpose an old dog (No. VI.) was injected with blood of young dogs suffering from biliary fever, and at different intervals, from 18th June, 1903, to 28th August, 1903. The total quantity of blood injected amounted to 115 c.c. blood.

II.

Experiments with Serum of a Fortified Dog (No. VI.), and Blood of a Dog (No. V.), which had Recovered from Piroplasmosis.

(Dog No. V. was suffering from Biliary Fever, 26th June, 1903, to 1st July, 1903.)

Remark.—All dogs used in the experiments were reared on the premises and were used soon after they were weaned.

Experiment 1.—To note whether the blood of Dog V. is virulent. 5th November, 1903, Puppy 16, injected with 2 c.c. defibrinated blood of Dog V., subcutaneously.

Result.—On 14th November, 1903, piroplasma canis was found in the blood of Puppy 16, which showed a fever reaction on that date. Knowing from previous experience that this dog would die, an injection was made with 20 c.c. serum of Dog VI. in order to come to some conclusion concerning the curative effect of the serum. Notwithstanding the injection the piroplasmas

increased during the next day, while some of them showed quite an abnormal shape. The puppy died during the night of the 16th November, 1903.

Conclusion.—The blood of Dog V. proved to be virulent.

Experiment 2.—To note the effects of a simultaneous injection of serum and virus (def. blood). Puppy 13 was injected on 5th November, 1903, with 20 c.c. serum, Dog VI., and 2 c.c. def. blood of Dog V., simultaneously and subcutaneously.

Result.—No reaction took place.

Experiment 3.—To note the effect of the injection of virus 24 hours after the serum had been injected. Puppy 14, injected on 5th November, 1903, with 20 c.c. serum of Dog VI., and on 6th November, 1903, 24 hours later, with 2 c.c. def. blood, Dog V.

Result.—No reaction took place.

Experiment 4.—To note the effect of the injection of serum 24 hours after the virus had been injected. Puppy 15, injected on 5th November, 1903, with 2 c.c. def. blood, Dog V., and on 6th November, 1903, viz., 24 hours later, with 20 c.c. serum.

Result.—No reaction took place.

Experiment 5.—To note the effect of a mixture of serum and virus injected 24 hours after the mixing. Puppy 17, injected on 5th November, 1903, with a mixture of 20 c.c. serum, Dog VI., and 2 c.c. def. blood, Dog V., mixed 24 hours previously.

Result.—No reaction took place.

Conclusion.—Blood of Dog V. was virulent, and produced disease in the control animal (Experiment 1). The simultaneous injection with serum and virus prevented the disease. The injection of serum 24 hours before or after virus prevented the disease; the serum and virus mixture proved not to be virulent. The serum of Dog VI. had, therefore, highly preventive properties when injected in large quantities. The different puppies lived for about six weeks, when, owing to too close confinement in rabbit cages, they died. Post-mortem was made in every instance, and microscopical examination proved the absence of piroplasma.

III.

Experiment 6.—To note the effect of smaller mixtures of serum than used in the former experiments. Puppy 18, injected on 18th November, 1903, with a mixture of 2 c.c. serum, Dog VI., and 2 c.c. def. blood, Dog V.

Result.—No reaction.

Experiment 7.—Puppy 19, injected on 18th November, 1903, with a mixture of 5 c.c. serum, Dog VI., and 2 c.c. def. blood, Dog V.

Result.—No reaction.

Experiment 8.—Puppy 20, injected on 18th November, 1903, with a mixture of 10 c.c. serum and 2 c.c. def. blood, Dog V.

Result.—No reaction.

Experiment 9.—To note the effect of virus which has been mixed with serum for 24 hours and which has been separated by centrifugalising and washed with aq. physiol. Puppy 21, injected on 18th November, 1903, with sediment of a mixture of 2 c.c. def. blood, Dog V., and 18 c.c. serum, Dog VI.

Result.—No reaction.

Conclusion.—The serum of Dog VI. prevented the disease when mixed in equal quantities with virulent blood. The impregnation of virulent blood with serum was sufficient to render it harmless.

Experiment 10.—To note the effect of simultaneous injection of virus and serum on animals of different weight, calculated on the base of 1 c.c. serum to 500 grammes live weight. Puppy 22 was used as control experiment, and was injected on 4th December, 1903, with 2 c.c. def. blood, Dog V. It contracted the disease and died of typical piroplasmosis on 16th December, 1903.

Experiment 11.—Puppy 23.—On 4th December, 1903, injected simultaneously with 32 c.c. serum, Dog VI., and 2 c.c. def. blood, Dog V.

Result.—Puppy 23 died on 11th December, 1903, or seven days after injection, too early to have contracted the disease from the injection. It must have contracted piroplasmosis by tick infection.

Experiment 12.—Puppy 24.—On 4th December, 1903, injected with 37 c.c. serum, Dog VI., and 2 c.c. def. blood, Dog V.

Result.—No reaction.

Experiment 13.—Puppy 25.—On 4th December, 1903, injected with 13 c.c. serum and 2 c.c. def. blood, Dog V.

Result.—No reaction.

Conclusion.—The serum had preventive action when injected at the rate of 1 c.c. to 500 grammes live weight. The Puppies 18, 19, 20, 21, 24 and 25, which survived the experiments, were tested on 28th December, 1903, with 5 c.c. def. blood of Dog V. They all contracted piroplasmosis and died with the exception of two (20 and 25), viz., Nos. 19 and 20 on 12th January, 1904, and Nos. 18 and 24 on 15th January, 1904. Thus the injection of virus and serum did not produce any immunity.

IV.

Experiments to Prove that the Blood of the Highly Hyper-immunised Dog VI. is Virulent, and that the Serum acts against this Virus in the same way as it did against the Virus of Dog V.

Experiment 14.—To prove that the blood of Dog VI. is virulent. Puppy 37, injected on 17th April, 1904, with 2 c.c. blood of Dog VI., freshly drawn and directly injected (not defibrinated).

Result.—Puppy 37 started to react six days after injection; a few piroplasms were seen on the seventh day. They increased daily, and the puppy died 14 days after inoculation.

Experiment 15.—Puppy 32, injected on 17th April, 1904, with 2 c.c. def. blood of Dog VI. The blood was defibrinated directly after withdrawal and injected with as little delay as possible.

Result.—Puppy 32 died on 2nd May, 1904, or 15 days after injection. There was but a slight rise of temperature. The usual symptom of jaundice was absent on *post-mortem*. The urine was red. There was also a splenic tumour, and piroplasma canis was fairly frequent.

Experiment 16.—Puppy 34, injected on 17th April, 1904, with 2 c.c. def. blood of Dog VI., which was kept for 48 hours in the ice box.

Result.—Puppy 34 started to react on 3rd May, 1904, on which date the piroplasma canis was present in the blood. It died on 5th May, 1904. Jaundice was absent. The urine was clear. Splenic tumour was present.

Conclusions.—The blood of Dog VI. proved to be virulent, injected either defibrinated or non-defibrinated, and, what is more remarkable, even after it had been standing for 48 hours.

Experiment 17.—To note the effect of serum of Dog VI. on blood of Dog VI., mixed and kept for 48 hours. Puppy 36.—On 17th April, 1904, injected with a 48 hours old mixture of 5 c.c. serum, Dog VI., and 2 c.c. def. blood of the same dog.

Result.—No reaction.

Experiment 18.—To note the effects of a simultaneous injection of serum, Dog VI., and of blood of Dog VI., inoculated at different places. Puppy 29.—On 17th April, 1904, injected with 5 c.c. serum, Dog VI., on one side of the body, and with 2 c.c. def. blood, Dog VI., on the other side.

Result.—Puppy 29 died on 3rd May, 1904, of piroplasmosis. Jaundice was absent. A big splenic tumour was present. The piroplasms were present in scanty numbers.

Experiment 19.—To note the effect of def. blood of Dog VI. 24 hours after the injection of serum of Dog VI. Puppy 30.—On 17th April, 1904, injected with 5 c.c. serum, Dog VI., and 24 hours later with 2 c.c. def. blood, Dog VI.

Result.—No reaction.

Conclusion.—With the exception of Experiment 18, the serum of Dog VI. proved to be preventive against its own virus. Puppy 29 probably died because the quantity of serum was not sufficient when simultaneously injected with virus.

V.

Experiments to Prove that the Serum of Dog VI. Contains an Immunising Substance which Resists Heating at 55° C.

Experiment 20.—To note the effect of a 48 hours old mixture of heated serum and def. virulent blood from which the serum was separated by the centrifugal machine and washing the red corpuscles with aq. physiol, 0·85 per cent. Puppy 35.—On 17th April, 1904, injected with 48 hours old mixture of 5 c.c. heated serum, Dog VI., and 2 c.c. washed and centrifuged blood of Dog VI.

Result.—No reaction.

Experiment 21.—To note the effects of a simultaneous injection of heated serum and washed virus. Puppy 33.—On 17th April, 1904, injected simultaneously with 5 c.c. heated serum, Dog VI., and 2 c.c. washed blood, Dog VI.

Result.—No reaction.

Experiment 22.—To note the effect of an injection of washed virus, 24 hours after heated serum has been injected. Puppy 28.—On 17th April, 1904, injected with 5 c.c. serum, Dog VI., and 24 hours later with 2 c.c. washed blood, Dog VI.

Result.—No reaction.

Conclusion.—The heated serum acted in every instance as a preventive. The conclusion is, therefore, that it contains a substance which is not destroyed by heating at 55° C.

Summary of Conclusions.

1. A dog which has recovered from an attack of piroplasmosis has acquired immunity against the disease.

2. The blood of an immune dog acts as virus when injected into a susceptible dog.

3. By hyper-immunising dogs with blood of dogs suffering from piroplasmosis, the serum acquires preventive properties.

4. The blood of a hyper-immunised dog is pathogenic when injected, either defibrinated or non-defibrinated, into susceptible dogs.

5. The serum of a hyper-immunised dog is preventive against the piroplasma canis of the same dog when injected into susceptible dogs.

6. The serum contains a preventive substance which is not destroyed at 55° C. It is, therefore, of a complex nature.

7. The mechanism of the production of a preventive serum in an immune dog seems to follow the same laws as exist in producing an anti-bacterial serum, with the essential difference, however, that the blood of a highly immunised dog remains infective.

Notes on the Immunity of the Piroplasmosis in Dog.

Experiment 1.—TEMPERATURES OF PUPPY 16.

Nov. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102⁸	102²	101⁸	99⁸	101²	100	102⁴	101²	102	101	102	104	103⁸	101⁶	103	101	101⁴	100⁴	104⁸	101⁶	105²

16		17	18	
101	100²	Died.		

Experiment 2.—TEMPERATURES OF PUPPY 13.

Nov. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102	101	101⁸	100⁸	102	102	102⁴	101²	101⁴	101²	103	99⁴	101²	101	104	101⁴	102²	101⁶	103⁸	102	102

16		17		18		19		20		21		22		23		24		25	
101⁴	103⁴	100⁴	102⁴	101	102⁴	102²	105	104	105⁶	102⁶	102	100⁶	100⁶	100	101	101	105²	101⁶	102⁶

Experiment 3.—TEMPERATURES OF PUPPY 14.

Nov. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	101^4	100^6	103	100^6	102^8	101^2	102	100^2	101^8	101	101^6	101^4	99^6	100^4	102^2	101^4	101^4	101^2	102	100^6	102

16		17		18		19		20		21		22	
100^2	101^6	99^8	101^4	99^6	101^6	99^2	101^8	100^2	102^4	101^8	102	100^6	102

Experiment 4.—TEMPERATURES OF DOG 15.

Nov. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	100	100^4	103	100^8	102^2	100^4	101^4	100^2	102^6	101^6	102	100^8	101^4	101^4	102^2	101^4	101^2	102	102^4	101^6	102^2

16		17		18		19		20		21		22	
100^6	101^4	100^8	100^8	100^6	101^4	100^8	101^8	100^4	102^2	101^6	101^8	101	102^2

Experiment 5.—TEMPERATURES OF PUPPY 17.

Nov. 6, 1903.		7		8		9		10		11		12		13		14		15		16	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102^4	101^4	102^8	101^6	102^6	100^8	103	100^2	102	100^2	100^6	99^6	101^4	100	101^0	100^6	102^4	100	101^4	100^5	102^4

17		18		19		20		21		22		23	
99^6	102^4	101^4	102	100^4	101^6	100^2	102^4	100^6	102^6	100^2	103	100	100^4

Experiment 6.—TEMPERATURES OF PUPPY 18.

Nov. 18, 1903.		19		20		21		22		23		24		25		26		27		28	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102	100^4	101^6	100^2	101^8	100^6	102^4	99^4	102^2	100	100	99^6	102^2	100^8	102^4	101	102	101	102	101^6	102^8

29		30		Dec. 1, 1903.		2		3		4		5	
100^4	102^2	100^8	103^2	100^6	102^4	100^8	102	100^6	102^6	99^4	102^4	100^6	102^2

Experiment 7—TEMPERATURES OF PUPPY 19.

Nov. 19, 1903.		20		21		22		23		24		25		26		27		28		29	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102^{8}	100^{4}	100^{8}	101	101	100^{6}	101^{6}	100^{4}	101^{8}	100	101^{6}	100^{2}	101^{8}	99^{8}	101^{8}	100^{6}	101^{8}	99^{8}	102	99^{4}	101^{4}

30		Dec. 1, 1903.		2		3		4		5	
100^{2}	100^{2}	101^{8}	102^{4}	100^{8}	101^{6}	101	102^{6}	101^{8}	102^{4}	100^{6}	101^{6}

Experiment 8.—TEMPERATURES OF PUPPY 2.

Nov. 19, 1903.		20		21		22		23		24		25		26		27		28		29	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102^{4}	102	101^{8}	101^{6}	101^{4}	100^{4}	102^{4}	101	101^{2}	100^{6}	101^{2}	101^{8}	103^{2}	101^{8}	102	101^{6}	102	101^{6}	103	101^{2}	102^{6}

30		Dec. 1, 1903.		2	
100^{6}	102^{4}	101^{8}	102^{4}	102^{2}	101^{6}

Experiment 9.—TEMPERATURES OF PUPPY 21.

Nov. 19, 1903.		20		21		22		23		24		25		26		27		28		29	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102^{4}	102	102	101^{2}	102^{6}	100^{8}	101	100^{8}	100^{5}	101	102^{4}	102^{4}	102^{4}	102^{2}	101^{8}	102	102^{4}	102^{2}	102^{4}	102	102^{6}

30		31		Dec. 1, 1903.		2		3		4		5		6	
101	103	101^{4}	102^{2}	101	101^{8}	101	102^{4}	101^{4}	102^{2}	102^{2}	102^{4}	101^{2}	102^{4}	102^{6}	102

Experiment 10.—TEMPERATURES OF PUPPY 22.

Dec. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102^{6}	101^{4}	103	101^{4}	102^{6}	100	101^{8}	99^{8}	102	100	103^{2}	100^{6}	102^{4}	102	103^{8}	101^{2}	101^{4}	101	102^{4}	99^{8}	102

16
Died.

Experiment 11.—TEMPERATURES OF PUPPY 23.

Dec. 5, 1903.		6		7		8		9		10		11	
M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102	100⁴	102⁴	101⁴	103²	101	105⁴	103	103	104	105⁴	103	Died.

Experiment 12.—TEMPERATURES OF PUPPY 24.

Dec. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	101⁴	100⁸	103	102²	101⁸	101	102	101	101⁸	100	102⁴	101	102⁴	101⁸	103²	103	103	101⁸	101²	102²	101⁶

16		17		18		19		20		21		22		23	
102	101⁸	101	101⁶	102	101⁶	101⁶	101²	101⁶	101⁸	101	102⁶	100⁶	101⁸	101	101²

Experiment 13.—TEMPERATURES OF PUPPY 25.

Dec. 5, 1903.		6		7		8		9		10		11		12		13		14		15	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	101²	99⁶	102²	101⁸	101⁴	100⁶	101⁴	100⁸	102	99⁸	101⁶	101	102	99⁸	100⁶	101	102²	100	102	99⁸	102⁶

16		17		18		19		20		21		22		23	
101	102²	100⁶	102⁶	100	101⁶	100⁸	101⁶	102	102²	101²	102²	102⁶	101⁶	100⁸	102⁴

Experiment 14.—TEMPERATURES OF PUPPY 37.

April 24, 1904.		25		26		27		28		29		30		May 1, 1904.		2		3		4	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	102⁴	101⁸	102⁸	101⁴	101²	102⁶	101⁶	102	101	102⁶	102⁶	103	104	103⁸	104	104	104²	103	104	104	104⁸

5		6	
103⁸	103	103⁸	Died.

Experiment 15.—TEMPERATURES OF PUPPY 32.

April 17, 1904.		18		19		20		21		22		23		34		25		26		27	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	101^{6}	99^{8}	100^{8}	99^{6}	100^{4}	99^{4}	100^{4}	99^{2}	100^{2}	100^{2}	101^{2}	99^{6}	101^{8}	99^{6}	101	99^{6}	101^{6}	100^{2}	101^{6}	99	100

28		29		30		May 1, 1904.		
98^{4}	100	99^{2}	100^{4}	100	102^{4}	100	96	Died.

Experiment 16.—TEMPERATURES OF PUPPY 34.

April 17, 1904.		18		19		20		21		22		23		24		25		26		27	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	—	101	100^{6}	100^{6}	101^{6}	100^{4}	100^{8}	100^{4}	100^{6}	101^{2}	101^{4}	101^{6}	101^{2}	101^{4}	101^{4}	99^{6}	101^{4}	101	102	101^{2}	101^{8}

28		29		30		May 1, 1904.		2		3		4		5
100	101^{2}	100^{8}	101	100	101	98^{6}	101^{4}	100	101	102^{4}	104^{8}	103^{2}	102^{2}	Died.

Experiment 17.—TEMPERATURES OF PUPPY 36.

April 18, 1904,		19		20		21		22		23		24		25		26		27		28	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
—	101	101^4	101^6	100^6	101^6	100^6	101^4	101^4	100^6	101	102^2	101^8	101^6	100^8	102^2	100^4	102	98^6	101	98^2	101

29		30		May 1, 1904,		2		3	
100^2	101	99^8	101^4	100	102	99^8	101^8	100^4	102

Experiment 18.—TEMPERATURES OF PUPPY 29.

April 18, 1904.		19		20		21		22		23		24		25		26		27		28	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100^4	100^2	100^6	100^8	100	100^8	100	101^8	101^4	101^6	100^8	102	101^4	102	101^4	102^2	101^6	101	99^6	100	98^4	98^4

29		30		May 1, 1904,		2		3
98	99	97	102	97	100^8	99	98	Died.

Experiment 19.—TEMPERATURES OF PUPPY 30.

April 18, 1904.		19		20		21		22		23		24		25		26		27		28	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101	100^{6}	101^{4}	101^{4}	101^{2}	101^{2}	100	101^{4}	a101^{8}	101^{4}	101^{6}	102	101^{4}	101	100^{6}	101^{8}	100^{8}	101^{4}	101	101^{8}	101	100^{6}

29		30		May 1, 1904.		2		3	
98^{2}	101	101^{2}	101^{2}	101	101^{6}	101	101^{2}	100^{8}	101

Experiment 20.—TEMPERATURES OF PUPPY 35.

April 18, 1903.		19		20		21		22		23		24		25		26		27		28	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
102^{4}	101^{4}	101	101	102^{2}	101	102^{4}	102^{8}	101^{6}	101^{4}	100^{6}	102	101^{6}	100^{8}	100^{8}	101^{2}	100^{6}	100^{6}	101^{6}	101^{2}	100^{4}	101

29		30		May 1, 1904.		2		3	
99^{2}	101^{4}	100^{4}							

I.

Heartwater in Cattle.

The study of this disease was undertaken with the object of obtaining some reliable data about the symptoms and morbid lesions which would help the observer in the field in arriving at a definite diagnosis. The material used was also utilised for hyper-immunising purposes. These latter experiments are still incomplete, and I propose to conduct them on a much larger scale next year. The different observations already made on sick and dead cattle have given us some information about the pathology of heartwater. The name of the disease is derived from a characteristic symptom, viz., the enormous filling of the pericardial sac with liquid. This is considered by farmers as the distinguishing lesion of the disease. Heartwater in cattle has long since been identified by the Transvaal farmer with a disease of the same name found in sheep and goats. He was led to this conclusion by a similar morbid lesion found in the heart-bag. He also observed that the disease in cattle appears under similar conditions as that in sheep and goats. Whilst this observation was correct, as will appear later, the farmer considered only cases of pure heartwater in which the characteristic symptom was present. My investigations have, however, revealed the fact that what is considered the constant symptom, although frequently met with in sheep and goats, is in the majority of cases absent in cattle. Hence the paucity of cases reported as heartwater, which for the most part were probably termed gall-sickness and bosch-sickness. The name bosch-sickness is applied to a disease which frequently occurs in cattle brought from the high veld into the warm bush veld, and is commonly found under similar conditions as heartwater. I am also under the impression that the term "dronk gall-sickness" is likewise applied to heartwater, more especially to sick animals which show symptoms indicating a disturbance of the brain. On the other hand, I am also aware that the term heartwater is applied to diseases in cattle which have not died from this disease, but on the *post-mortem* of which an increased amount of liquid is found in the heart-bag. Indeed, East Coast Fever, where this morbid change is frequently observed, has often been and still is, mistaken for heartwater.

II.

Cause of the Disease.

There is no doubt that heartwater is due to a micro-organism which enters the blood stream of animals. This opinion is based on the fact that the disease is easily communicated by inoculation with the blood of sick or recently recovered animals into healthy, susceptible ones; from sheep to goats and cattle, and *vice versa*. Blood seems to contain the virus under all circumstances. The disease is easily transmitted by subcutaneous inoculation, and more easily by injection into the jugular vein. The liquid of the heart-bag fails to produce the disease in most cases. It failed in all my experiments. This indicates that the microbe has to be looked for in the blood, particularly in the red corpuscles. Up to the present, every investigator who has conscientiously worked on the subject has failed to demonstrate the organism, either by the microscope or by cultures. These facts indicate a remarkable similarity between heartwater and horse-sickness. As far as I am aware it has not yet been shown that the virus of heartwater passes through a Berkefield or Chamberland filter, as is the case in horse-sickness. On the contrary, experiments seem to indicate that

this does not take place. The virus of heartwater is easily destroyed ; the blood retains its virulency hardly more than 48 hours. Heartwater is one of the diseases which we cannot diagnose by the microscope. Indeed, the naked eye diagnosis will often be impossible, because the characteristic clinical symptoms are frequently wanting, sometimes difficult, and then only based on circumstantial evidence.

After the inoculation of blood from a sick animal into a healthy one, a certain period is required to elapse before any symptoms in the infected animal are observed. This time usually lasts from eight to ten days. In rare cases it may be as short as five days and as long as fifteen days. The disease itself runs from two to six days, and even more.

III.

Symptoms of Heartwater in Sheep and Goats.

The first sign is always a rise of temperature. This may be the only symptom, and the animal suddenly dies, although seemingly quite healthy. Usually, however, the fever keeps on rising, and may reach 106° F., 107° F., and even 108° F. The sheep or goat may then stop feeding, become prostrated, and die without any typical heartwater symptoms. But a good many cases sometimes show symptoms before death of a brain affection so pronounced that one is struck by the idea that the name of " brainwater " would better fit the disease than that by which it is now known. For instance, a constant movement of the lower jaw, as if chewing something, is noticed ; a constant movement of the tongue may be observed, extending and withdrawing it continually. In other cases the animal may be seen licking the ground. It continually turns around the post to which it may be tied, and all of a sudden drops to the ground. Fits set in, the neck being extended backwards and the legs kept in regular motion. These symptoms may pass to return ; then they indicate the appearance of death. Some of the animals bleat some time before they die, others pass away quietly.

The symptoms of the digestive organs may vary greatly. There is sometimes foam at the mouth, the throat may be slightly swollen, rumination may stop even before feeding. The bowels are usually normal, but in some cases the animal is slightly costive, and, again, diarrhœa may be present.

The symptoms may thus vary considerably, but I wish to lay particular stress on the symptoms caused by the diseased condition of the brain, as described above, because, as will be seen later, a striking resemblance occurs in cattle, whether they are artificially infected with the disease or whether they contract it naturally.

IV

Post-Mortem Lesions.

The principal lesion in heartwater of sheep and goats is the increase of liquid in the heart-bag, which condition gave the disease its common name. This liquid coagulates easily soon after the bag is opened, and is sometimes even found coagulated in *post-mortem* of some standing. The liquid is, as a rule, clear, but it may also be found bloodstained. It must be borne in mind here, that not every excess of liquid in the pericardial sac has to be interpreted as being due to the malady in question. Cachectical condition of the body, due to bad pasture, infections by internal parasites and other causes, may produce a similar

collection of liquid. Here, however, one has nearly always to do with a poor carcase, whereas the same symptom in a good-conditioned animal points to the specific disease.

The pericardium itself is usually thickened, and has a gelatinous character, both in aspect and structure. In other cases, both the liquid in the bag and the derangement in the pericard may be missing.

The endocard (inner lining) of the left heart cavity has usually a mottled appearance, caused by blood spots; sometimes the hæmorrhages are present in streaks and also in patches. The blood is generally well coagulated, the clot of normal consistency and colour. There is also liquid found in the pleural cavity, which behaves similar to the one found in the heart-bag. The lungs are often in a dropsical state, full of liquid in the inter-lobular tissues, which may be found distended, and there is then foam present in the bronchial tubes, and even in the windpipe.

The abdominal organs may be found in quite a healthy condition. The spleen is, in the majority of cases, slightly enlarged, and its pulpa is slightly softened. The liver may be found in a state of congestion, dark in colour, the gall-bladder distended by a viscid thickish green or yellow bile. The kidneys are sometimes also in a hyperæmic condition, which is indicated by the red colour of a section of that organ. The bladder may be full or empty, and the urine is always clear.

The intestinal track shows different variations in the different cases. The third stomach may be found dry or soft; the mucous membrane of the stomach and bowels may be quite normal; but sometimes lesions of a slight catarrh or even of a hæmorrhagic inflammation are present. These conditions are indicated by a redness of the mucous membrane, which may be general, in streaks, in patches, or in form of a stronger filling of the blood vessels, and forming then a distinct arborization or tree-like appearance. The whole extension of the intestines, part or parts of it, may be involved in this pathological change. The membrane is usually thickened in such parts, and even a gelatinous infiltration of the mesentery belonging to it may be present. I have even noticed, in exceptional cases, that the mucous membrane of the lower part of the small intestines had become necrotic, and that the cœcum and colon had an exudation of blood on the surface. This is certainly, however, a rare occurrence.

V.

Geographical Distribution of Heartwater.

Heartwater of sheep and goats is met with only in a certain part of the Transvaal, namely, in the bush veld. There, sheep and goat breeding of high bred animals is impossible, and only the common Kaffir goat and fat-tailed sheep are found in any number, probably because these breeds have, in the long run of time, become immune. Not all parts of the bush veld are, however, equally bad; the worst ones are certainly the lower lying, and, therefore, also warmer, countries. The season also has some influence, inasmuch as during the summer time the disease is in such parts more prevalent and virulent. Heartwater is a malady which seems to stick on the veld. The Boer farmers know that since ages, and, accordingly, when an outbreak occurred in their flocks whilst they were in the winter veld, a rapid change of field was made, when the disease would soon come to a standstill. They also know that heartwater is not contagious, and once a flock was out of a heartwater field, up again in the high veld and mixed with other sheep or goats, such animals would not take the disease.

VI.

Propagation of the Disease.

Lounsbury, of Capetown, has demonstrated that heartwater is a disease propagated by a certain species of tick, viz., the bont tick (Amblyomma Habræum). He showed that it is only this species of tick which acts as carrier of the infection. In a series of careful experiments he proved that the infection does not go through the egg, but is taken either as a larva or a nympha from a sick animal and communicated in the later stage, which is as a nympha or an adult. Lounsbury was good enough to send me heartwater-infected nymphæ, which I placed in my laboratory on healthy Angora goats. They all contracted typical heartwater, and served for my first experimental researches.

The bont tick is an inhabitant of the bush veld, as every careful observer must know. It does not thrive in the high veld, nor even in the middle veld. It has since ages been brought there by cattle returning from the bushveld The male tick may be noticed on such cattle for many weeks and months, but the female is not known to breed in the colder districts. In the neighbourhood of Pretoria we find the bont tick behind the Magaliesberg, whereas it is not found on this side of the range.

Heartwater is only found in the same parts of the country where the bont tick exists.

VII.

Heartwater Cases in Cattle.

The tenor of this article being of an inductive character, the enumeration of some carefully observed experimental cases of heartwater may illustrate points more clearly.

Case 1 (calf heifer, two years old).—An Africander heifer, about two years old, was injected on July 24th, 1903, with 10 c.c. blood, intrajugularly. The blood was taken from a goat, which contracted heartwater from bont tick infection. Sixteen days later the temperature began to rise; became normal again after six days, during which time there seemed nothing to be amiss with the animal. When the temperature, however, rose to 105·6° F., the heifer was bled, and the defibrinated blood was injected into a goat, a sheep, and a calf (about 18 months old). The goat contracted the disease, so did the calf; its history is as follows :—

Case 2.—On the ninth day after inoculation, this period represents the incubation time, the temperature rose and was high for four days, when the animal, which was feeding all the time, showed visible symptoms of sickness. On the morning of that day the animal had distinct attacks of fits, there was a constant chewing movement of the lower jaw, just as if chewing the cud, and at the same time there was a constant nodding of the head, accompanied by extension and withdrawal of the tongue. A foamy saliva collected on the lips and the edges of the mouth, and dropped in long strings. The back of the animal was arched, the neck and head stretched out, the hind legs placed under the abdomen. When the animal was forced to move it did so with apparently stiff legs turned outside, and the walk was staggering, and it kept on moving until it dropped. It would then rise again, but all of a sudden intermittent spasms took place all over the body. These spasms may be produced by touching the animal, by forcing it to move, and by frightening it. I noticed them occurring as soon as a fly settled on the eyelids. Finally the animal drops after most pitiful bellowing, struggling with its legs, with stretched-out head. It was then killed.

Post-mortem was made directly after death. The condition of the animal was good. The lungs were in a slight dropsical condition (œdema of the lungs). There was a *little liquid* in the heart-bag. The outside of the heart was diffusely studded with hæmorrhages ; the lining of the left ventricle showed blood spots. The liver was normal, so was the gall-bladder. The spleen was enlarged. Kidneys and urine were normal. The mucous membrane of the fourth stomach was thickened. The folds were swollen ; the mucous membrane of the duodenum and all through the lengths of the smaller intestines and the colon were congested.

The microscopical examination of the blood gave negative results.

Case 3.—Africander calf, about 18 months old, was injected with 20 c.c. undefibrinated blood of a goat which was suffering from heartwater. After an incubation time of six days the temperature began to rise, gradually ascending up to 106·8° F. on the evening of the eleventh day. The day after the temperature dropped, and at that time a very serious brain symptom developed. The calf turned constantly round the pole to which it was tied up. At intervals it dropped, rose again, and constantly pushed forward ; the head was extended straight out, constant chewing movements of the lower jaws were present, and saliva beaten to foam hung around the lips. The eyes were staring, the eyeballs were slightly protruding, and the blood vessels of the sklera were strongly congested. Finally the calf dropped, not to get up again ; fits set in all over the body ; the head was extended, and convulsions followed. Death set in about eight hours after the first described symptom was noticed.

Post-mortem.—The condition of the animal was pretty good ; rigor mortis was present. There was some liquid in the peritoneal cavity, and there were blood spots on the serosa of the intestines. The *heart-bag* contained about 500 *c.c.* of a slightly red tinged liquid. The blood was blackish, but well coagulated. There were black spots on the lining of the left ventricle. The lungs were œdematous. The liver was of normal appearance. The gall-bladder was contracted, and contained brown bile. The kidneys were strongly reddened in section. The spleen was slightly enlarged. The mucous membrane of the fourth stomach was in a state of strong congestion, especially the fundus and pyloric parts ; there were blood spots on different places. The mucous of the duodenum was swollen and bile-stained. The mucosa of the smaller intestines had, in the first parts, a slate colour mixed with red patches and arborization of injected blood vessels. The membrane is slightly thickened. Towards the end of the mucous membrane of this particular intestine was still more swollen, the congestion more strongly pronounced, and the hæmorrhagic patches more numerous. The coecum and colon were strongly congested. There was a distinct increase of liquid in the chambers of the brain and under the duramater. Increased liquid was also found in the spinal cord. Microscopical examination gave negative results.

Case 4.—Africander calf, about two years old, was injected with 20 c.c. undefibrinated blood into the jugular vein of a goat which was suffering from heartwater. After an incubation time of 11 days the rise of temperature began ; the fever lasted five days. On the last day but one the calf was noticed to be very ill. It stood with a drooping head, taking no notice of what was happening around him. On the morning of the last day the animal was lying with extended head. There was slight diarrhœa present. *Post-mortem* was made about on hour after death. The condition of the cadaver was moderate. Rigor mortis was present. The blood was well coagulated and of normal colour, so was also the beef. There was some *yellow liquid* (*about* 500 *c.c.*) in the pleural cavity. The lungs were strongly œdematous, the interlobular spaces distended. The

heart-bag was filled with yellow liquid and enormously distended. The liquid coagulated rapidly after blood was added to it. The heart-bag was infiltrated with the same liquid, and had a gelatinous structure. There were blood spots on the lining of the left ventricle. The liver was congested; it had rather a reddish, glossy appearance. The gall-bladder was half filled with normal bile. The spleen was normal, the kidneys were hyperæmic, and the urine was normal. The lymphatic glands of the mesentery were slightly enlarged. The fundus of the fourth stomach was deep red, with numerous blood spots. The mucous membrane was thickened, the folds standing prominently out. There was a slight catarrhalic condition of the duodenum. In the lower parts of the smaller intestines the hyperæmic condition very pronounced, and numerous punctiform blood spots were present. In the cœcum, colon, and rectum was the congestion even more intense, and long red streaks were very marked. The whole picture called to mind similar conditions in rinderpest, with the difference that the peyer's patches were not attacked. The brain was slightly hyperanæmic, the dura mater was slightly infiltrated with liquid; there was also some liquid under the dura mater.

Case 5 (Calf 220).—A calf about 18 months old was injected on January 3rd, 1904, with blood of Sheep 96, suffering at that time from heartwater. The original strain of heartwater virus was, in this case, obtained from a sheep which died in Nelspruit from the disease. The *post-mortem* lesions of Sheep 96 were shortly, as follows:—Yellow liquid in heart-bag and pleural cavirt. Lungs slightly œdematous. Hæmorrhages on endocard of the left ventricle. Spleen enlarged and pulpy. Liver congested. Kidneys hyperæmic and capsula infiltrated. Mucous membrane of the fourth stomach slightly reddened. Parts of the small intestines were swollen and strongly congested. This is the usual *post-mortem* record of heartwater in sheep.

The disease ran in Calf 220 in incubation time of 10 days, then the temperature began to rise, and reached, three days later, 106·4° F. The symptoms of a serious trouble, however, set in only on the morning of the last (fourth) day, when irregular muscular spasms were noticed, which preceded usually a shock-like shaking of the whole body. Towards mid-day the calf lay down. It kept the head bent to the right side, there was a constant chewing movement of the lower jaw, accompanied by extension and withdrawal of the tongue and curling of the upper lip; at the same time a noise like sucking air was heard. Finally the head was resting on the right leg, which the animal began to bite furiously, so that a wound was produced. The calf died in the evening at six o'clock.

Post-mortem was made immediately after death. The rigor mortis was not yet present. The blood was not yet coagulated. The beef had a normal colour. The lungs were normal. There was *no liquid in the heart-bag.* The endocard of both ventricles was dotted with blood spots and streaks. The liver was normal, and so were the kidneys. The contents of the third stomach were soft. The folds of the fourth stomach were slightly swollen. The mucous membrane of the small intestines, as well as of the colon and cœcum, was, in its whole extension, thickened. It was reddened in patches in the smaller intestines, and in streaks in the thick intestines. From the outside the intestines had a swollen appearance. When the head was cut off, in the region of the first vertebra, a rather large quantity of clear liquid escaped from the spinal canal. The brain also had an appearance as if infiltrated with liquid.

Case 6 (Calf 216, two years old).—This animal was injected on January 12th, 1904, with 20 c.c. blood into the jugular vein of Sheep 97. The strain of the virus was also originally obtained from the sheep dying of heartwater in Nelspruit. Sheep 97 had served as a control in the foregoing experiment,

and had been infected with blood of Sheep 96, as was Calf 220. Sheep 97 died on January 13th, 1904, and had the typical heartwater lesions, viz., heart-bag full of liquid hæmorrhages on endocard, slightly enlarged spleen, congested kidneys, and an enlarged bile-bladder.

The incubation time in Calf 216 lasted nine days, then the temperature rose, and the disease lasted twelve days, when the animal had to be killed, it having been in a comatose state for two days. Shortly before the animal lay down, and during its coma, it had convulsive spasms. These convulsions were renewed whenever the animal was touched. There was a peculiar blinking movement of the eyelids.

The *post-mortem* was made soon after death, before rigor mortis had set in. The beef had a normal colour. There was *no liquid* in the peritoneal or pleural cavity, and but little in the heart-bag. The lungs were retracted, pale, and there was some foam in the windpipe. There were subendocardial hæmorrhages in the left ventricle. The liver was normal, the bile green and liquid. The spleen and the kidneys were normal. The mucous membrane of the stomach and of the intestines was slightly swollen, without much reddening. The brain was strongly hyperæmic.

Case 7 (Calf 236).—This animal was injected on December 27th, 1903, with 20 c.c. blood of Sheep 92, intrajugularly. Sheep 92 had been injected with a virus obtained from a sick sheep in Nelspruit, and died on December 26th, 1903. It had typical heartwater lesions, viz., there was about a litre of yellow liquid in the chest; the heart-bag was filled with the same liquid, and the pericard was infiltrated. The endocard of the left ventricle was dotted with blood spots over the whole surface.

Calf 236 showed a rise of temperature after an incubation time of about seven days. The reaction was somewhat irregular, but attained over 106° F. the last few days.

On January 11th, or fourteen days after the injection, this animal died suddenly in the early morning, without having shown any outward symptoms the day before. The *post-mortem* was made before rigor mortis had set in. The beef had a normal colour. There was about two litres of liquid in the pleural cavity, also an increased amount in the heart-bag. The lungs were very œdematous, the interlobular tissue much enlarged. The mediastinal tissue and the tissue along the aorta was infiltrated with copious hæmorrhages. There were extended petechiæ in the left ventricle of the heart. The liver, spleen, and kidneys were normal. The mucous membrane of the fourth stomach was slightly reddened, the folds were very much thickened; the mucous lining of the duodenum and also that of the coecum was thickened, and coagulated blood stuck on the surface. There were very small ulcers on the mucous membrane of the small intestines, which were but slightly swollen. The rectum had long red streaks on the mucous membrane, which was thickened.

VIII.

Analyses of the Experimental Cases.

All seven animals, which were injected with heartwater virus, contracted the disease and six died. The injected cattle were imported, either from the Cape Colony or Texas. The experiments show the high susceptibility of foreign cattle. The incubation time averaged from nine to ten days and the disease about six days. The shortest period of disease lasted four days, the longest twelve days.

The only animal which resisted the disease, had merely a high temperature. It showed no outward symptoms, and, in fact, this case might have escaped observation. Notwithstanding which, its blood proved fatal to a calf and goat.

All the animals which died showed symptoms resembling those found in sheep and goats. Irritation of the nervous system was present in four cases. These symptoms are especially characteristic and may be considered typical.

The striking lesion from which the name is taken, was absent in three cases and distinct in two. The most marked lesions were a gastritis and an enteritis, which in one case, were slightly noticeable, but were plain in the rest. The hæmorrhagic nature of the inflammation was sometimes so pronounced that local irritation set up by a strong poison might readily be surmised to be the cause of it. There was also some likeness to the lesions of rinderpest and East Coast Fever.

Heartwater must be considered as a septicæmia, due to non-visible and non-cultivable micro-organism. This being so, it is possible to understand the variations in the symptoms of the different animals.

The experimental cases prove that heartwater in goats and sheep is inoculable into cattle and, again, from cattle into goats and sheep. This demonstrates the unicity of the disease, although the *post-mortem* lesions vary somewhat. In sheep and goats which contract the disease naturally, viz., by ticks, the liquid in the heart-bag is an almost constant sign. This is not always the case in the same animals when they have contracted the disease by inoculation with virulent blood taken from cattle. Here, likewise, the lesions in the intestines may be the only significant symptoms. In making several inoculation passages from sheep to sheep, the typical heartwater lesions will reappear. In other cases, the typical disease was apparent even after the first inoculation. It should be stated that the Cape experts have produced the disease in cattle with pathogenic bont tick nymphæ, which had been feeding both on sick goats and on sick cattle. They have also made similar observations in regard to *post-mortem* lesions as already mentioned.

IX.

Spontaneous Cases of Heartwater.

The following cases were observed amongst a mob of cattle belonging to the rinderpest station, Pretoria. They had been imported from the northern districts of the Cape Colony, where redwater and heartwater are unknown. During the course of immunisation against rinderpest, they also became immune against ordinary redwater, this being a natural result of injection of blood from animals which are immune against this disease. The cattle were first kept on the premises of the laboratory and were then removed to a farm behind the Magaliesberg, where several outbreaks of a disease occurred, which, when compared with the experimental cases of heartwater, had to be declared identical with them.

1. One Cape ox, 2½ years old, was hyper-immunised against rinderpest, arrived three days ago from Wonderboom, where he had been for some weeks. He was noticed to be ill for about two days when he got suddenly bad and could not get up any more. He showed spasms of the head, of the neck, and the front legs. Diarrhœa was present. He died during the night.

Post-mortem was made early in the morning. Condition was good. Rigor mortis was present. The beef had a normal colour; the blood was partially not coagulated. There was a yellow liquid in the peritoneal cavity. The lungs were slighly œdematous, so were the mediastinal lymphatic glands. The heart-bag contained a considerable amount of yellow liquid, which had rather a brownish tinge. The outside of the heart had a brick-red colour, and big red, blackish hæmorrhagic spots were on the left endocard. The liver was

enlarged; contained much blood. The walls of the bile-bladder were thickened, and there was but little bile. The spleen was slightly congested, its pulpa softened. Both kidneys were deep red; the urine was normal.

The mucous membrane of the fourth stomach was much swollen, particularly in the folds, which were as thick as a finger. The pyloric part was reddened. The mucosa of the duodenum was bile-stained; that of the small intestines was reddened, the blood vessels strongly injected. The contents of the bowels were of a brownish red colour. The lower part of the intestines was covered with mucous; there was a slate-grey discolouration present. Peyer's patches were normal. The mucous membrane of the cœcum and colon was thickened; there were grey and red patches; there were also red streaks in the rectum.

The brain was strongly congested. Microscopical examination of the blood from the heart, liver and spleen proved the absence of piroplasmas of any description.

2. 8th June, 1903.—Cape ox, about 3½ years old. Same history as previous case. Was noticed to be ill since about a day, showing nervous symptoms, such as spasms of the head, extension and withdrawal of the tongue, biting the ground, staring look; when he finally dropped, he kept moving his legs in a trotting action.

Post-mortem was made about 1½ hours after death. The condition was fair. Rigor mortis was not yet present; the blood was not yet coagulated. The beef had a normal colour. There was some liquid in the peritoneal cavity. The lungs were normal. The heart-bag was empty; the left ventricle showed blood spots. The liver was congested. The gall-bladder was thickened; the bile was viscid. The spleen was normal, so were the kidneys and the urine.

The serous membranes of the bowels were infiltrated with yellow liquid. The third stomach was somewhat dry. The fourth stomach had a swollen mucous membrane; the folds were very thick, the mucous membrane was uniformly reddened, and there were numerous hæmorrhagic spots. The mucous lining of all the small intestines was generally congested and studded with fine blood spots. Colon and cœcum were swollen, and on the mucosa were long hæmorrhagic streaks which extended into the rectum. The anus was deep red. Microscopical examination proved the absence of any blood parasites.

3. On December 12th, 1903, a Cape Ox, No. 448, which had been hyper-immunised against rinderpest and was also immune against ordinary redwater, was noticed to be ill. With an elevated temperature, the illness kept on for some days, but the animal finally rallied and recovered. The examination of its blood revealed certain endoglobular parasites, which I have described in my article on "The Piroplasma of the Immune Ox," as the form of piroplasma bigeminum which, under certain conditions, may increase in the blood. The disease in the ox was declared to be "gall-sickness," a name, which, as I have repeatedly pointed out, is very vague, and is useful as a convenient cloak with which to conceal the ignorance of cattle diseases in general.

X.

Experiments to prove that the Spontaneous Cases were Heartwater.

In order to arrive at a definite conclusion as to what disease Ox 448 was suffering from, an inoculation was made with blood into a Sheep, No. 102 and a two year old Cape Ox, No. 233, each animal receiving 20 c.c. defibrinated blood into the jugular vein on December 12th, 1903. The following is the result of the inoculation:—

(*a*) Sheep 102.—On the eighth day after inoculation, a rise of temperature began, which on the same day reached 107°. The temperature ocillated during the following days, but reached a second time, eight days later, 107·2° F. The animal recovered without showing any other symptom, save losing condition.

(*b*) Ox 233.—This ox collapsed on the ninth day after inoculation and died suddenly, having previously shown an irregular temperature, which dropped to sub-normal the day before death.

The *post-mortem* was made soon after death, the blood being still uncoagulated. The beef and serous membranes of the different cavities had a normal colour. The lungs were partially retracted, the interglobular tissue was emphysematous. There was but little liquid in the pericardial bag, whose base was infiltrated with the same liquid, and so also was the mediastinum. There was no liquid in the pleural cavity. The heart was flabby; a few petechiæ were on the surface of the heart and one on the endocard. The colour and consistency of the liver was normal, with slight hyperæmia. The gall-bladder was half filled with liquid yellow bile. The spleen was enlarged to about three times its normal size; its pulpa was hyperæmic and the Malpighian bodies were scarcely recognisable. The capsula of the kidney was slightly œdematous, likewise the calix of the kidney; the organ itself was of a normal colour and of normal consistency. The bladder was filled with yellow urine.

The mucosa of the fourth stomach was slightly swollen without any discolouration. The mucous lining of the duodenum was still more swollen and bile-stained; so also was the mucosa of the small intestines, which was contracted into folds, covered with a viscid mucous and reddened in patches. The mucous membranes of the colon and of the cœcum were also swollen, but without any discolouration. The rectum was normal.

The blood of this ox, taken after his death, was injected into a two year old calf and an old Transvaal ox and a sheep. The two cattle did not show any reaction, they proved to be immune, but the Sheep, No. 103, which was injected at the same time, had a high fever reaction, the temperature rising as high as 106·4° F. However, it recovered.

In order to elucidate the problem of the disease in Ox 448, whose blood produced a high fever in Sheep 103, this latter animal was tapped during the reaction, and the blood injected into sheep and cattle at different dates.

(1.) 21st January, 1904.—Sheep 104, injected with 20 c.c. This animal showed a distinct but a typical fever reaction, lasting several days, without any outward symptoms. It recovered. It was tested on the 8th April with a strain of heartwater virus, taken from a sick sheep. It proved to be immune.

(2). 22nd January, 1904.—Cape Ox 230, about three years old, was injected with 20 c.c. undefibrinated blood, After an incubation time of nine days, the temperature attained 105° F., at which point it kept during the next three days, in order to drop sub-normal on the thirteenth day, when the ox was found to be dying. The symptoms of brain irritation were present. The animal was lying on one side, constantly biting the leg on which the head was laid. Profuse salivation was present. Shortly before death the spasms set in all over the body and the animal died bellowing.

The *post-mortem* was made about four hours after death. Rigor mortis was present. The condition was good. The blood had not yet completely coagulated. There was a slight dropsical condition of the lungs. The liquid in the heart-bag was increased. There were blood spots on the surface of the heart and on the endocard of the right ventricle; and a patchy hæmorrhagic

infiltration on the endocard of the left ventricle. The liver was hyperæmic, and the gall-bladder was filled with liquid bile. The spleen was slightly enlarged. The kidneys were hyperæmic; and the calix œdematous. The stomach and the intestines showed a swollen appearance outside. The folds of the fourth stomach were thickened. The mucus membrane was reddened. The mucous membranes of the duodenum of the whole length of the small intestines, of the colon and cœcum were uniformly swollen, with hyperæmic patches showing blood extravasations. In some parts it was slate coloured in patches and streaks. The contents of the larger intestines were watery.

(3). 1st February, 1904.—Sheep 102 was tapped again, and this time four three year old oxen, all imported from the Cape, were injected with 50 c.c. blood, directly drawn and not defibrinated. Out of the four animals, only one did not contract the disease (No. 229). The first animal died 22 days after the injection of virus, it showed an incubation time of 14 days and a fever reaction of seven days, the highest temperature reaching 105° F. This ox showed rapid loss of condition during the fever reaction. The day previous to death, serious symptoms were noticed. The ox refused to feed, and stopped chewing the cud. Attacks of fits were frequent, the animal falling suddenly down and the peculiar blinking movement of the eyelids was present.

The *post-mortem* was made directly after death, before the cadaver was cold. The condition was poor. The beef had a normal colour. There was no liquid in heart-bag, chest or peritoneal cavity. The heart was also normal. The liver was hyperæmic. The gall-bladder was large and full of green bile. The spleen was slightly enlarged. The kidneys were hyperæmic; the tissues of the hilus and calix were infiltrated with yellow liquid. The mucous membrane of the stomach was reddened, the folds were swollen. The mucous membrane of the small intestines was swollen throughout the whole length and folded; it was strongly reddened, and on some parts were distinct hæmorrhages. The mucous membrane of the cœcum and colon was less swollen, but red streaks were noticed. The serous membrane of the intestines were infiltrated with liquid. In cutting off the head, a fair amount of liquid escaped. There was nothing peculiar noticed in the brain.

The second animal (Ox 228), also a three year old Cape Ox, died 23 days after inoculation. It had an incubation time of 15 days, an irregular fever reaction, which lasted eight days; the temperature rose on one evening up to 106° F. This animal showed, during his last days, loss of condition and would not feed properly; during the latter days, increased salivation was seen, foam hanging round the mouth; but there were never any alarming symptoms present. The ox died during the night.

Post-mortem was made in the morning. The cadaver was in a rather poor condition. Rigor mortis was present. There was a considerable amount of yellow liquid in the chest. The lungs were dropsical; in cutting into the lung tissue, some liqiud was noticed to run out freely. The heart-bag was bursting full of yellow liquid. The pericard was thickened with liqiud. Both heart ventricles were filled with well coagulated blood. The surface of the heart had a patchy appearance through confluent petechiæ; the same was noticed on the endocard. The spleen was slightly enlarged and soft. The liver was slightly icteric. The gall-bladder contracted, and there was but little normal bile. The vicinity of the kidneys was infiltrated with gelatinous looking liquid; the organ itself was hyperæmic; the calix œdematous and dotted with blood spots. The urine-bladder was filled with yellow urine. The mesenteric tissue of the intestines was strongly infiltrated with liquid. The contents of the third stomach were soft. The mucous membrane of the fourth

stomach was very much thickened, over half an inch, and presented a uniform hæmorrhagic surface. The duodenum and also the ileum and jejunum were swollen on their whole extension; the mucous membrane of the lower ilium was, particularly, much thickened. The whole surface of the mucous lining was reddened with numerous hæmorrhages. Peyer's patches were normal. The mucosa of the cœcum and colon were also swollen and reddened in streaks and patches.

The third Ox (No. 227) died 32 days after inoculation. After an incubation time of 14 days, an irregular fever reaction was observed, the temperature rising on one occasion, up to 106° F. The fever reaction kept on oscillating, and usually dropped in the morning to normal and sometimes to sub-normal. The animal lost condition from day to day, its appetite was wanting; there were never, however, any distinct symptioms.

The *post mortem* was made soon after death, before rigor mortis had set in. The cadaver was in a poor condition. There were gelatinous infiltrations of the intermuscular tissue. All serous membranes were infiltrated with liquid, as for instance, the pericard, the mesentery, and the intestines. The base of the heart and the horizontal and vertical grooves presented a gelatinous looking aspect. The lungs were normal. The spleen was also normal. The liver was rather small. The bile was normal. The surrounding tissue of the kidneys and the pelvis was also infiltrated. The third stomach was dry and hard. On the mucous membranes of the fourth stomach were relics of old hæmorrhages. The mucosa of the small intestines was swollen in its whole length, and small ulcers were present. The mucosa of the cœcum and colon was thickened and reddened. The mucous lining of the swollen rectum was slate-coloured and hard dung was present.

(4). On 3rd February, 1904, viz., during the last days of the fever reaction of Sheep 102, a final bleeding was made, and a young Goat, No. 106, which was born on the premises, was injected with 10 c.c. non-defibrinated blood. After an incubation time of seven days this animal contracted fever, which reached 107·8° F. The kid died on the 12th day after inoculation. The day previous to his death it was constantly lying, being paralyzed in the hind legs and, therefore, unable to rise. It showed peculiar spasms in the muscle of the head, and when touched, started a pitiful moaning.

The *post-mortem* revealed a healthy appearance of the organs of the chest. It was particularly noticed that the heart-bag was empty. The mucous membrane of the fourth stomach was swollen and reddened, and so also, intensely, was the mucous membrane of the small intestines. The cœcum and colon were less reddened.

Not satisfied with the *post-mortem* of Goat 106, which died on the 15th February, a sheep was injected on 23rd February, 1904, with 10 c.c. blood of Ox 227, which, at that time, was undergoing reaction. This sheep showed, after an incubation time of nine days, a reaction; the fever gradually rising and reaching on the day before death, 106·8° F. It died almost suddenly

The *post-mortem* was made soon after death. The blood was not yet coagulated and the cadaver was in good condition. There was an *increase of liquid* in the heart-bag. There were numerous hæmorrhages in the left ventricle and also, but less so, in the right. The lungs were slightly œdematous. The liver was hyperæmic. The bile normal. The kidneys were hyperæmic. The spleen was slightly enlarged; its pulpa soft. The mucous membrane of the fourth stomach was congested; the whole length of the small intestines was

in a state of cattarrh, with red patches, more or less regularly distributed. The mucous membrane of the cœcum and colon was intensely reddened and uniformly marked throughout its whole length. The blood of all the sick animals was microscopically examined with negative results. A further experiment was made with blood of Ox 448, which was ten days old, and a calf and a sheep were inoculated, but both remained healthy, indicating that the blood had lost its virulency.

II.

Conclusions.

1. Heartwater of sheep and goats is inoculable into cattle and *vice versa.*

2. The transmission takes place under natural conditions by means of infected nymphal or adult bont ticks.

3. The disease varies in its symptoms in cattle, sheep and goats.

4. The lesion, which is believed to have a certain differential diagnostic value is not always present.

5. The common morbid symptoms of heartwater in cattle are the lesions found in the digestive tract.

6. The injection of virulent blood from cattle into sheep and goats is not always fatal.

7. Imported cattle are highly susceptible to heartwater.

TEMPERATURES OF OX 228.

February 1, 1904.		2		3		4		5		6		7		8		9		10		11	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100⁶	105⁶	100²	102⁴	99⁴	102⁶	99⁴	102⁴	99⁸	103²	100²	102⁴	100⁴	103⁶	100⁶	103⁸	100	99⁸	99⁶	103⁴	99⁸	103

12		13		14		15		16		17		18		19		20		21		22		23	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
98⁸	102⁶	99²	103	98⁴	102⁶	98⁶	105⁴	101	105⁴	99⁴	102⁶	101⁴	104⁶	102⁶	105⁶	104⁸	106	103⁴	102⁸	100	102⁴	Dead.	

TEMPERATURES OF OX 227.

February 1, 1904.		2		3		4		5		6		7		8		9		10		11	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100	103	99^{8}	102^{4}	99^{4}	103^{4}	98^{4}	101^{4}	99	102^{8}	99^{4}	102^{8}	100	103	99	103^{8}	98^{6}	100^{4}	97^{6}	103	100^{2}	102^{8}

12		13		14		15		16		17		18		19		20		21		22	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
99^{4}	102^{2}	99^{4}	102^{4}	97	102^{6}	98^{8}	103^{8}	100^{2}	104^{4}	98^{6}	103	101^{4}	104^{4}	101	104^{8}	102^{4}	106	102^{8}	104	101	103^{2}

23		24		25		26		27		28		29		March 1, 1904.		2		3		4	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101	103^{6}	98	104^{4}	98^{4}	—	—	104^{2}	99^{8}	104	101^{6}	104^{8}	101^{6}	104^{2}	101^{6}	104^{4}	100^{2}	103	97^{8}	—	100	Died.

Experiment 6.—TEMPERATURES OF CALF 216.

January 12, 1904.		13		14		15		16		17		18		19		20		21		22	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100⁶	102⁴	100⁴	101⁶	99	100⁸	99⁴	102⁶	100	102⁴	100	102⁶	99⁸	102⁴	99²	100⁶	100	98⁴	98⁸	102⁶	100	103²

23		24		25		26		27		28		29		30		31		February 1, 1904.		2	
102	105⁴	102²	102⁸	102⁴	103⁴	101⁶	105	101⁶	104²	103⁶	105²	103⁴	105²	105	105⁴	100⁶	101²	98	102²	100	

Experiment 7.—TEMPERATURES OF CALF 236.

December 22. 1903.		23		24		25		26		27		28		29		30		31		January 1, 1904.	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
100⁸	102⁸	100⁶	102⁴	99⁶	101	100⁴	101⁸	100	101⁶	101	102²	101²	104⁶	101⁴	103⁴	101	102⁶	101	104	101⁶	100⁴

2		3		4		5		6		7		8		9		10		11		
101	102⁶	101	103⁸	102	104⁴	102	105	101⁴	103⁴	100⁸	106⁴	104	105⁴	105⁶	106²	104	102⁴	98⁶	Died.	

10 C. OX 233.

January 12, 1904.		13		14		15		16		17		18		19		20		21	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
101²	102⁸	100⁶	102⁴	100²	101⁶	100⁶	104²	100⁴	104	100	104²	100⁶	103⁶	98	98	95⁶	100	95⁴	Died.

2.—TEMPERATURES OF OX 230.

December 21, 1903.		22		23		24		25		26		27		28		29		30		31	
M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E	M	E
98	102⁶	97⁸	100	98	104²	99⁸	102²	99⁶	101⁸	98⁶	102⁴	98⁴	101⁸	99⁶	102⁶	98⁸	102⁶	99²	102²	99⁸	101⁸

January 1, 1904.		2		3		4		5		6		7		8		9		10		11	
100²	102⁶	100²	101⁴	99²	102²	99⁴	102²	99²	102⁴	99	103⁴	99⁶	103⁴	98⁶	102⁴	99	100⁶	100⁸	103⁴	101	105

12		13		14		15		16	
102²	104⁶	103	104⁸	98⁴					

Horse-Sickness.—Introduction.

Results from Former Experiments.

1. The blood of an animal which is suffering from any form of horse-sickness (dunkop, dikkop, or blue tongue) is virulent when injected into other susceptible animals.

2. Susceptible animals are the horse, the ass, and their bastards. The ass is the least susceptible; only a small percentage contract the disease, and a still smaller number succumb to the inoculation. The mule is as susceptible as the horse, but there is a larger percentage of recoveries.

3. Horse-sickness is not a contagious disease, but easily inoculable.

4. The micro-organism of the disease is not known. It passes through the finest Chamberland Filter, and is, therefore, ultra-visible. Hitherto it has not been cultivated on any artificial media.

5. The different clinical forms of horse-sickness are due to the same virus. The blood of an animal suffering from dunkop may produce dikkop, and *vice versa*.

6. The blood is virulent, so is the serum of the blood, the exudates, extracts with physiol. water of brain, lung, heart, liver, spleen, kidneys, and muscle. The urine is not virulent.

7. The minimal quantity of blood required to produce the disease may be 0·001 c.c. This quantity is not, under all circumstances, virulent. A dose of 1 c.c. of blood is virulent.

8. Serum of a sick horse does not always produce the disease when subcutaneously injected in the quantity of 1 c.c.

9. The disease may be produced by injection of virus into the skin, under the skin, into the lungs, into the pleural cavity, into the peritoneal cavity, into the trachea, into the jugular vein, into the brain, and through the mouth.

10. The surest methods of producing the disease are the intrajugular and the subcutaneous injections. Injection of virus into the trachea is not always followed by the disease, and as much as 5 c.c. of virulent serum was supported. Horse-sickness virus, given through the mouth, will only produce the disease when large quantities are used. Quantities of 100 c.c. never caused the disease. Quantities above 150 c.c. always caused the disease.

11. The blood of an animal suffering from horse-sickness is virulent as soon as the reaction has begun. It is sometimes virulent before this reaction, likewise a few days after it has passed.

12. Blood from a sick animal retains its virulency for at least two years. During this time it may become putrid, but it will still produce the disease.

13. Virus which has been dried in the incubator, or at the temperature of the room for twelve hours, loses its virulency.

14. The temperature of the ice-box does in no way influence the virulency of the horse-sickness virus.

15. Heating at 45 C. for five days does not alter the virulency of horse-sickness blood.

16. It is possible to increase the virulency of virus by passing it through a series of susceptible animals.

17. The average incubation time lasts seven days. The average time of horse-sickness reaction lasts from five to six days. The shortest incubation time is noticed after injection of virus into the jugular vein. The length of the incubation time depends somewhat on the quantity of virus. In cases of recovery the average reaction lasts ten days. The majority of recoveries (56·5 per cent.) showed the symptoms of dikkop.

18. Animals which have recovered from horse-sickness acquire immunity. This immunity is not, however, complete for every animal. It may break down at any time (relapses, aanmaning), but such relapses end fatally only in the minority of cases. The experiments undertaken to ascertain the percentage of relapses of immune horses exposed in a notoriously unhealthy locality, viz., Elandshoek, resulted as follows :—Out of 18 "salted horses," whose immunity had been tested, 6 contracted horse-sickness (viz., 33·3 per cent), of which 2 died (11·1 per cent.).

19. The relapses occurred in two animals which had recovered from Dikkop, and in four animals which had recovered from dunkop.

The Serum Treatment Applied to Horse-Sickness.

I.—*To note whether the serum of an animal (mule), which has proved to be immune against horse-sickness, has protective properties when injected in large quantities into a susceptible horse simultaneously with virus*

Remark.—Mule 112 had been injected on 24-6-02 with 150 c.c. virus, Horse 124, subcutaneously, and did not react. It was accordingly immune.

Experiment No. 1.—16-8-02.—Horse 136, injected with 1,000 c.c. serum of Mule 112, subcutaneously, and 1 c.c. virus of Horse 102, subcutaneously, into the tail.

Result.—Horse 136 died on 29-8-02, viz., 13 days after injection, from horse-sickness.

Conclusion.—One litre of serum from an immune—not hyper-immunised—animal did not protect against 1 c.c. of virus, both injected subcutaneously.

II.—*To note whether the serum of an immune horse—which has been hyper-immunised with virulent blood—protects a susceptible animal when injected in large quantities with virus, simultaneously.*

Remark.—The Horse S.H. 1 had been hyper-immunised to the extent of 2,020 c.c. virulent blood from various horses.

Experiment No. 2.—Horse 138.—10-9-02, injected 1000 c.c. serum, S.H. 1, subcutaneously, and 1 c.c. virus of Horse 102, into the tail, simultaneously. A reaction resulted on 21-9-02, or 11 days after the injection. It lasted seven days. This animal was tapped during the reaction on 25-9-02, and 10 c.c. of the defibrinated blood was injected into a horse, No. 160. No reaction ensued in this horse, but later, when Horse 160 was retested with a virus—the virulency of which was established—it proved to be immune.

8-10-02.—Horse 138, reinjected with 1 c.c. virus, 102. No reaction.
24-10-02.—Horse 138, reinjected with 5 c.c. virus, No. 102. No reaction.
13-11-02.—Horse 138, reinjected with 50 c.c. virus, No. 102. No reaction.
1-12-02.—Horse 138, reinjected with 100 c.c. virus, No. 174. No reaction.

Result.—This horse was killed on 30-1-03, it having shown symptoms of farcy.

Conclusion.—The injection of 1,000 c.c. serum of a horse hyper-immunised to the extent of 2,020 c.c. virus did not protect the horse against the disease—produced by the simultaneous injection of virus—but modified the attack by which the animal acquired immunity.

Note.—The absolute proof as to whether Horse 138 had an attack of horse-sickness is lacking, the control animal, in which blood of the reaction was injected, having been immune. Nevertheless, the incubation time and also the course of the fever reaction warrants us in concluding that this case was one of a slight attack of horse-sickness.

III.—*To note whether smaller quantities than a* 1,000 *c.c. of fortified serum will produce the same effect as in No. II.*

Remark.—Horse S.H. 1 had been immunised in the meantime to the extent of 3,520 c.c. virulent blood.

Experiment No. 3.—13-10-02.—Horse 163, injected subcutaneously with 800 c.c. serum of Horse S.H. 1, and 1 c.c. virus, 102, into the tail, simultaneously.

Result.—Horse 163 died on 29-10-02, or 16 days after the injection, from horse-sickness.

Experiment No. 4.—13-10-02.—Horse 170, injected with 600 c.c. serum, subcutaneously, of Horse S.H. 1, and 1 c.c. virus, No. 102, into the tail, simultaneously.

Result.—Horse 170 died on 27-10-02, or 14 days after the injection, from horse-sickness.

Experiment No. 5.—13-10-02.—Horse 162, injected with 400 c.c. serum of Horse S.H. 1, subcutaneously, and 1 c.c. virus, Horse 102, into the tail, simultaneously.

Result.—Horse 162 died on 2-11-02, or 20 days after the injection, from horse-sickness.

Conclusion.—The serum of Horse S.H. 1, hyper-immunised to the extent of 3,520 virulent blood, in quantities of 800 c.c. and less, did not protect against the simultaneous injection of virus.

IV.—*To note whether* 100 *c.c. serum of a highly hyper-immunised horse will protect against a simultaneous injection of virus.*

Remark.—Horse S.H. 1 was hyperimmunised to the extent of 8,020 c.c. virulent blood up to the date of the experiment.

Experiment No. 6.—22-11-02.—Horse 220, injected subcutaneously with 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus, Horse 102, subcutaneously.

Result.—Horse 220 died of horse-sickness on 7-12-02, or 15 days after the injection.

Conclusion.—The injection of 100 c.c. of a fortified serum did not protect a horse against a simultaneous injection of virus.

V.—*To note whether* 100 c.c. *serum of a still higher immunised horse will protect against a simultaneous injection of virus.*

Remark.—Horse S.H. 1 was, up to the time of the experiment, hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 7.—Horse 302.—1-5-03, injected with 100 c.c. serum of Horse S.H. 1, and 1 c.c. virus, Horse No. 295.

Result.—Horse 302 died 10-5-03 from horse-sickness.

Conclusion.—The injection of 100 c.c. serum of a horse hyper-immunised to the extent of 9,020 c.c. virulent blood did not protect against a simultaneous and subcutaneous injection of virus.

VI.—*To note whether* 50 *and* 100 *c.c. serum of a hyper-immunised horse, injected into the jugular vein, will protect against a simultaneous subcutaneous injection of virus.*

Remark.—Horse S.H. 1 had been hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 8.—22-11-02.—Mule 205, injected with 50 c.c. serum of Horse S.H. 1, intrajugularly, and 1 c.c. virus, Horse 102, subcutaneously.

Result.—Mule 205 died on 5-12-02, or 13 days after injection, from horse-sickness.

Experiment No. 9.—22-11-02.—Mule 203, injected with 100 c.c. serum, Horse S.H. 1, intrajugularly, and 1 c.c. virus, subcutaneously.

Result.—Mule 203 died on 10-12-02, from horse-sickness, viz., 18 days after injection.

Experiment No. 10.—Mule 198.—On 22–11–02, injected with 100 c.c. serum, S.H. 1, into the jugular vein, and 1 c.c. virus, Horse 102, subcutaneously.

Result.—Mule 198 died on 30–11–02, or eight days later, from horse-sickness.

Experiment No. 11.—Mule 201.—On 22–11–02, was injected with 100 c.c serum, S.H. 1, into the jugular vein, and 1 c.c. virus, Horse 102, subcutaneously.

Result.—No reaction. Animal proved to be immune.

Conclusion.—The injection of 50 c.c. and 100 c.c. serum into the jugular vein did not protect against a simultaneous inoculation of virus subcutaneously. One mule was found to be immune.

VII.—*To note the effect of serum—from a mixture of serum and virus—after the serum had been separated by centrifugation.*

Remark.—Horse S.H. 1 had been immunised to the extent of 3,520 c.c. virulent blood. The mixture was made with 100 c.c. serum and 1 c.c. virus. It stood for five days, and was then centrifugalised.

Experiment No. 12.—Horse 152, injected on 20–10–02 with serum alone, from a mixture of 100 c.c. serum and 1 c.c. virus of Horse 153.

Result.—No reaction ensued from the injection. The Horse 152 was tested on its immunity on 4–11–02 with 1 c.c. virus, Horse 122. No reaction took place. Re-tested on 19–11–02 with 10 c.c. virus of Horse 102. Horse 152 contracted horse-sickness and died on 3–12–02.

Remark.—Horse S.H. 1 had been hyper-immunised to the extent of 5,520 c.c. virulent blood. The mixture consisted of 100 c.c. serum, Horse S.H. 1, and 10 c.c. virus of Horse 158. It stood for three days, and was then separated.

Experiment No. 13.—11–11–02.—Horse 212 was injected with serum of a mixture of 100 c.c. serum and 10 c.c. virus.

Result.—No reaction.—Horse 212 was tested on, its immunity on 26–11–02, or 15 days later, with 25 c.c. virus of Horse 216. No reaction resulted from this injection. It was re-tested on its immunity on 7–2–03, with 1 c.c. virus of Horse 262. It contracted horse-sickness, and on 15–2–03, or eight days later, died from horse-sickness.

Conclusion.—The serum alone, from a mixture of serum and virus, was not virulent. It protected susceptible animals against a subsequent inoculation of virus, injected 15 days after the serum.

VIII.—*To note whether* 100 *c.c. of a fortified serum, injected subcutaneously, protects against a subsequent injection of* 1 *c.c. virus* 24 *hours after the serum.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 5,520 c.c. virulent blood.

Experiment No. 14.—Mule 202.—8–11–02, injected with 100 c.c. serum of Horse S.H. 1. 9–11–02, injected subcutaneously with 1 c.c. virus of Horse 158.

Result.—On 25–11–02—Mule 202, was injected with 20 c.c. virus of Horse No. 216. The injection of virus produced no reaction. The mule was used for another experiment (*vide* Experiment 19), and died on 15–2–03, from horse-sickness.

Conclusion.—The injection of 100 c.c. of a fortified serum protected a susceptible mule against the inoculation of 1 c.c. virus injected 24 hours later, and also against 20 c.c. virus injected 17 days later.

IX.—*To note whether* 100 *c.c. of a fortified serum protects against a subsequent injection of* 1 *c.c. virus when inoculated* 10 *days after the serum.*

Remark.—Horse S.H. 1 had been hyper-immunised to the extent of 3,520 virulent blood.

Experiment No. 15.—Horse 182.—26–10–02 injected with 100 c.c. serum, Horse S.H. 1, subcutaneously. On 5–11–02 injected with 1 c.c. virus of horse 102.

Result.—Horse 182 died on 15-11-02, or 10 days after the injection of virus, from horse-sickness (dikkop).

Conclusion.—The subcutaneous injection of 100 c.c. serum did not protect a susceptible horse against a subsequent inoculation of virus injected 10 days later.

X.—*To note whether* 100 *c.c. of a fortified serum protects against a subsequent injection of* 1 *c.c. virus when inoculated* 15 *days after the serum.*

Remark.—Horse S.H. 1 had been hyperimmunised to the extent of 3,520 virulent blood.

Experiment No. 16.—Horse 187.—26-10-02, injected with 100 c.c. serum, Horse S.H. 1. On 11-11-02, injected with 1 c.c. virus, Horse 102.

Result.—No reaction from the inoculation of 1 c.c. virus. This horse was tested on its immunity on 1-12-02, by a subcutaneous injection of 10 c.c. virus, Horse No. 174. It died nine days later, on 10-12-02, from horse-sickness.

Conclusion.—The injection of 100 c.c. fortified serum protected a susceptible horse against the inoculation of 1 c.c. virus made 15 days after the serum.

XI.—*To note whether the subcut aneous injection of* 50 *c.c. of a fortified serum will protect against* 1 *c.c. virus injected* 24 *hours later into the jugular vein.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 virulent blood. Serum of Horse 168 was used as virus.

Experiment No. 17.—Horse 229.—On 4-1-03, injected with 50 c.c. serum, Horse S.H. 1, subcutaneously. On 5-1-03, injected with 1 c.c. virus (virulent serum), intrajugularly.

Result.—Horse 229 did not react from the injection of 1 c.c. virus. It was tested on its immunity on 7-2-03 with virus, Horse 262. It died eight days later (15-2-03) from horse-sickness.

Conclusion.—The subcutaneous injection of 50 c.c. serum protected a horse against 1 c.c. virus (virulent serum) injected into the jugular vein 24 hours later.

XII.—*To note whether the injection of* 100 *c.c. fortified serum into the jugular vein will protect against a subsequent subcutaneous inoculation of virus made* 24 *hours later.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 5,520 c.c. virulent blood.

Experiment No. 18.—Mule 200.—8-11-02, injected with 100 c.c. serum, intrajugularly. 9-11-02 injected with 1 c.c. virus of Horse No. 158.

Result.—The mule contracted horse-sickness and died 14 days after the injection (23-11-02).

Conclusion.—The intrajugular injection of 100 c.c. serum did not protect a mule against a subcutaneous injection of 1 c.c. virus made 24 hours later.

XIII.—*To note whether the intrajugular injection of* 100 *c.c. of a fortified serum will protect against a subsequent subcutaneous injection of virus when made more than* 20 *days after the serum was injected.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 virulent blood. As virus was used, virulent serum, Horse 168.

Experiment No. 19.—Mule 202 (*vide* Experiment 14).—On 6-12-02, injected with 100 c.c. serum, intrajugularly. On 29-12-02, viz., 23 days later, injected with 1 c.c. virulent serum, subcutaneously.

Result.—On 7-2-03, the mule was tested for its immunity, and 5 c.c. virulent blood of Horse 262 was injected subcutaneously. Mule 202 contracted horse-sickness and died on 15-2-03, viz., eight days after the injection.

Conclusion.—The injection of 100 c.c. fortified serum into the jugular vein protected the mule against an injection of 1 c.c. virus (virulent serum) made 23 days later.

XIV.—*To note the effect of an injection of a 24 hours old mixture of 100 c.c. immune serum and 1 c.c. virus injected subcutaneously.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 3,520 c.c. virulent blood.

Experiment No. 20.—Horse 173.—11-10-02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 153, taken the day previously.

Result.—No reaction took place. Horse 173 was tested for its immunity on 8-11-02, with 1 c.c. virus of Horse 158, injected subcutaneously. The horse developed glanders, and died 18 days after the injection of virus.

Conclusion.—There is no proof that horse 173 was not already immune before injection.

XV.—*To note the effect of an injection of a mixture 24 hours old of 100 c.c. immune serum and 5 c.c. virus, injected subcutaneously.*

Remark.—Horse S.H. 1 had been hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 21.—Horse 267.—On 7-2-03, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 5 c.c. virus of Horse 262. The mixture was 24 hours old.

Result.—Horse 267 died on 16-2-03, from horse-sickness, viz., nine days after injection.

Experiment No. 22.—Horse 265.—On 7-2-03, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 5 c.c. virus of Horse 262. The mixture was 24 hours old.

Result.—Horse 265 died 18 days after the injection (25-2-03) from horse-sickness.

Experiment No. 23.—Horse 248.—On 7-2-03, injected with a mixture of 100 c.c. serum and 5 c.c. virus. The mixture was 24 hours old.

Result.—Horse 248 showed no reaction. It was tested for its immunity on 4-3-03 with 50 c.c. virulent blood of Horse 279. It died of horse-sickness on 12-3-03, or eight days after injection.

Experiment No. 24.—Mule 151.—On 7-2-03, injected with a mixture of 100 c.c. serum and 5 c.c. virulent blood of Horse 262. The mixture was 24 hours old.

Result.—Mule 151 contracted horse-sickness and died on 17-2-03, or 10 days after the injection.

Conclusion.—The mixture of 100 c.c. serum and 5 c.c. virus, 24 hours old, produced the disease in three of the four experiments. It proved not to be virulent in a susceptible horse (Experiment No. 23).

XVI.—*To note the effect of an injection of a mixture, 24 hours old, of 100 c.c. immune serum and 10 c.c. virus, injected subcutaneously.*

Remark.—(Compare Experiments 48 and 74).—Horse 181 had been injected 33 days before with serum from a mixture of serum and virus. Horse S.H. 1 had been hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 25.—7-2-03.—Horse 181 was injected with a mixture of 100 c.c. serum and 10 c.c. virus of Horse 262, 24 hours old.

Result.—No reaction took place. Horse 181 was tested for its immunity on 5-3-03 with 100 c.c. virulent blood of Horse 278. It contracted horse-sickness and died on 14-3-03, or nine days after the injection of virus.

Conclusion.—The 24 hours old mixture of 100 c.c. serum and 10 c.c. virus did not prove to be virulent. It is possible that the injection, made 33 days previously, had some protecting influence.

XVII.—*To note the effect of a mixture of* 100 *c.c. serum and* 5 c.c. *virus*, 24 *hours old, when injected into the jugular vein.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 26.—Horse 257.—On 7-2-03, injected intrajugularly with a mixture of 100 c.c. serum, Horse S.H. 1, and 5 c.c. virus, Horse 262, 24 hours old.

Result.—Horse 257 died of horse-sickness on 16-2-03, or nine days after the injection.

Conclusion.—The 24 hours old mixture of 100 c.c. serum and 5 c.c. virus injected into the jugular vein proved to be virulent.

XVIII.—*To note the effect of the subcutaneons injection of a mixture of serum and virus*, 48 *hours old.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood. Serum of Horse 168 was used as virus.

Experiment No. 27.—Horse 236.—On 31-12-02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 5 c.c. virulent serum of Horse 168.

Result.—No reaction took place. Horse 236 had to be killed on 27-1-03 on account of glanders.

Conclusion.—The mixture of 100 c.c. serum and 5 c.c. virus, 48 hours old, did not prove to be virulent. There is no evidence that Horse 236 was immune previous to the injection.

XIX.—*To note the effect of the intrajugular injection of a mixture of serum and virus*, 48 *hours old.*

Remark.—Horse S.H. 1, was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 28.—Horse 228.—On 5-1-03, injected intrajugularly with a mixture of 100 c.c. serum of Horse S.H. 1, and 0·5 c.c. virulent serum of Horse 168.

Result.—Horse 228 died on 14-1-03 from equine piroplasmosis.

Experiment No. 29.—Horse 259.—On 16-1-03, injected intrajugularly with a mixture of 100 c.c. serum of Horse S.H. 1 and 0·5 c.c. virulent serum of Horse 168.

Result.—No reaction ensued. On 7-2-03, Horse 259 was tested for its immunity by an injection of 10 c.c. virus of Horse 262. The horse died 12 days after injection from horse sickness (dikkop).

Experiment No. 30.—Horse 258 was injected on 16-1-03 with a mixture of 100 c.c. serum and 0·5 c.c. virulent serum of Horse 168, injected into the jugular vein.

Result.—No reaction ensued. On 7-2-03, Horse 258 was tested for its immunity with an injection of 5 c.c. virus of Horse 262. The horse died 10 days after injection from horse-sickness (16-2-03).

Conclusion.—A mixture of 100 c.c. serum, Horse S.H. 1, and 0·5 c.c. virulent serum did not produce the disease.

There is some doubt whether 0·5 c.c. serum of a sick horse is sufficient to act as virus (compare No. 8 of introduction).

XX.—*To note the effect of a subcutaneous injection of a mixture of* 100 *c.c. serum and* 1 *c.c. virus three days old.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 3,520 virulent blood.

Experiment No. 31.—Horse 175.—On 13-10-02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 153.

Result.—No reaction. On 12–11–02, tested for its immunity with 1 c.c. virus of Horse 102. No reaction. Tested a second time on 1–12–02 with 10 c.c. virus of Horse 174. Horse 175 contracted horse-sickness and died 11 days after the injection (11–12–02).

Remark.—Horse S.H. 1 hyper-immunised to the extent of 5,520 c.c. virulent blood.

Experiment No. 32.—Mule 207.—On 11–11–02, injected with a mixture of 100 c.c. serum and 1 c.c. virus of Horse 158, three days old.

Result.—No reaction. Re-injected on 16–11–02 with 1 c.c. virus of Horse 102. No reaction. Tested on 1–12–02 with 10 c.c. virus of Horse 174. Died on 16–12–02 from horse-sickness.

Conclusion.—A mixture of 100 c.c. serum and 1 c.c. virus, three days old, did not produce the disease in two susceptible animals.

XXI.—*To note the effect of a subcutaneous injection of a mixture of* 100 *c.c. serum and* 10 *c.c. virus, three days old.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 33.—Horse 225.—On 8–12–02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 10 c.c. virus of Horse 174.

Result.—Horse 225 died 13 days later (21–12–02) from horse-sickness.

Experiment No. 34.—Horse 224.—On 8–12–02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 10 c.c. virus of Horse 174.

Result.—Horse 224 died on 28–12–02, or 20 days later, from debility.

Conclusions.—There is no proof that Horse 224 was immune previous to the injection. The three days old mixture of 100 c.c. serum and 10 c.c. virus proved to be virulent in one case (Experiment 33).

XXII.—*To note the effect of a subcutaneous injection of a mixture of* 100 *c.c. serum and* 50 *c.c. virus, three days old.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 35.—Horse 218.—On 8–12–02, injected with a three days old mixture of 100 c.c. serum and 50 c.c. virus of Horse 174.

Result.—Horse 218 died on 16–12–02, or eight days after injection, from horse-sickness.

Conclusion.—The three days old mixture of 100 c.c. serum and 50 c.c. virus proved to be virulent.

XXIII.—*To note the effect of a mixture of* 100 *c.c. serum and* 0·5 *c.c. virus, three days old, injected intrajugularly.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 5,520 virulent blood.

Experiment No. 36.—Horse 216.—On 11–11–02, injected with a mixture of 100 c.c. serum and 0·5 c.c. virus of Horse 158 into the jugular vein.

Result.—Horse 216 died on 24–11–02 from horse-sickness.

Conclusion.—The mixture of 100 c.c. serum and 0·5 c.c. virus, injected intrajugularly, proved to be virulent.

XXIV.—*To note the effect of a subcutaneous injection of a mixture of* 100 *c.c. serum and* 1 *c.c. virus five days old.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 2,020 c.c. virulent blood.

Experiment No. 37.—Horse 134.—On 17–9–02, injected with a mixture of 100 c.c. serum and 1 c.c. virus of Horse 132. The mixture was five days old.

Result.—Horse 134 died on 29-9-02 from debility. No lesions of horse-sickness were present.

Experiment No. 38.—Horse 158.—On 24-9-02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 146. The mixture was five days old.

Result.—No reaction from the injection. Tested after 13 days (9-10-02) for its immunity with 1 c.c. virus of Horse 102, subcutaneously. Horse 158 died on 2-11-02, or 23 days after the injection of virus, from horse-sickness.

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 3,520 c.c. virulent blood.

Experiment No. 39.—Horse 174.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum, Horse S.H. 1 and 1 c.c. virus of Horse 169.

Result.—No reaction ensued from this injection. Horse 174 was tested for its immunity on 20-11-02, or 24 days later, with 1 c.c. virus of Horse 102, subcutaneously. Horse 174 died on 1-12-02 from horse-sickness (viz., 11 days after the injection of virus).

Experiment No. 40.—Horse 184.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 169.

Result.—No reaction ensued from this injection. Horse 184 was tested on its immunity on 5-11-02, or 11 days later, with 1 c.c. of virus, Horse 102, subcutaneously. It died on 18-11-02, or 13 days after the injection of virus, from debility

Experiment No. 41.—Horse 185.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum and 1 c.c. virus, Horse 169.

Result.—No reaction ensued from this injection. Horse 185 was tested for its immunity on 5-11-02, or 11 days after the injection, with 1 c.c. virus of Horse 102, subcutaneously. It died from horse-sickness on 17-11-02, or 12 days after the injection of virus.

Experiment No. 42.—Horse 189.—On 26-10-02, injected with a mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 169.

Result.—No reaction ensued from this injection. Horse 189 was tested for immunity on 11-11-02, or 16 days later, with 1 c.c. virus of Horse 102, injected intrajugularly. No reaction followed this injection. It was tested again on 26-11-02, or 31 days after the first injection, with 50 c.c. virus, subcutaneously. Horse 189 contracted horse-sickness and died on 9-12-02, or 14 days after the last injection.

Experiment No. 43.—Horse 193.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum and 1 c.c. virus, Horse 169.

Result.—No reaction ensued from this injection. On 5-11-02, or 10 days after the first injection, Horse 193 was tested for its immunity with 10 c.c. virus, Horse '102, subcutaneously. No reaction followed from this injection. On 20-11-02, or 25 days after the first injection, Horse 193 was tested again for its immunity with 50 c.c. virus of Horse 166. It then contracted horse-sickness and died on 28-11-02, or eight days later, from horse-sickness.

Experiment No. 44.—Horse 191.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum and 1 c.c. virus of Horse 169.

Result.—No reaction ensued from this injection. Horse 191 was tested with 1 c.c. virus, injected intrajugularly. It contracted horse-sickness and died on 15-11-02, or 10 days later.

Experiment No. 45.—Horse 181.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 169.

Result.—No reaction (Horse 181 was later used for Experiments, Nos. 25 and 74). It was tested for immunity on 5-3-03 with 100 c.c. defibrinated blood, Horse 278, and died of horse-sickness on 14-3-05, or nine days after the injection.

Experiment No. 46.—Mule 151.—On 26-10-02, injected with a five days old mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 169.

Result.—No reaction. Mule 151 was later used for Experiment No. 24 and died of horse-sickness.

Experiment No. 47.—Mule 150.—On 26-10-02, injected with a mixture of 100 c.c. serum and 1 c.c. virus of Horse 169, five days old.

Result.—No reaction ensued from the first injection. It was tested for its immunity on 11-11-02 with 1 c.c. virus. No reaction took place after this. It was tested again on 26-11-02 with 10 c.c. virus. A reaction took place after seven days, from which the animal recovered. It proved to be immune after the reaction and was hyper-immunised.

Experiment No. 48.—Mule 179.—On 26-10-02, injected with a mixture, five days old, of 100 c.c. serum and 1 c.c. virus of Horse 169.

Result.—No reaction took place. It was killed later, on account of an accident. (Compare also Experiments Nos. 55 and 95.)

Remark.—Serum Horse 160 was used. This horse was hyper-immunised to the extent of 9,510 c.c. virulent blood.

Experiment No. 49.—Horse 376.—On 10-10-03, injected with a five days old mixture of 100 c.c. serum of Horse 160, and 1 c.c. virus of Horse 350.

Result.—No reaction took place. This horse was not tested for its immunity.

Conclusion.—In no instance was the injection of a five days old mixture of 100 c.c. serum and 1 c.c. virus followed by a reaction. The injection protected against a subsequent inoculation of virus made, respectively, 10 days and 16 days later, but did not protect against a subsequent inoculatlon of virus made, respectively, 10, 11, 13 and 24 days later.

XXV.—*To note the effect of a subcutaneous injection of a five days old mixture of* 100 *c.c. serum and* 10 *c.c. virus.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 3,520 virulent blood.

Experiment No. 50.—Horse 177.—On 19-10-02, injected with a five days old mixture of 100 c.c. serum, Horse S.H. 1, and 10 c.c. virus of Horse 153.

Result.—No reaction ensued from this injection. On 18-11-02, viz., 30 days after the first injection, Horse 177 was tested with 1 c.c. virus of Horse 102. No reaction followed. On 4-12-02, viz., 44 days after the first injection, Horse 177 was re-injected with 10 c.c. virus of Horse 174. It then contracted horse-sickness and died on 12-12-02, viz., eight days after the last injection of virus.

Conclusion.—The five days old mixture of 100 c.c. serum and 10 c.c. virus did not produce horse-sickness. The injection protected at least 30 days against a subsequent injection of virus.

XXVI.—*To note the effect of a five days old mixture of* 10 *c.c. serum and* 1 *c.c. virus, injected into the jugular vein.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 3,520 c.c. virulent blood.

Experiment No. 51.—Horse 192.—26-10-02, injected with a five days old mixture of 100 c.c. serum and 1 c.c. virus of Horse 102, injected intrajugularly.

Result.—Horse 192 died of horse-sickness on 3-11-02, or eight days later.

Conclusion.—The five days old mixture of 100 c.c. serum and 1 c.c. virus, injected into the jugular vein produced horse-sickness.

XXVII.—To note the effect of a 22 days old mixture of 100 c.c. serum and 10 c.c. virus, injected intrajugularly.

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 52.—Mule 222.—On 1-12-02, injected with a 22 days old mixture of 100 c.c. serum, S.H. 1, and 10 c.c. virus, intrajugularly.

Result.—No reaction ensued from this injection. Tested on 29-12-02, with 10 c.c. virulent serum of Horse 168. No reaction ensued from this inoculation. Re-tested on 22-1-03 with 20 c.c. virus of Horse 102, intrajugularly. Mule 222 contracted the disease and died seven days later from horse-sickness.

Conclusion.—The 22 days old mixture of 100 c.c. serum and 10 c.c. virus, injected intrajugularly, did not produce the disease. The injection protected against a subsequent injection of virus, made 29 days later.

XXVIII.—*To note the effect of a 15 days old mixture of 100 c.c. serum and 10 c.c. virus injected into the jugular vein.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 53.—Horse 249.—On 14-1-03, injected with a 15 days old mixture of 100 c.c. serum and 10 c.c. virulent serum of Horse 168 into the jugular vein.

Result.—No reaction. Tested after 22 days with 10 c.c. virus of Horse 262. Died three days later from horse-sickness, or 25 days after the injection of the mixture.

Conclusion.—The 15 days old mixture produced the disease after a long incubation time.

XXIX.—*To note the effect of one month old mixture of 100 c.c. serum and 10 c.c. virus injected into the jugular vein.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 54.—Horse 264.—On 6-2-03, injected with a mixture of 100 c.c. serum and 10 c.c. virulent serum of Horse 168.

Result.—No reaction. This horse was used later for Experiment No. 79. It then contracted horse-sickness, recovered, and was later hyper-immunised.

Experiment No. 55.—Mule 179.—On 6-2-03, injected with a one month old mixture of 100 c.c. serum and 10 c.c. virulent serum of Horse 168.

Result.—No reaction. (Compare also Experiments Nos. 48 and 95.) This mule was killed later, owing to an accident.

XXX.—*To note the effect of an 85 days old mixture of serum and virus injected intrajugularly.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 56.—Horse 256.—On 14-1-03, injected intrajugularly with a mixture 85 days old, of 100 c.c. serum and 1 c.c. virus of Horse 169.

Result.—No reaction. Tested 7-2-03, or 24 days later, with 10 c.c. virus of Horse 262. Died on 12-2-03 from glanders.

Experiment No. 57.—Horse 265.—On 14-1-03, injected with a mixture, 85 days old, of 100 c.c. serum and 5 c.c. virus of Horse 169.

Result.—No reaction. This horse died on 1-2-03, or 15 days after injection, from torsion of the colon.

Conclusion.—Mixture, 85 days old, of 100 c.c. serum and 5 c.c. virus, injected into the jugular vein, did not produce the disease.

XXXI.—*To note the effect of the deposit of a mixture of serum and virus, after the deposit had been separated by centrifugation and washed out with physiological water.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 3,520 c.c. virulent blood. The mixture was made with 100 c.c. serum and 1 c.c. virus. It stood for five days and was then centrifugalised and washed with aq. physiol.

Experiment No. 58.—Horse 166.—19-10-02, injected with the deposit of a mixture of serum, Horse S.H. 1, and 1 c.c. virus of Horse 153, after it had been washed with aq. physiol.

Result.—No reaction resulted from the injection. Horse 166 was tested for its immunity on 4-11-02 with 1 c.c. virus of Horse 102. It contracted horse-sickness and died on 13-11-02, or nine days later.

Conclusion.—The washed out deposit of a five days old mixture of 100 c.c. serum and 1 c.c. virus, did not prove to be virulent.

XXXII.—*To note the effect of the deposit of serum and virus after it had been separated by centrifugation.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 5,520 c.c. virulent blood. The mixture was made with 100 c.c. serum and 10 c.c. virus, and was three days old. It was centrifugalised and the red corpuscles were injected.

Experiment No. 59.—Horse 208.—11-11-02, injected with deposit of a mixture of 100 c.c. serum and 10 c.c. virus of Horse 158.

Result.—Horse 208 died on 22-11-02, or 11 days after the injection of the deposit, from horse-sickness.

Conclusion.—The deposit of a mixture of 100 c.c. serum and 10 c.c. virus, proved to be virulent.

XXXIII.—*To note the effect of an intrajugular injection of a mixture of* 100 *c.c. serum and* 5 *c.c. virus,* 24 *hours old, into an animal which,* 24 *hours previously, had been injected with* 10 *c.c. serum.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 60.—Horse 244.—On 6-2-03, injected with 10 c.c. serum, Horse S.H. 1, subcutaneously. On 7-2-03, injected into the jugular vein with a 24 hours old mixture of 100 c.c. serum and 5 c.c. virus, Horse 262.

Result.—Horse 244 died on 20-2-03 from horse-sickness, 13 days after the injection.

Experiment No. 61.—Horse 251.—On 6-2-03, injected with 10 c.c. serum, Horse S.H. 1, subcutaneously. On 7-2-03, injected with a 24 hours old mixture of 100 c.c. serum and 5 c.c. virus, Horse 262.

Result.—Horse 251 died on 18-2-03, or 11 days after the injection, from horse-sickness.

Conclusion.—The subcutaneous injection of 10 c.c. serum, made 24 hours previously, to the intrajugular injection of a 24 hours old mixture of serum and virus, did not prevent the development of the disease.

XXXIV.—*To note the effect of an intrajugular injection of a mixture of* 100 *c.c. serum and* 5 *c.c. virus,* 24 *hours old, into an animal which,* 24 *hours previously, had been injected with* 20 *c.c. serum.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 62.—Horse 252.—On 6-2-03, injected with 20 c.c. serum. On 7-2-03, injected into the jugular vein with a 24 hours old mixture of 100 c.c. serum and 5 c.c. virus of Horse 262.

Result.—Horse 252 died on 15-2-03, or eight days later, from horse-sickness.

Experiment No. 63.—Horse 253.—On 6-2-03, injected with 20 c.c. serum. On 7-2-03, injected into the jugular vein with a 24 hours old mixture of 100 c.c. serum and 5 c.c. virus of Horse 262.

Result.—Horse 253 died on 17-2-03, or 10 days later, from horse-sickness.

Conclusion.—The subcutaneous injection of 20 c.c. serum, made 24 hours previously to the intrajugular injection of a 24 hours old mixture of serum and virus, did not prevent the development of the disease.

XXXV.—*To note the effect of an intrajugular injection of a mixture of* 100 *c.c. serum and* 5 *c.c. virus*, 24 *hours old, into an animal which*, 24 *hours previously, had been injected with* 50 *c.c. serum.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 64.—*Horse* 260.—On 6-2-03, injected with 50 c.c. serum. On 7-2-03, injected with a 24 hours old mixture of 100 c.c. serum and 5 c.c. virus, intrajugularly.

Result.—Horse 260 died on 17-2-03, or 10 days later, from horse-sickness.

Experiment No. 65.—Horse 263.—On 6-2-03, injected with 50 c.c. serum On 7-2-03, injected with a 24 hours old mixture of 100 c.c. serum and 5 c.c. virus, intrajugularly.

Result.—Horse 263 died on 15-2-03, or nine days later, from horse-sickness.

Conclusion.—An injection of 50 c.c. serum, made 24 hours previous to the intrajugular injection of a mixture of 100 c.c. serum and 5 c.c. virus, did not prevent the development of the disease.

XXXVI.—*To note the effect of an inoculation of a two days old mixture of* 100 *c.c. serum and* 5 *c.c. virus* (*virulent serum*) *into the jugular vein* 24 *hours after a sub-cutaneous injection of* 50 c.c. *serum.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 66.—Horse 196.—4-1-03, injected subcutaneously with 50 c.c. serum, Horse S.H. 1. On 5-1-03, injected with a two days old mixture of 100 c.c. serum, Horse S.H. 1, and 5 c.c. virulent serum of Horse 168.

Result.—No reaction ensued. Horse 196 had to be killed on 27-1-03 on account of glanders.

Conclusion.—There is no proof that Horse 196 was immune previous to the injection.

XXXVII.—*To note the effect of an injection of a mixture of* 100 *c.c. serum and* 5 *c.c. virus* (*virulent serum*) *two days old, followed* 27 *days later by an injection of* (1) vide *Experiment* 67, 100 *c.c. serum and* 20 *c.c. virus* (*virulent serum*); (2) vide *Experiment* 68, *of* 100 *c.c. serum and* 30 *c.c. virus* (*virulent serum*); *and* (3) vide *Experiment* 69, *of* 100 *c.c. serum and* 50 *c.c. virus* (*virulent serum*).

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 67.—Horse 243.—On 31-12-02, injected subcutaneously with 100 c.c. serum and 5 c.c. virulent serum of Horse 168. The mixture was

two days old. No reaction took place. On 27–1–03, injected with a four days old mixture of 100 c.c. serum, Horse S.H. 1, and 20 c.c. virulent serum of Horse 168.

Result.—No reaction took place. Horse 243 was tested on 16–2–03, or 20 days after the last injection, which 20 c.c. virus of Horse 212, and died on 26–2–03 from horse-sickness.

Conclusion.—The mixture of 100 c.c. serum and 5 c.c. virulent serum two days old did not produce the disease; nor did a subsequent inoculation of a four days old mixture of 100 c.c. serum and 20 c.c. virulent serum. The horse did not acquire immunity from these injections.

Experiment No. 68.—Horse 233.—On 31–12–02, injected subcutaneously with a two days old mixture of 100 c.c. serum and 5 c.c. virulent serum of Horse 168. No reaction took place. On 27–1–03, injected with a mixture of 100 c.c. serum and 30 c.c. virulent serum; the mixture was four days old.

Result.—No reaction took place. Horse 233 was killed on account of glanders on 19–2–03, that is, 23 days after the last inoculation.

Experiment No. 69.—Horse 241.—On 31–12–02, injected subcutaneously with a two days old mixture of 100 c.c. serum and 5 c.c. virulent serum of Horse 168. No reaction took place. On 27–1–03, injected with a four days old mixture of 100 c.c. serum and 50 c.c. virulent serum of Horse 168.

Result.—No reaction took place. Horse 241 was killed on 12–2–03, or 15 days later, on account of glanders.

Conclusion.—The mixture of 100 c.c. immune serum and 5 c.c. virulent serum did not produce a reaction; nor did a subsequent inoculation of a mixture of serum and virulent serum, or, respectively, 30 c.c. and 50 c.c. virulent serum to 100 c.c. immune serum, produce the disease. There is no proof that these two horses were susceptible to horse-sickness.

XXXVIII.—*To note the effect of the injection of a three days old mixture of* 100 *c.c. immune serum and* 10 *c.c. and* 20 *c.c. virus, injected subcutaneously, and a subsequent inoculation* 23 *days later of a mixture of* 100 *c.c. immune serum and* 20 *c.c. and* 40 *c.c. virus (virulent serum).*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 70.—Horse 223.—On 8–12–02, injected with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 10 c.c. virus of Horse 174. No reaction took place. On 1–1–03, injected with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 20 c.c. virulent serum of Horse 168. No reaction took place.

Result.—Horse 223 was tested on its immunity on 22–1–03, with 20 c.c. virus of Horse 102, subcutaneously. It contracted horse-sickness and died on 4–2–03, or 13 days later.

Experiment No. 71.—Horse 217.—On 8–12–02, injected with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 20 c.c. virus of Horse 174. No reaction took place. On 1–1–03, injected with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 40 c.c. virulent serum of Horse 168. No reaction took place.

Result.—Horse 217 was tested on 22–1–03 with 40 c.c. virus of Horse 102. It contracted horse-sickness and died on 31–1–03, or nine days later.

Conclusion.—The injection of a mixture of 100 c.c. immune serum and 10 c.c. and 20 c.c. virulent serum did not produce the disease; nor did a subsequent inoculation, 23 days later, of 100 c.c. immune serum and 20 c.c. and 40 c.c. virulent serum produce the disease. The horse did not acquire immunity from these two injections.

XXXIX.—*To note the effect of the injection into the jugular vein of a three days old mixture of* 100 *c.c. immune serum and* 1 *c.c. virus into an animal which,* 24 *hours previously, had been injected subcutaneously with a three days old mixture of* 100 *c.c. serum and* 1 *c.c. virus.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 5,520 c.c. virulent blood.

Experiment No. 72.—Mule 199.—On 11–11–02, injected subcutaneously with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 158. On 12–11–02, injected into the jugular vein with a three days old mixture of 100 c.c. serum and 1 c.c. virus of Horse 158.

Result.—No reaction. This animal proved to be immune against horse-sickness, and was later hyper-immunised.

XL.—*To note the effect of the injection of a mixture of* 100 *c.c. immune serum and* 1 *c.c. virus into the jugular vein five days after the subcutaneous injection of a three days old mixture of* 100 *c.c. immune serum and* 1 *c.c. virus.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 5,520 virulent blood.

Experiment No. 73.—Mule 206.—On 11–11–02, injected subcutaneously with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 158. On 16–11–02, injected into the jugular vein with a three days old mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 158.

Result.—No reaction took place from these injections. Mule 206 died of acute glanders on 27–1–02.

Conclusion.—There is no proof that Mule 206 was susceptible to horse-sickness.

XLI.—*To note the effect of an injection of a mixture of* 50 *c.c. immune serum and* 1 *c.c. virus (virulent serum) into the jugular vein of an animal which,* 24 *hours previously, had been injected with* 50 *c.c. immune serum, subcutaneously.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 8,020 c.c. virulent blood.

Experiment No. 74.—Horse 181.—On 4–1–03 injected subcutaneously, with 50 c.c. serum, Horse S.H. 1, and on 5–1–03, injected with a mixture of 50 c.c. serum, Horse S.H. I, and 1 c.c. virulent serum of Horse 168. No reaction took place from these injections.

Result.—Horse 181 was used for Experiments, Nos. 25 and 45. No reaction. Tested on 5–3–03 with 100 c.c. virus of Horse 278. Died on 15–3–03, 10 days later from horse-sickness.

Conclusion.—Horse 181 did not acquire immunity by an inoculation of 50 c.c. serum injected 24 hours previously to the mixture of 50 c.c. serum and 1 c.c. virus.

XLII.—*To note the effect of a subcutaneous injection of a five days old mixture of* 100 *c.c. immune serum and* 10 *c.c. virus five days after a subcutaneous injection of a five days old mixture of* 100 *c.c. immune serum and* 1 *c.c. virus.*

Remark.—Horse 160 was hyper-immunised to the extent of 9,510 c.c. virulent blood.

Experiment No. 75.—Horse 387.—On 10–11–03, injected with a five days old mixture of 100 c.c. serum of Horse 160, and 1 c.c. virus of Horse 350. On 16–11–03, injected with a five days old mixture of 100 c.c. serum of Horse 160 and 10 c.c. virus of Horse 369.

Result.—No reaction took place. Horse 387 was tested on 7–5–04 with 5 c.c. virulent serum and died of horse-sickness on 13–5–04.

Conclusion.—The injection of a five days old mixture of 100 c.c. serum and 1 c.c. virus, followed five days later by a five days old mixture of 100 c.c. serum and 10 c.c. virus, did not produce the disease nor gave immunity.

XLIII.—*To note the effect of a subcutaneous injection of a five days old mixture of 100 c.c. immune serum and 20 c.c. virus five days after the subcutaneous injection of a five days old mixture of 100 c.c. immune serum and 1 c.c. virus.*

Remark.—Horse 160 was hyper-immunised to the extent of 9,510 c.c. virulent blood.

Experiment No. 76.—Horse 358.—On 10–11–03, injected with a five days old mixture of 100 c.c. serum of Horse 160 and 1 c.c. virus of Horse 350. On 16–11–03, injected with a five days old mixture of 100 c.c. serum of Horse 160 and 20 c.c. virus of Horse 369, subcutaneously.

Result.—No reaction took place. Horse 358 was not tested for its immunity.

Conclusion.—There is no proof that Horse 358 was susceptible to horse-sickness.

XLIV.—*To note the effect of an injection of serum and virus mixtures of different ages, repeated every fifth day, and with virus increasing in quantity, followed by pure virus.*

Remark.—Horse 160 was hyper-immunised to the extent of 9,510 c.c. virulent blood.

Experiment No. 77.—Horse 362, injected as follows:—9–10–03, with a mixture of 100 c.c. serum, Horse 160, and 1 c.c. virus of Horse 352. The mixture was 48 hours old. 14–10–03, with a mixture of 100 c.c. serum, Horse 160, and 5 c.c. virus of Horse 350; the mixture was five days old. 21–10–03, with a mixture of 100 c.c. serum, Horse 160, and 10 c.c. virus of Horse 350; the mixture was five days old. 26–10–03, with a mixture of 100 c.c. serum, Horse 160, and 20 c.c. virus of Horse 350; the mixture was five days old. 31–10–03, injected with 5 c.c. virus of Horse 350. 5–11–03, injected with 10 c.c. virus of Horse 350.

Result.—Horse 362 contracted horse-sickness five days after the injection of the first dose of pure virus, and died on 11–11–03.

Experiment No. 78.—Mule 368, injected as follows:—9–10–03, with a mixture of 100 c.c. serum, Horse 160, and 1 c.c. virus of Horse 352; the mixture was 48 hours old. 14–10–03, with a mixture of 100 c.c. serum, Horse 160, and 5 c.c. virus of Horse 350; the mixture was five days old. 21–10–03, with a mixture of 100 c.c. serum, horse 160, and 10 c.c. virus of Horse 350; the mixture was five days old. 26–10–03, with a mixture of 100 c.c. serum, Horse 160, and 20 c.c. virus of Horse 350; the mixture was five days old. 31–10–03, with 5 c.c. virus of Horse 350. 5–11–03, with 10 c.c. virus of Horse 350.

Result.—No reaction took place. This mule proved to be immune, and was later hyper-immunised.

Conclusion.—The repeated injections of mixtures of serum with increasing quantities of virus proved virulent in a Horse (362).

XLV.—*To note the effect of repeated injections of serum into the jugular vein after a simultaneous injection of serum and virus had been made subcutaneously.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood (*vide* Experiment 54).

Experiment No. 79.—Horse 264.—1–5–03, injected with 1 c.c. virus, Horse 295, and 100 c.c. serum, Horse S.H. 1, subcutaneously. 2–5–03, injected with 100 c.c. serum, intrajugularly. 5–5–03, injected with 100 c.c. serum, intrajugularly. 8–5–03, injected with 100 c.c. serum, intrajugularly. 11–5–03, injected with 100 c.c. serum, intrajugularly. 14–5–03, injected with 100 c.c. serum, intrajugularly. 17–5–03, injected with 100 c.c. serum, intrajugularly.

Result.—Reaction began on 10–5–03. It was slight. The fever never rose above 103·6. Symptoms of dikkop were present. The animal became very weak, but recovered. This horse was used later for hyper-immunisation.

Experiment No. 80.—Horse 301.—1–5–03, injected with 1 c.c. virus of Horse 295, and 100 c.c. serum, Horse S.H. 1, subcutaneously. 2–5–03, injected with 100 c.c. serum, intrajugularly. 5–5–03, injected with 100 c.c. serum, intrajugularly. 8–5–03, injected with 100 c.c. serum, intrajugularly.

Result.—Reaction began on 12–5–03, and lasted seven days, then the temperature dropped and a second reaction took place. There was a slight swelling noticed above the eyes. During the first reaction no change was noticed in the horse's health, but during the second reaction it became distinctly ill.

Conclusion.—The repeated injection of serum after a simultaneous injection of serum and virus modified the attack of the horse-sickness reaction.

XLVI.—*To note the effect of the injection of a mixture of equal parts of normal horse, mule, and donkey serum, simultaneously with a subcutaneous injection of immune serum and virus, both inoculated in different places. The injections of immune and normal serum to be repeated.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 81.—20–5–03, Mule 312 injected in different places:—On left side with 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus of Horse 295. On the right side, injected with 200 c.c. of a mixture of normal horse, mule, and donkey serum. 21–5–03, injected intrajugularly with 100 c.c. serum, Horse S.H. 1, and subcutaneously with a mixture of normal horse, mule, and donkey serum. 24–5–03, injected as under 21–5–03. 27–5–03, injected as under 21–5–03. 30–5–03, injected as under 21–5–03.

Result.—Incubation lasted six days. The fever reaction never rose above 103·6° F., and ended fatally on 31–5–03 (horse-sickness).

Experiment No. 82.—Horse 313.—20–5–03, simultaneous injection of 1 c.c. virus and 100 c.c. serum, Horse S.H. 1, in different places of right side of horse, and 200 c.c. mixture of normal horse, mule, and donkey serum, on left side.

Result.—The incubation time lasted eight days. There was a reaction during four days, which could not be with certainty identified with that of horse-sickness, but probably was so. The mule was tested on 22–6–03 with 10 c.c. virus, 300, injected intrajugularly. No reaction ensued. The animal became debilitated, and had to be killed on 6–7–03.

Experiment No. 83.—Mule 315.—20–5–03, simultaneous injection of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus, 295, in different places on right side, and a mixture of normal horse, mule, and donkey serum on left side. 21–5–03, injected 100 c.c. immune serum, intrajugularly, and 200 c.c. mixture of normal horse, mule, and donkey serum.

Result.—The incubation lasted eight days. The reaction ended fatally after 13 days (horse-sickness).

Conclusion.—The injections of immune and normal serum, after a simultaneous injection of virus and serum inoculated at different places, did not prevent the outbreak of the disease. One animal recovered from the reaction of horse-sickness.

XLVII.—*To note the effect of repeated inoculations at intervals with immune serum heated at 55° C. for half an hour, following a simultaneous injection of serum and virus.*

Remark.—Horse S.H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 84.—Horse 215.—20–5–03, injected simultaneously with 100 c.c. serum, Horse S.H. 1, heated to 55° C., and 1 c.c. virus, 295, subcutaneously. 21–5–03, injected intrajugularly with 100 c.c. serum, S.H. 1, heated to 55° C. 24–5–03, injected intrajugularly with 100 c.c. serum, S.H. 1, heated to 55° C. 27–5–03, injected intrajugularly with 100 c.c. serum, S.H. 1, heated to 55° C. 30–5–03, injected intrajugularly with 100 c.c. serum, S.H. 1, heated to 55° C. 2–6–03, injected intrajugularly with 100 c.c. serum, S.H. 1, heated to 55° C.

Result.—The incubation time lasted seven days. The reaction lasted seven days; then the temperature dropped. A second reaction took place, and the horse died on 14–6–03. The *post-mortem* proved absence of horse-sickness. The horse died of debility.

Experiment No. 85.—Horse 309.—20–5–03, injected simultaneously as Horse 215 (*vide* above). Subsequent inoculations as in Horse 215 (*vide* above). Last inoculation was made on 30–5–03.

Result.—The temperature rose gradually from the fifth day after the first injection, and rose till the 12th day, when the horse died of horse-sickness.

Experiment No. 86.—Mule 310.—Treated as Horse 215.

Result.—Reaction started five days after the injection, and lasted 13 days, when the mule died of horse-sickness.

Conclusion.—The repeated inoculations with heated serum did not protect the horses and one mule against a simultaneous injection of serum and virus.

XLVIII.—*To note the effect of repeated injections of serum into the jugular vein after a simultaneous injection of serum and virus was made subcutaneously; the first three serum injections to be made* 6, 12, *and* 18 *hours respectively, after the simultaneous injection.*

Remark.—Horse S.H. 1 had been hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 87.—Mule 321.—On 4–6–03, injected subcutaneously with 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus, simultaneously at 10 a.m. At 4 p.m. of the same date, viz., 6 hours later, 100 c.c. serum was injected into the jugular vein. 5–6–03, injected 100 c.c. serum into the jugular vein. 8–6–03, injected 100 c.c. serum into the jugular vein. 11–6–03, injected 100 c.c. serum into the jugular vein. 14–6–03, injected 100 c.c. serum into the jugular vein. 17–6–03, injected 100 c.c. serum into the jugular vein. 20–6–03, injected 100 c.c. serum into the jugular vein.

Result.—On the 21st day after the first injection a reaction took place, which lasted seven days. It was diagnosed as acute glanders, and the mule was shot.

Experiment No. 88.—Mule 320.—4–6–03, injected subcutaneously with 100 c.c. serum and 1 c.c. virus, 300, simultaneously, at 10 a.m. At 10 p.m. of the same date injected intrajugularly with 100 c.c. serum, Horse S.H. 1. 5–6–03, injected 100 c.c. serum into the jugular vein. 8–6–03, injected 100 c.c. serum into the jugular vein. 11–6–03, injected 100 c.c. serum into the jugular vein. 11–6–03, injected 100 c.c. serum into the jugular vein. 14–6–03, injected 100 c.c. serum into the jugular vein. 17–6 03, injected 100 c.c. serum into the jugular vein. 20–6–03, injected 100 c.c. serum into the jugular vein.

Result.—After an incubation period of eight days a reaction started which lasted six days, and which was typical for horse-sickness. This animal was tested with virus. It proved to be immune, and was hyper-immunised.

Experiment No. 89.—Mule 326.—4-6-03, injected with 100 c.c. serum and 1 c.c. virus, 300, subcutaneously at 4 p.m. On 5-6-03, injected with 100 c.c. serum, intrajugularly at 10 a.m. 6-6-03, injected 100 c.c. serum, intrajugularly. 9-6-03, injected 100 c.c. serum, intrajugularly. 12-6-03, injected 100 c.c. serum, intrajugularly. 15-6-03, injected 100 c.c. serum, intrajugularly. 18-6-03, injected 100 c.c. serum, intrajugularly. 21-6-03, injected 100 c.c. serum, intrajugularly.

Result.—Reaction began 13 days after the first injection. It lasted eight days, when distinct symptoms of acute glanders developed, and the animal was killed.

Conclusion.—The repeated intrajugular injections of serum after a simultaneous injection of serum and virus modified the horse-sickness reaction.

XLIX.—*To note the effect of repeated serum injections into the jugular vein, following an injection of serum and virus mixture into the jugular vein.*

Experiment No. 90.—Mule 323.—4-6-03, mixture of 100 c.c. serum, Horse S.H. 1, and 1 c.c. virus, 300, injected into the jugular vein, at 10 a.m. 4-6-03, injected into the jugular vein with 200 c.c. serum at 4 p.m. 5-6-03, injected 100 c.c. serum into the jugular vein. 8-6-03, injected 100 c.c. serum into the jugular vein. 11-6-03, injected 100 c.c. serum into the jugular vein. 14-6-03, injected 100 c.c. serum into the jugular vein.

Result.—The temperature began to rise three days after the first injection. It never rose above 104° F. The animal died on 15-6-03 from horse-sickness.

Conclusion.—Repeated intrajugular injections of serum did not prevent horse-sickness due to the injection of a mixture of serum and virus, intrajugularly.

L.—*To note the effect of injections of serum into the jugular vein started at twelve hours interval, after the simultaneous injections of serum and virus.*

Remark.—Horse 147 was hyper-immunised to the extent of 10,050 c.c. virulent blood.

Experiment No. 91.—Horse 289.—24-6-03, injected with 100 c.c. serum, Horse 317, and 1 c.c. virus, 300, at 10 a.m., subcutaneously. At 10 p.m. of the same date, injected intrajugularly with 100 c.c. serum, Horse 147. 25-6-03, injected with 100 c.c. serum, 147, intrajugularly. 28-6-03, injected with 100 c.c. serum 147, intrajugularly. 1-7-03, injected with 100 c.c. serum 147, intrajugularly. 4-7-03, injected with 100 c.c. serum 147, intrajugularly.

Result.—Horse 289 began to react seven days after the first injection The reaction lasted six days, when the animal died of horse-sickness.

Experiment No. 92.—Horse 296.—24-6-03, treated as above on 24-6-03, but with only one injection of serum afterwards, viz., on 25-6-03.

Result.—Hæmoglobinuria was noticed the first day after injection. The temperature kept on rising till the seventh day, when the animal died of horse-sickness and hæmolysis (1-7-03).

Conclusion.—Repeated injections of serum into the jugular vein, after a simultaneous injection of serum and virus, did not modify the horse-sickness reaction. Serum of horse 147 proved to be hæmolytic in one case.

LI.—*To note the effect of the simultaneous injection of virus, intrajugularly, and of serum, subcutaneously, followed six hours later by an injection of serum intrajugularly.*

Remark.—Horse 147 was hyper-immunised to the extent of 10,050 c.c. virulent blood.

Experiment No. 93.—Horse 317.—6-7-03, injected with 100 c.c. serum, subcutaneously, and 1 c.c. virus, 300, intrajugularly. Injected a second time, six hours later, with 200 c.c. serum, Horse 147.

Result.—The horse showed hæmoglobinuria, and died on 12-7-03 from hæmolysis.

Experiment No. 94.—Horse 297.—6-7-03, injected with 100 c.c. serum, subcutaneously, and 1 c.c. virus, 300, intrajugularly. Injected a second time, six hours later, with 200 c.c. serum, Horse 147.

Result.—The horse showed hæmolysis, and died on 9-7-03.

Conclusion.—Serum, Horse 147, proved to be hæmolytic for both horses.

LII.—To note the effect of a simultaneous injection of virus and serum into the jugular vein, followed by serum, intrajugularly.

Remark.—Horse 147 was hyper-immunised to the extent of 10,050 c.c. virulent blood.

Experiment No. 95.—Mule 179.—6-7-03, injected intrajugularly, 100 c.c. serum, Horse 147, and 1 c.c. virus, 300. It was reinjected six hours later with 200 c.c. serum, Horse 147, intrajugularly. 7-7-03, reinjected into the jugular vein, 100 c.c. serum, heated at 55° C. for half an hour, and again six hours later with 100 c.c. heated serum. 10-7-03, 100 c.c. heated serum injected into jugular vein. 13-7-03, 100 c.c. heated serum injected into jugular vein. 13-7-03, 100 c.c. heated serum injected into jugular vein.

Result.—No reaction. Mule 179 had to be killed, owing to an accident. (Compare Experiments 48 and 55.)

Experiment No. 96.—Horse 298.—6-7-03, injected into the jugular vein with 100 c.c. serum, Horse 147, and 1 c.c. virus, 300. 7-6-03, two injections into the jugular vein at six hours interval of 100 c.c. serum, Horse 147, heated at 55° C.

Result.—The Horse 298 died of hæmolysis on 10-7-03.

Conclusion.—The serum, Horse 147, proved to be hæmolytic for the horse, but not for the mule. No proof that Mule 179 was immune previous to the injection.

LIII.—To note the effect of large quantities of serum injected into the jugular vein simultaneously with virus, subcutaneously.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 97.—Horse 342.—24-7-03, injected into the jugular vein with 500 c.c. serum, Horse S. H. 1, and at the same time subcutaneously with 1 c.c. virus, 335.

Result.—Horse 342 died on 4-8-03, or 10 days later, of horse-sickness.

Experiment No. 98.—Mule 340.—24-7-03, injected into the jugular vein with 500 c.c. serum, Horse S. H. 1, and at the same time subcutaneously with 1 c.c. virus, 335.

Result.—Mule 240 died on 3-8-03 from horse-sickness.

Conclusion.—Large quantities of serum injected into the jugular vein did not prevent the disease due to the simultaneous subcutaneous injection of virus.

LIV.—To note the effect of injections of serum into the jugular vein at short intervals after a simultaneous injection of virus, subcutaneously, and serum, intrajugularly.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 99.—Horse 334.—On 24-7-03, injected with 100 c.c. serum, Horse S. H. 1, intrajugularly, and 1 c.c. virus, 300, subcutaneously. Six hours

later, on the same date, injected with 200 c.c. serum, intrajugularly. On 25-7-03, injected again with serum (200 c.c.), Horse S. H. 1, into the jugular vein.

Result.—Horse 334 died on 3-8-03, from horse-sickness.

Experiment No. 100.—Mule 345.—24-7-03, treated in the same way as Horse 334.

Result.—There was a reaction which started on the ninth day and lasted up to the nineteenth day. It was a typical horse-sickness reaction.

This mule was killed on 27-9-03 on account of debility.

Conclusion.—The injections of serum into the jugular vein at short intervals, after a simultaneous injection of virus, subcutaneously, and serum, intrajugularly, modified the horse-sickness reaction of the mule, but not of the horse.

LV.—To note the effect of repeated subcutaneous injections of serum at short intervals, after a simultaneous injection of serum and virus was made subcutaneously.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 101.—Horse 339.—On 24-7-03, injected subcutaneously with 100 c.c. serum, Horse S. H. 1, and 1 c.c. virus. Six hours later on the same date, injected subcutaneously with 200 c.c. serum, Horse S. H. 1. 25-7-03, injected with 200 c.c. serum, subcutaneously.

Result.—Horse 339 died on 5-8-03, from horse-sickness.

Experiment No. 102.—Mule 344.—On 24-7-03, injected subcutaneously with 100 c.c. serum, Horse S. H. 1, and 1 c.c. virus. Six hours later on the same date, injected simultaneously with 200 c.c. serum, Horse S. H. 1. 25-7-03, injected with 200 c.c. serum, subcutaneously.

Result.—Mule 344 did not show any reaction, and when tested later proved to be immune. It has been hyper-immunised since then.

Conclusion.—The repeated subcutaneous injections of serum at short intervals, after simultaneous injections of serum and virus, did not prevent death from horse-sickness in the horse.

LVI.—To note the effect of a simultaneous injection of serum into the jugular vein and virus, subcutaneously, preceded 24 hours by an injection of serum.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 9,020 c.c. virulent blood.

Experiment No. 103.—Horse 336.—26-7-03, injected subcutaneously at 10 a.m. with 100 c.c. serum, Horse S. H. 1. On 27-7-03, injected with 100 c.c. serum into the jugular vein and 1 c.c. virus, 335, subcutaneously. At 4 p.m. of the same date, reinjected into the jugular vein with 200 c.c. serum.

Result.—Horse 336 died ten days later, on 6-8-03, from horse-sickness.

Experiment No. 104.—Horse 337.—26-7-03, injected subcutaneously at 10 a.m. with 100 c.c. serum, Horse S. H. 1. On 27-7-03, reinjected with 100 c.c. serum into the jugular vein and 1 c.c. virus, subcutaneously. At 4 p.m. of the same date, reinjected with 200 c.c. serum into the jugular vein.

Result.—No reaction was noticed in this horse. It was tested on 10-10-03 with 1 c.c. virus, 350, and proved to be immune. It was killed on November 2nd, 1903, being then in a very poor condition.

Conclusion.—An injection of serum 24 hours previous to the simultaneous injection of serum, intrajugularly, and virus, subcutaneously, followed by an injection of serum into the jugular vein, did not prevent the development of the disease, neither did it modify the horse-sickness reaction.

LVII.—To note the effect of serum injected intrapectorally on the one side and virus, intrapectorally, on the other side of the animal.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 13,020 c.c. virulent blood.

Experiment No. 105.—Mule 1.—1-10-03, injected with 500 c.c. serum, Horse S. H. 1, intrapectorally, and 1 c.c. virus, 333, intrapectorally, on the opposite side.

Result.—Mule 1 died on 8-10-03 from horse-sickness.

Conclusion.—The simultaneous intrapectoral injection of serum and virus did not prevent the disease.

LVIII.—To note the effect of serum injected intrapectorally and virus, subcutaneously, on the opposite side.

Remark.—Horse S. H. 1 was hyperimmunised to the extent of 13,020 c.c. virulent blood.

Experiment No. 106.—Mule 2.—1-10-03, injected with 100 c.c. serum, intrapectorally, on the one side and 1 c.c. virus, 333, on the other, subcutaneously.

Result.—Mule 2 died on 9-10-03. The diagnosis not certain (Debility).

LIX.—To note the effect of a mixture of fortified serum of a horse, a mule and an ass.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 13,020 c.c virulent blood.

Mule 201 was hyper-immunised to the extent of 9,150 c.c. virulent horse. blood.

Donkey 306 was hyper-immunised to the extent of 2,350 c.c. virulent horse blood.

Experiment No. 107.—10-10-03, Horse 349 injected simultaneously with a mixture of serum, Horse S. H. 1, of Mule 201 and of Donkey 306, and 1 c.c. virus, 350, on the opposite side.

Result.—Horse 349 died of hæmolysis on 15-10-03.

Conclusion.—The mixture of fortified horse, mule and donkey serum proved to be hæmolytic.

Experiment No. 108.—Mule 355.—10-10-03, injected simultaneously with a mixture of serum, Horse S. H. 1, of Mule 201 and of Donkey 306, and 1 c.c. virus, 350, on the opposite side.

Result.—Mule 355 suffered severely from hæmolysis, passing red urine. It had a typical horse-sickness reaction, and recovered. It was bled during this reaction on 22-10-03, and 5 c.c. of its blood was tested on Horse ·374, which contracted horse-sickness and died.

Conclusion.—The mixture of fortified horse, mule and donkey serum proved to be hæmolytic. The mixture modified the reaction of horse-sickness in the mule.

LX.—To note the effect of a mixture of several sera of fortified horses and mules injected simultaneously with 1 c.c. virus.

Remark.—Horse S. H. 1 was hyper-immunised to the extent of 15,020 c.c. virulent blood. Horse 160 was hyper-immunised to the extent of 9,510 virulent blood. Horse 172 hyper-immunised to the extent of 7,150 virulent blood. Mule 150 hyper-immunised to the extent of 9,550 virulent blood. Mule 199 hyper-immunised to the extent of 9,000 virulent blood. Mule 201 hyper-immunised to the extent of 11,150 virulent blood.

Experiment No. 109.—Horse 331.—5-11-03, injected with 500 c.c. of a serum mixture of equal parts of serum, S. H. 1, Horse 160, Horse 172, and Mules 150, 199 and 201, simultaneously with 1 c.c. virus, 350.

Result.—Horse 331 died of hæmolysis on 8-11-03.

Conclusion.—The mixture of sera of fortified horses and mules proved to be hæmolytic.

Experiment No. 110.—Mule 386.—5-11-03, injected with 500 c.c. serum, mixture of equal parts of serum, Horse S. H. 1, Horse 160, Horse 172, and Mules 150, 199, and 201, simultaneously with 1 c.c. virus, 350.

Result.—Mule 386 showed reaction typical for horse-sickness. It was not tested for its immunity, and was destroyed on 10-3-04, suffering from epizootic lymphangitis.

Conclusion.—The mixture of several sera of fortified horses and mules proved not to be hæmolytic for the mule and modified the horse-sickness reaction.

LXI.—*To note the effect of a serum from a mule which has passed through an attack of hæmolysis, added to a mixture of different sera, which had a hæmolytic effect before, and injected simultaneously with* 1 *c.c. virus, subcutaneously.*

Remark.—The mixture was made with 1,000 c.c. serum, Mule 355 (*vide* Experiment 108), and equal parts of serum, Horses S. H. 1, 160, 172 and Mules 150, 199, and 201, to make up a litre (compare LX.). The mixture was heated to 55° C. for half an hour, and then kept for three days.

Experiment No. 111.—Horse 378.—13-11-03, injected with one litre of the above mixture and 1 c.c. virus, 369, subcutaneously.

Result.—Horse 378 died on 17-11-03 from hæmolysis.

Experiment No. 112.—Mule 356.—13-11-03, injected with one litre of the above mixture and 1 c.c. virus, 369, subcutaneously.

Result.—Mule 356 died of hæmolysis on 21-11-03.

Conclusion.—The serum of a mule which had passed through an attack of hæmolysis did not prevent the hæmolysis after being mixed with several hæmolytic sera.

LXII.—*To note the effect of a mixture of different fortified horse sera, to which was added an equal quantity of ox serum, simultaneously injected with* 1 *c.c. virus.*

Remark.—S. H. 1 hyper-immunised to 15,020 c.c. virulent blood; 160 hyper-immunised to 9,510 c.c. virulent blood; 172 hyper-immunised to 7,150 c.c. virulent blood.

Experiment No. 113.—Horse 346.—20-11-03, injected with a mixture of serum, Horses S. H. 1, 160 and 172, equal parts to make up 500 c.c., together with 500 c.c. ox serum on one side of the body and 1 c.c. virus, 369, on the other side.

Result.—Horse 346 died on 26-11-03, with lesions of hæmolysis and pulmonary lesions of horse-sickness.

Conclusion.—The mixture of serum of fortified horses, to which was added normal ox serum, proved to be hæmolytic.

LXIII.—*To note the effect of a mixture of different fortified horse sera, to which was added an equal quantity of sheep serum, simultaneously injected with* 1 *c.c. virus.*

Remark.—S. H. 1, 15,020 c.c. virulent blood. 160, 9,510 c.c. virulent blood. 172, 7,150 c.c. virulent blood.

Experiment No. 114.—Horse 384.—20-11-03, injected with a mixture of horse serum S. H. 1, 160, 172, equal parts to make up 500 c.c., together with 500 c.c. sheep serum on one side and 1 c.c. virus, 369.

Result.—No reaction took place. This horse was not tested for immunity.

Conclusion.—The mixture of fortified horse sera, to which normal sheep serum was added, proved not to be hæmolytic. There is no proof that Horse 384 was immune previous to the injection.

LXIV.—*To note the effect of a mixture of different fortified horse, mule and donkey sera, mixed with an equal amount of serum,* 355 (*supposed to be anti-hæmolytic*), *simultaneously with* 1 *c.c. virus.*

Remark.—S. H. 1, 15,020 c.c. virulent blood. 160, 9,510 c.c. virulent blood. 172, 7,150 c.c. virulent blood. 147, 10,050 c.c. virulent blood. 150, 9,550 c.c virulent blood. 199, 9,000 c.c. virulent blood. 201, 11,150 c.c. virulent blood. 306, 2,850 c.c. virulent blood.

Experiment No. 115.—Horse 360.—20-11-03, injected with 500 c.c. mixture sera of Horses S. H. 1, 160, 172, 147, Mules 150, 199, 201, Donkey 306 and the supposed anti-hæmolytic serum, 355, on one side and 1 c.c. virus, 369, on the othe rside.

Result.—Horse 360 died on 5-12-03 of hæmolysis.

Conclusion.—Serum, Mule 355, did not prevent the hæmolytic action of the different sera.

LXV.—*To note the effect of an injection of different fortified sera of horses, mules and donkeys, preceded* 24 *hours by an injection of a serum supposed to be anti-hæmolytic and* 1 *c.c. virus, subcutaneously.*

Remark.—Compare Experiment No. 115.

Experiment No. 116.—Horse 348.—19-11-03, injected 500 c.c. serum, 355 (anti-hæmolytic), subcutaneously. 20-11-03, injected with mixture 500 c.c. made up by equal parts of serum, Horse S. H. 1, 160, 172, 147, Mules 150, 199, 201 and Donkey 306 and 1 c.c. virus, 369.

Result.—Passed through a reaction, during which symptoms of hæmolysis were present. It did not acquire immunity. It died June 27th, 1904, from a spontaneous attack of horse-sickness.

Conclusion.—The injection of serum, Mule 355, 24 hours previously to the injection of hæmolytic sera, did not prevent hæmolysis. The reaction was not a horse-sickness reaction

LXVI.—*To note the effect of a simultaneous injection of different fortified sera of horses, mules and donkeys, with* 1 *c.c. virus, five days after an injection had been made with a supposed anti-hæmolytic serum.*

Remark.—Compare Experiment No. 115.

Experiment No. 117.—Horse 391.—27-11-03, injected with 500 c.c. serum, Mule 355, heated to 55° C. 2-12-03, injected with 500 c.c. of a mixture of the following sera: Horses S. H. 1, 147, 160, 172, Mules 150, 199, 201 and Donkey 306, and 1 c.c. virus, 369, simultaneously and subcutaneously.

Result.—Horse 391 died on 6-12-03, from hæmolysis.

Conclusion.—The injection of a supposed anti-hæmolytic serum, Mule 355, five days previous to the injection of hæmolytic serum mixture did not prevent hæmolysis.

LXVII.—*To note the effect of the injection of a mixture of different fortified sera of horses, mules and donkeys, simultaneously with* 1 c.c. *virus, subcutaneously.*

Remark.—Compare Experiment No. 115.

Experiment No. 118.—Horse 388.—10-12-03, injected with 500 c.c. of a mixture of serum, Horses S. H. 1. 160, 147, 172, Mules 199, 201, 150 and Donkey 306, and 1 c.c. virus, subcutaneously.

Result.—Horse 388 died 16-12-03 from hæmolysis.

LXVIII.—*To note the effect of a mixture of different fortified sera of horses, mules and donkeys, simultaneously with* 1 *c.c. virus, intrajugularly.*

Remark.—Compare Experiment No. 115.

Experiment No. 119.—Horse 392.—10-12-03, injected subcutaneously with 500 c.c. of a mixture of serum, Horse S. H. 1, 147, 160, 172, Mules 199, 201, 150 and Donkey 306, simultaneously with 1 c.c. virus, 369, intrajugularly.

Result.—Horse 392 died on 15-12-03, from hæmolysis.

Conclusion.—The simultaneous injection of a mixture of different fortified sera of horses, mules and a donkey proved to be hæmolytic.

LXIX.—*To note the effect of a simultaneous injection of fortified sera of different horses and virus.*

Experiment No. 120.—Horse 408.—10-12-03, injected with 500 c.c. of a mixture of serum, Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 369, intrajugularly.

Result.—Horse 408 died on 22-12-03 from horse-sickness.

Experiment No. 121.—Horse 402.—10-12-03, injected with 500 c.c. of a mixture of serum, Horses S. H. 1, 160 and 172, subcutaneously, and simultaneously 1 c.c. virus, 369, subcutaneously.

Result.—There was an apparently slight reaction, during which the morning temperature alone kept above normal; the evening temperature was always normal. The horse was tested on 7-12-03 on its immunity, and died from horse-sickness on 15-3-04.

Experiment No. 122.—Horse 399.—10-12-03, injected with 375 c.c. of a mixture of serum, Horses S. H. 1, 160 and 172 subcutaneously and 1 c.c. virus, 369, subcutaneously.

Result.—There was a distinct reaction in Horse 399, starting from the ninth day and lasting ten days; the temperature, however, never reached more than 103° C. in the evening. Horse 399 was tested, 9-4-04, with 5 c.c. virus, 447. It contracted horse-sickness, and died on 15-3-04.

Experiment No. 123.—Horse 396.—10-12-03, injected with 250 c.c. of a mixture of serum, Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 369, subcutaneously.

Result.—Died of debility on 23-12-03.

Conclusion.—The Horse 408 injected subcutaneously with serum and virus, simultaneously into the jugular vein, contracted horse-sickness and died. The animals injected with serum and virus, subcutaneously, passed through a slight reaction, but did not acquire immunity.

LXX.—*To note the effect of a simultaneous injection of fortified sera of different horses, mixed with ox serum, inoculated simultaneously with virus.*

Remark.—Compare Experiment No. 115.

Experiment No. 124.—Horse 359.—10-12-03, injected with 500 c.c. of a mixture of sera, Horses S. H. 1, 160, 172, and 500 c.c. ox serum, simultaneously with 1 c.c. virus, 369.

Result.—Horse 395 died from debility on 12-24-03.

Experiment No. 125.—Horse 406.—10-12-03, injected with a mixture of 375 c.c. serum of Horses S. H. 1, 160, 172, added to 500 c.c. ox serum and 1 c.c. virus, 369.

Result.—No reaction. Horse 406 was tested with virus on 25-6-04 (virus, Horse 385), and died of horse-sickness on 28-6-04.

Experiment No. 126.—Horse 405.—10-12-03, injected with 250 c.c. mixture of serum of Horses S. H. 1, 160, 172, added to 500 c.c. ox serum, simultaneously with 1 c.c. virus, 369.

Result.—Horse 405 died of debility on 23-12-03.

Conclusion.—The mixture of different fortified horse sera, mixed with ox serum, injected simultaneously with virus did not produce horse-sickness, nor resulted in immunity.

LXXI.—*To note the effect of a simultaneous injection of a mixture of different fortified horse sera, subcutaneously, with virus, also subcutaneously.*

Remark.—Compare Experiment No. 115.

Experiment No. 127.—Horse 393.—28-12-03, injected with 400 c.c. of a mixture of serum of Horses S. H. 1, 160, 172, subcutaneously, and simultaneously with 1 c.c. virus, 390.

Result.—No reaction. Not tested for immunity.

Experiment No. 128.—Horse 401.—28-12-03, injected with 300 c.c. mixture of sera of Horses S. H. 1, 160, 172, subcutaneously, and simultaneously with 1 c.c. virus, 390.

Result.—No reaction. Tested with 5 c.c. virus, 447, subcutaneously, on 7-3-04. Died of horse-sickness on 16-3-04.

Conclusion.—The subcutaneous simultaneous injection of a mixture of different fortified horse sera and virus did not produce the disease, nor did it produce immunity.

LXXII.—*To note the effect of a simultaneous injection of a mixture of different fortified horse sera injected into the jugular vein and virus injected subcutaneously.*

Remark.—Compare Experiment No. 115.

Experiment No. 129.—Horse 407.—28-12-03, injected with a mixture of 400 c.c. serum, Horses S. H. 1, 160, 172, into the jugular vein, and 1 c.c. virus, 390, subcutaneously.

Result.—Horse 407 showed no reaction. Not tested for immunity.

Experiment No. 130.—Horse 404.—28-12-03, injected with a mixture of 300 c.c. serum, Horses S. H. 1, 160 and 172 into the jugular vein, and 1 c.c. virus, 390, subcutaneously.

Result.—Horse 404 died from horse-sickness on 12-1-04, viz., 14 days later.

Conclusion.—The simultaneous injection of a mixture of different fortified horse sera into the jugular vein, with virus, subcutaneously, produced the disease in one instance. There is no proof that Horse 407 was immune previous to the injection.

LXXIII.—*To note the effect of a mixture of different fortified horse sera injected into the jugular vein, simultaneously with virus injected into the jugular vein.*

Remark.—Compare Experiment No. 115.

Experiment No. 131.—Horse 394.—28-12-03, injected with a mixture of 400 c.c. serum, Horses S. H. 1, 160 and 172, and 1 c.c. virus, 390, intrajugularly and simultaneously.

Result.—Horse 394 died on 13-1-04 from horse-sickness, viz., 16 days after the injection.

Conclusion.—The mixture of different fortified horse sera injected into the jugular vein simultaneously with virus did not prevent the disease.

LXXIV.—*To note the effect of a simultaneous subcutaneous injection of different fortified horse sera, together with a large quantity of virus.*

Remark.—Compare Experiment No. 115.

Experiment No. 132.—Horse 403.—28-12-03, injected subcutaneously with a mixture of 500 c.c. serum, Horses S. H. 1, 160, and 172 and 100 c.c. virus, 390, simultaneously.

Result.—Horse 403 died on the ninth day of January, 1904, viz., 12 days after the injection.

Conclusion.—The simultaneous injection of serum and a large quantity of virus produced horse-sickness.

LXXV.—*To note the effect of a simultaneous subcutaneous injection of different horse sera and virus into mules, virus in varying quantities.*

Remark.—Compare Experiment No. 115.

Experiment No. 133.—Mule 433.—11-1-04, injected with a mixture of 300 c.c. serum, Horses S. H. 1, 160 and 172, subcutaneously, and simultaneously with 1 c.c. virus, 390, also subcutaneously.

Result.—A reaction took place from 23-1-04 to 27-1-04. Mule 433 was tested with 5 c.c. virus, 447, on 7-3-04, viz., 56 days after the injection. This mule was sent to Nelspruit, and exposed to natural infection. It was still alive on 30-6-04.

Experiment No. 134.—Mule 431.—11-1-04, injected with a mixture of 300 c.c. serum, Horses S. H. 1, 160 and 172, and 10 c.c. virus, subcutaneously and simultaneously.

Result.—A slight reaction took place. Tested with virus, 447, on 7-3-04, viz., 56 days after the injection. Sent to Nelspruit, 11-4-04. Still alive on 30-6-04.

Experiment No. 135.—Mule 429.—11-1-04, injected with a mixture of 300 c.c. serum, Horses S. H. 1, 160, and 172, and 30 c.c. virus, 390, subcutaneously and simultaneously.

Result.—Reaction took place from the 18th to the 23rd January, 1904. Tested for immunity with 5 c.c. virus, 447, on 7-3-04. Sent to Nelspruit on 11-4-04. Still alive on 30-6-04.

Conclusion.—The subcutaneous simultaneous injection of different horse sera and different quantities of virus modified the horse-sickness reaction in mules.

LXXVI.—*To note the effect of the simultaneous intrajugular injection of different fortified horse sera and virus.*

Remark.—Compare Experiment No. 115.

Experiment No. 136.—Mule 430.—7-1-04, injected with a mixture of 300 c.c. serum, Horses S. H. 1, 160 and 172, intrajugularly, and 1 c.c. virus, 390, intrajugularly and simultaneously.

Result.—A reaction took place from the 20th to 27-1-04. Tested with 5 c.c. virus, 447, on 7-3-04. Sent to Nelspruit on 11-4-04. Still alive on 30-6-04.

Conclusion.—The simultaneous intrajugular injection of different fortified horse sera and virus modified the horse-sickness reaction in mules.

LXXVII.—*To note the effect of the subcutaneous injection of different fortified horse sera on mules which were injected previously with virus, subcutaneously.*

Remark.—Compare Experiment No. 115.

Experiment No. 137.—Mule 415.—12-1-04, injected with 1 c.c. virus, 390, and three hours later with a mixture of 300 c.c. serum of Horses S. H. 1, 160 and 172.

Result.—Reaction from the 19th to the 23rd. Tested on 7-3-04 with 5 c.c. virus, 447. This mule was later hyper-immunised.

Experiment No. 138.—Mule 432.—12-1-04, injected with 1 c.c. virus, 390, and six hours later, 300 c.c. mixture of serum, Horses S. H. 1, 160 and 172.

Result.—Reaction took place from the 20th to 27-1-04. Tested on 7-3-04 with 5 c.c. virus, 447. Exposed at Nelspruit on 11-4-04. Killed, owing to bad infection of epizootic lymphangitis, on 30-6-04.

Conclusion.—The subcutaneous injection of different horse sera into mules after the infection with virus was followed by a modified reaction of horse-sickness

LXXVIII.—*To note the effect of the injection of a mixture of different fortified horse and mule sera, virus injected simultaneously and subcutaneously.*

Remark.—Compare Experiment 115.

Experiment No. 139.—Horse 438.—1-2-04, injected with 400 c.c. mixture serum, Horses S. H. 1, 160 and 172, and Mules 150, 199 and 201, simultaneously with 30 c.c. virus, subcutaneously.

Remark.—Compare Experiment No. 115.

Result.—Horse 438 contracted hæmolysis and died, 12-1-04, with symptoms of horse-sickness.

Experiment No. 140.—Mule 246.—1-2-04, treated as Horse 438, with 300 c.c. serum.

Result.—No reaction. Tested on immunity with 5 c.c. virus on 7-3-04. Still alive on 30-6-04.

Conclusion.—The mixture of different fortified horse and mule sera produced hæmolysis in the horse but not in the mule.

LXXIX.—*To note the effect of a mixture of different fortified horse and mule sera, injected subcutaneously, and the virus intrajugularly.*

Remark.—Compare Experiment No. 115.

Experiment No. 141.—Horse 359.—On 1-2-04, injected with a mixture of 400 c.c. serum, Horses S. H. 1, 160 and 172, and Mules 150, 199 and 201.

Result.—Died of hæmolysis on 8-2-04. Symptoms of horse-sickness were also present.

Experiment No. 142.—Mule 424.—1-2-04, injected with a mixture of 300 c.c. serum, Horses S. H. 1, 160 and 172 and Mules 150, 199 and 201.

Result.—Died on 18-2-04 of hæmolysis.

Conclusion.—The mixture of different sera of horses and mules proved to be hæmolytic.

LXXX.—*To note the effect of the subcutaneous injection of a mixture of different horse and mule sera injected after the virus.*

Remark.—Compare Experiment No. 115.

Experiment No. 143.—Horse 435.—1-2-04, injected with 1 c.c. virus, 414, and six hours later, 400 c.c. serum, Horses S. H. 1, 160 and 172, and Mules 150, 199 and 201, subcutaneously.

Result.—Horse 435 died, 11-2-04, from the effect of hæmolysis. Symptoms of horse-sickness were also present.

Experiment No. 144.—Mule 409.—1-2-04, injected with 1 c.c. virus, 414, and six hours later, 300 c.c. serum, Horses S. H. 1, 160 and 172 and Mules 150, 199 and 201, subcutaneously.

Result.—Hæmoglobinuria and reaction. Tested on 7-3-04 with 5 c.c. virus, 447. Was killed owing to debility, 18-3-04.

Conclusion.—The injection of a mixture of different fortified sera of horse and mules, injected after virus, proved to be hæmolytic. The mule recovered from the disease.

LXXXI.—*To find the minimum quantity of serum required to modify the horse-sickness reaction in a mule when virus is injected into the jugular vein simultaneously and the serum subcutaneously.*

Remark.—Compare Experiment No. 115.

Experiment No. 145.—Mule 418.—15-2-04, injected with 400 c.c. of a mixture of different sera, Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 419, intrajugularly.

Result.—Reaction lasting from 22-2-04 to 1-3-04. Exposed in Nelspruit 11-4-04. Still alive on 30-6-04.

Experiment No. 146.—Mule 417.—15-2-04, injected with a mixture of 300 c.c. different sera, Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, intrajugularly.

Result.—Reaction took place from 22nd to 27th. Exposed in Nelspruit on 11-4-04. Still alive on 30-6-04.

Experiment No. 147.—Mule 413.—15-2-04, injected with a mixture of 200 c.c. different sera, Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 419, intrajugularly.

Result.—Reaction from 25-2-04 to 3-3-04. This animal was used for hyper-immunisation.

Conclusion.—200 c.c. of a mixture of different fortified horse sera were sufficient to modify the horse-sickness reaction in a mule.

LXXXII.—*To note the effect of the simultaneous injection of serum, subcutaneously, and virus, intrajugularly, followed by a second injection of serum during the incubation stage.*

Remark.—*Vide* Experiment No. 115.

Experiment No. 148.—Mule 449.—29-2-04, injected with a mixture, serum, Horses S. H. 1, 160 and 172, subcutaneously and 1 c.c. virus, 446, intrajugularly. 3-3-04, injected with 100 c.c. of the serum mixture.

Result.—Reaction. Was used for hyper-immunisation. Died on 25-4-04 from colic.

Experiment No. 149.—Mule 464.—29-2-04, injected with a mixture of serum, Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 446, intrajugularly. 3-3-04, injected with 100 c.c. serum mixture.

Result.—Reaction. Used for hyper-immunisation.

Experiment No. 150.—Mule 467.—29-2-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160 and 172, and 1 c.c. virus, 446, intrajugularly. 5-3-04, injected subcutaneously with 100 c.c. of a serum mixture.

Result.—Reaction. Exposed at Nelspruit on 11-4-04. Still alive on 30-6-04.

Experiment No. 151.—Mule 468.—29-2-04, injected with 300 c.c. of a mixture of Horses S. H. 1, 160 and 172, and 1 c.c. virus, 446, intrajugularly. 5-3-04, injected subcutaneously with 100 c.c. of a serum mixture.

Result.—Reaction. Dikkop. Used for hyper-immunisation.

Conclusion.—All mules recovered from horse-sickness. The second injection of serum modified the reaction.

LXXXIII.—*To note the effect of the simultaneous injection of* 300 *c.c. serum mixture, subcutaneously, and* 1 *c.c. virus, intrajugularly.*

Remark.—*Vide* Experiment No. 115.

Experiment No. 152.—Mule 540.—29-2-04, injected with 300 c.c. serum, mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus, 446, intrajugularly.

Result.—Reaction. Hæmolysis. Died on 10-3-04, with symptoms of horse-sickness.

Experiment No. 153.—Mule 451.—29-2-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus, 446, intrajugularly.

Result.—Long reaction. Hæmoglobinuria. Died on 19-3-04 from the effects of hæmolysis.

Experiment No. 154.—Mule 462.—29-2-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus, 446, intrajugularly.

Result.—Reaction. Symptoms of dikkop. This mule was later hyper-immunised.

Experiment No. 155.—Mule 463.—29-2-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus 446, intrajugularly.

Result.—Reaction. Symptoms of dikkop. This mule was later hyper-immunised.

Experiment No. 156.—Mule 465.—29-2-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus, 446, intrajugularly.

Result.—Reaction ; dikkop ; still alive ; not tested.

Experiment No. 157.—Mule 466.—29-2-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus 446, intrajuuglarly.

Result.—Reaction. Not tested.

Conclusion.—All mules had horse-sickness reactions. Those tested with pure virus proved to have acquired immunity. The serum was hæmolytic for two mules.

LXXXIV.—*To note the effect of a simultaneous injection of serum, subcutaneously, and a large quantity of virus, intrajugularly.*

Remark.—Compare Experimeut No. 115.

Experiment No. 158.—Mule 448.—1-3-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160, and 172, subcutaneously, and 5 c.c. virus, intra-jugularly.

Result.—No reaction. Proved to be immune ; was later hyperimmunised.

Experiment No. 159.—Mule 459.—1-3-04, injected with 300 c.c. of a serum mixture of Horses S. H. 1, 160 and 172, subcutaneously, and 5 c.c. virus, intrajugularly.

Result.—No reaction. Not tested

Conclusion.—Both animals were immune previous to the injection of virus and serum.

LXXXV.—*To note the effect of a mixture of fortified serum of horses hyper-immunised with horse virus and of horses hyper-immunised with mule virus, and of mules hyper-immunised with virulent mule blood, simultaneously with* 1 *c.c. virus, intrajugularly.*

Remark.—Compare Horses S. H. 1, 160, 172, with Experiment No. 115. Horse 264 hyper-immunised with virulent mule blood to 5,000 c.c. Horse 301 hyper-immunised with virulent mule blood to 5,000 c.c. Mule 320 hyper-immunised with virulent mule blood to 7,000 c.c.

Experiment No. 160.—Horse 346.—18-3-04, injected subcutaneously with 400 c.c. of a serum mixture of Horses S. H. 1, 160, 172, 264, 301, and Mule 320, and 1 c.c. virus, 447, intrajugularly.

Result.—Horse 346 died from hæmolysis, 26-3-04.

Experiment No. 161.—Horse 453.—18-3-04, injected with 400 c.c. of a serum mixture of Horses S. H. 1, 160, 172, 264, 301 and Mule 320, and 1 c.c. virus, 447, intrajugularly.

Result.—Died of hæmolysis, 22-3-04.

Experiment No. 162.—Horse 454.—18-3-04, injected with 400 c.c. of a serum mixture of Horses S. H. 1, 160, 172, 264, 301, and Mule 320, and 1 c.c. virus, 447, intrajugularly.

Result.—Died of hæmolysis, 25-3-04.

Conclusion.—The mixture of different fortified sera of horses and a mule proved to be hæmolytic.

LXXXVI.—*To note the effect of the simultaneous injection of serum, subcutaneously, 24 hours previously to a simultaneous injection of virus, intrajuguarly, and serum, subcutaneously.*

Remark.—Horses S. H. 1, 160 and 172. Compare Experiment No. 115.

Experiment No. 163.—Mule 353.—18-3-04, injected with 200 c.c. of a serum mixture of Horses S. H. 1, 160 and 172. 19-3-04, injected with 200 c.c. of a serum mixture and 5 c.c. virus, 447, intrajugularly. 21-3-04, injected with 200 c.c. of a serum mixture, intrajugularly.

Result.—Died of hæmolysis, 20-4-04.

Experiment No. 164.—Mule 469.—18-3-04, injected with 200 c.c. serum mixture, S. H. 1, 160 and 172. 19-3-04, injected subcutaneously with 200 c.c. of a serum mixture and 5 c.c. virus, 447, intrajugularly.

Result.—No reaction. Hyper-immunised.

Conclusion.—The mixture of sera proved to be hæmolytic for one mule. The other mule was immune previous to the injection.

LXXXVII.—*To note the effect of a mixture of different quantities of fortified horse sera, simultaneously injected with virus, intrajugularly, and after an interval of three days with serum alone.*

Remark.—Compare Experiment No. 115.

Experiment No. 165.—Mule 422.—5-4-04, injected with a mixture of 100 c.c. serum of Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 447, intrajugularly. 8-4-04, injected with 100 c.c. serum mixture.

Result.—Mule 422 died, 14-4-04, from horse-sickness.

Experiment No. 166.—Mule 480.—5-4-04, injected with a mixture of 200 c.c. serum of Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 447, intrajugularly. 8-4-04, injected with 100 c.c. serum mixtur.

Result.—Mule 480 died during night 11-12-4-04. It had a spontaneous infection of horse-sickness prior to inoculation.

Experiment No. 167.—Mule 485.—5-4-04, injected with a mixture of 300 c.c. serum of Horses S. H. 1, 160 and 172, subcutaneously, and 1 c.c. virus, 447, intrajugularly. 8-4-04, injected with 100 c.c. serum.

Result.—Reaction of horse-sickness. Not tested.

Conclusion.—100 c.c. serum is not sufficient to modify the horse-sickness reaction in mules.

LXXXVIII.—*To note the effect of a simultaneous injection of different quantities of serum, subcutaneously, and virus, intrajugularly, followed by an injection of serum three days later. The serum mixture was heated to 55° C. for half an hour.*

Remark.—*Vide* Experiment No. 115.

Experiment No. 168.—Mule 427.—5-4-04, injected with 100 c.c. serum of Horses S. H. 1, 160, 172, and 1 c.c. virus, 447, intrajugularly. Serum heated at 55° C. 8-4-04, injected with 100 c.c. serum.

Result.—Died 17–4–04 from horse-sickness.

Experiment No. 169.—Mule 483.—5–4–04, injected with 200 c.c. serum mixture of Horses S. H. 1, 160, 172, and 1 c.c. virus 647, intrajugularly. Serum heated to 55° C. 8–4–04, injected with 100 c.c. serum mixture heated

Result.—No reaction. Not tested.

Experiment No. 170.—Mule 482.—5–4–04, injected with 300 c.c. serum of Horses S. H. 1, 160 and 172, and 1 c.c. virus, 447, intrajugularly. Serum heated. 8–4–04, reinjected with 100 c.c. serum.

Result.—Reaction; dikkop. Used for hyper-immunisation.

Conclusion.—Compare LXXXVII. 100 c.c. of a heated serum mixture was not sufficient to modify the horse-sickness reaction in a mule.

LXXXIX.—*To note the effect of different fortified horse sera injected separately and simultaneously with virus, intrajugularly, and followed by serum.*

Remark.—*Vide* Experiment No. 115.

Experiment No. 171.—Mule 484.—5–4–04, injected with 100 c.c. serum, Horses S. H. 1, 160 and 172, each lot separately, and 1 c.c. virus, 447, intrajugularly. 8–5–04, reinjected with 100 c.c. serum.

Result.—Died of horse-sickness, 17–4–04.

Experiment No. 172.—Mule 486.—5–4–04, injected with 200 c.c. serum, Horses S. H. 1, 160 and 172, each lot separately, and 1 c.c. virus, 447, intrajugularly. 8–5–04, reinjected with 100 c.c. serum.

Result.—Reaction. Not tested.

Experiment No. 173.—Mule 479.—5–4–04, injected with 300 c.c. serum, Horses S. H. 1, 160 and 172, each lot separately, and 1 c.c. virus, 447, intrajugularly. 8–5–04, reinjected with 100 c.c. serum.

Result.—No reaction. Used for hyper-immunisation.

Conclusion.—100 c.c. of different sera, each injected separately, did not modify the horse-sickness reaction in a mule.

General Conclusions.

1. The serum of immune animals which were not hyper-immunised has no preventive action.
2. The serum of hyper-immunised animals has a preventive action:—
   (*a*) When mixed with virus;
   (*b*) When injected before virus;
   (*c*) When injected in large quantities simultaneously with virus
3. The serum of one animal exclusively has a less preventive action than the mixture of several sera.
4. The sera of hyper-immunised animals proved to be more hæmolytic in horses than in mules.
5. The simultaneous injection of serum and virus modifies, in the majority of cases, the horse-sickness reaction in mules.
6. Of the different experiments which are worthy of consideration as a method of immunising mules against horse-sickness, namely, Nos. 133, 134, 135, 136, 145, 146, 147, 148, 149, 150, 152, 153, 154, 155, 156, 157, 158, 159, 163, 164, 166, 167, 169, 170, 172 and 173. That is to say, 69·2 per cent. of mules recovered from horse-sickness; 19·33 per cent. proved to be immune, and 11·54 per cent. died of hæmolysis.
7. The simultaneous injection of virus, intrajugularly, and serum, subcutaneously, followed by an injection of serum within the incubation period, produced the most satisfactory results.
8. All mules which recovered from the horse-sickness reaction proved to be immune when tested with virus, or when exposed to natural infection.

Notes on Hæmolysis.

I.

The experiments with serum for a preventive treatment of horse-sickness resulted in the death of several animals. This was attributed to hæmolysis. The serum experiments (*vide* "The Serum Treatment applied to Horse-sickness," page 1: 2) give an insight into the difficulties which we have experienced in the course of our work. It now remains to consider their results purely from the standpoint of the hæmolysis. This is of the utmost importance, since the occurrence of hæmolysis debars us from introducing the inoculation method of mules against horse-sickness—a difficulty which, so far, we have not yet been able to overcome.

Under the term hæmolysis, we understand the effusion of the hæmoglobine from the red corpuscle. This is due to the death of the red corpuscle, when the hæmoglobine escapes from the cell and dissolves in the surrounding liquid. All substances which cause the death of the erythrocytes and, thereby, the effusion of hæmoglobine into the serum, are now commonly termed "hæmolysines," and are ranged under the class of blood poisons. A good many substances are known to act in the way described, particularly the hæmolysines which are produced by the blood of an animal belonging to one species injected into an animal of a different species. The serum of the injected animal will dissolve the blood cells in vitro, of such animals from which the blood for the injection was derived. And, when this hæmolytic serum is injected into the susceptible animal, toxic symptoms and death are usually observed.

Ehrlich and Morgenroth have also shown that it is possible to produce a hæmolytic serum in individuals belonging to a certain species, after injection of blood of animals of the same species. The above authorities gave the name of "Isolysines" to these hæmolysines, to distinguish them from the "Heterolysines," which, as already shown, are produced by the injection of blood of a foreign species.

II.

The production of our preventive serum for horse-sickness is based on the same principle employed in the production of preventive rinderpest serum. Large quantities of freshly-drawn and defibrinated blood were injected subcutaneously and, in some cases, intrajugularly. In an extensive experience with rinderpest serum, I have never come across a hæmolytic serum, viz., an "Isolysine," which had a toxic effect or caused the death of an ox. Ehrlich and Morgenroth state that, in order to obtain an isolytic serum, they first dissolved the blood corpuscles used for the injections. In my own investigations, defibrinated blood only was used. Basing these experiments on Ehrlich's lateral chain theory, it was deemed advisable to hyper-immunise the horse, the mule and the donkey, since these animals are susceptible to horse-sickness. The animals were hyper-immunised as follows:—

1. Horses injected with virulent horse blood (S.H. 1, 160, 172, 147).
2. Horses injected with virulent mule blood (264, 301).
3. Mules injected with virulent horse blood (150, 199, 201).
4. Mules injected with virulent mule blood (320).
5. Asses injected with virulent horse blood (306).

III.

The experiments to ascertain the preventive value of the serum of a hyper-immunised horse, were begun with the serum of a horse, which was periodically injected with virulent horse blood (Horse S.H. 1). The number of experiments in which this blood was exclusively used, amounted to 93 horses and mules. In no instance were symptoms of hæmolysis observed. It was naturally concluded that the serum in question was not hæmolytic. The serum of a second horse (160), hyper-immunised in the same way, was used in four experiments and, again, no hæmolysis was observed. The serum of a third horse (147) was used in six experiments (91, 92, 93, 94, 95, 96). Here it should be stated that this horse was once injected with defibrinated blood into its jugular vein, whereas the other two were not. I do not attach any importance to this intrajugular injection. Five horses and one mule were used in these six experiments. The result was that four horses showed the symptoms of hæmolysis and died. One horse and the mule were not affected by the serum.

I am not prepared to generalise from this single experiment with regard to mules, but I do believe that a deduction for horses, at least, is permissible; since out of five horses, only one was not affected by the serum. This conclusion then, is that a hæmolytic serum does not prove toxic for every horse. This is in accordance with the experience of Ehrlich and Morgenroth, who found that an isolysine does not act, *in vitro*, on the blood cells of various animals of the same species in the same way. The above experiments with serum of the different three horses also show that some horses do not produce a hæmolytic serum, as for instance, Horse S.H. 1. The question arises:— Was the hæmolytic action of the serum of Horse 147 due to the intrajugular injection of defibrinated blood? For my own part, I do not think so. Koch has lately published his reports on horse-sickness. He hyper-immunised his horses by intrajugular injection with defibrinated blood, and did not observe a hæmolytic serum.

IV.

After it was ascertained that a mixture of the various horse sera had a markedly preventive effect on mules, experiments were continued along this line. Forty-one animals, viz., 32 mules and nine horses, were injected with a mixture of the three horse sera (namely, S.H. 1, 160, and 172). Three mules died from hæmolysis. All these horses were hyper-immunised with virulent horse blood. It is impossible to tell whether the hæmolysis was due to one serum, which alone had hæmolytic action, or whether the combination of the three sera led to this action. From these results it is plain that a preventive serum produced in horses by the injection of virulent horse blood, may prove hæmolytic for some mules.

V.

Experiments were also made with a mixture of the three horse sera, which, as shown above, did not prove hæmolytic for horses, together with three mule sera (Nos. 150, 199, and 201). These mules had been hyper-immunised with horse blood. Eight animals were injected, viz., four horses and four mules. The result was that all four horses died of hæmolysis. Of the mules, one died, one recovered from hæmolysis, and the serum had no effect on the other two. Considering that the horse serum mixture was not hæmolytic for horses, it may be concluded that the mule serum was hæmolytic for horses. With regard to the mules, no definite conclusion is possible, since the hæmolysis might have been due to the horse serum, the mule serum, or possibly, to both.

VI.

Still guided by the same reflections as in the former experiments, the serum of the three horses was used mixed with the serum of two horses and a mule, hyper-immunised with virulent mule blood. The result was, that all three horses which were injected with this mixture died of hæmolysis. If we consider, as hitherto, that the mixture of the horse sera was harmless for horses, then the hæmolysis must have been due to the serum of horses hyper-immunised with mule blood, or to the mule serum hyper-immunised with mule blood. And we should expect that the serum of horses and mules, hyper-immunised with mule blood, should not become hæmolytic for horses, or only to a limited extent. The conclusion would be, therefore, that hæmolysines for horse blood are produced in horses injected with mule blood, and in mules injected with mule blood.

VII.

The mixture of serum of a hyper-immunised horse, a mule and an ass, all injected with horse blood, was also tested. The horse died of hæmolysis, the mule contracted hæmolysis, but recovered. In this instance, the hæmolysis in the horse might possibly have been expected, and in a lesser degree, also in the mule. Thus it may be said that the hyper-immunisation of animals with horse blood belonging to the two different varieties of the genus equi and of their bastards, produces a serum which is hæmolytic, not only for the horse, but also for the mule, at any rate, when these sera are mixed.

VIII.

Having noticed that the mixture of the three non-hæmolytic sera has but a slight preventive effect in horses against horse-sickness, it was thought advisable to experiment with the serum of animals of a different species, in conjunction with the immune serum. For this purpose, four horses were treated (compare Experiments 113, 124, 125, and 126). The result was, that one of the four horses succumbed with lesions of hæmolysis. Therefore, the addition of a serum of an ox, added to a mixture of non-hæmolytic serum, seems to cause hæmolytic properties. Of course, it is quite probable that the ox serum alone possesses hæmolytic properties, with reference to horse blood.

IX.

In one of the experiments (Mule 355, Experiment 108) one mule showed lesions of hæmolysis and horse-sickness, from which it recovered. It might reasonably be expected—taking the lateral chain theory into consideration—that the serum of this mule had acquired anti-hæmolytic properties. Accordingly, it was considered desirable to test the serum with regard to its anti-hæmolytic effect. The experiments were made as follows :—

1. Injection of a mixture of hæmolytic sera and supposed anti-hæmolytic serum.
2. Injection of supposed anti-hæmolytic serum, 24 hours previously to the injection of hæmolytic sera.
3. Injection of supposed anti-hæmolytic serum, five days previous to the injection of hæmolytic serum.

Result.—Ad. 1. The three horses and one mule which were injected with a mixture of hæmolytic and supposed anti-hæmolytic sera, contracted hæmolysis. Only one recovered. (Compare Experiments 111, 112, 115, and 116.)

Ad. 2 and 3. Both horses which were treated in this way died of hæmolysis.

Conclusion.—The serum of the mule, which had recovered from an attack of hæmolysis, had no anti-hæmolytic effect in the above quoted experiments.

X.

Before proceeding to summarize these experiments, I wish to make it plainly understood that these experiments were not undertaken for the precise purpose of ascertaining the hæmolytic effect of the various sera produced by blood injection of horses and mules into various susceptible animals.

The conclusion from the hæmolytic coincidences would be as follows :—

1. The hyper-immunisation of horses with defibrinated horse blood tends to produce a serum which may be hæmolytic for horses and mules (Horse 147).
2. The hyper-immunisation of horses with mule blood tends to produce a serum which may be hæmolytic for horses and mules (264 and 301).
3. The hyper-immunisation of mules with horse blood tends to produce a serum which may be hæmolytic for horses and mules (150, 199, and 201).
4. The hyper-immunisation of mules with mule blood tends to produce a serum which may be hæmolytic for horses and mules (320).
5. The hyper-immunisation of donkeys with horse blood tends to produce a serum which may be hæmolytic for horses and mules (306).
6. There seems to be a certain definite relation between the hæmolytic effect of a mixture of different sera and the number of the several sera contained in the mixture.
7. The addition of ox serum to a non-hæmolytic mixture of immune horse sera may prove to be hæmolytic.
8. The serum of a mule which passes through an attack of hæmolysis, did acquire anti-hæmolytic properties.

Morbid Lesions found in Animals which Succumbed to the Effect of a Hæmolytic Serum.

Horse 296.—Injected on 24-6-03 with serum of Horse 147 (compare Experiment 92).

27-6-03.—Hæmoglobinuria.

28-6-03.—Hæmoglobinuria.

29-6-03.—Hæmoglobinuria ; urine slightly clearer.

30-6-03.—Hæmoglobinuria ; the urine contained a few epithelial cells.

Horse 296 died on the evening of July 1st, 1903. *Post-mortem* was made on the morning of the 2nd July, 1903. Rigor mortis was present. The condition was good. The blood was rather watery. The symptoms of a general icterus (jaundice) were present. The lungs were slightly œdematous. The heart muscle was very pale. There was a black clot in the left ventricle and a white and black clot, well differentiated, in the right ventricle. There were a few hemorrhages on the endocard of the left ventricle. The liver was slightly jaundiced ; the bile ducts were filled with bile. The spleen was slightly enlarged and its pulpa was somewhat softened. The kidneys were enlarged, but had rather a pale colour. The mucous membrane of the stomach had a normal appearance; that of the duodenum was bile-stained, thickened and showed

the so-called zebra stripes. The same condition was found in the jejunum and ilium. The mucosa of the cœcum and colon was uniformly slate-coloured The microscopical examination of smears of all organs proved the absence of endoglobular parasites

Horse 297.—Injected on July 6th, 1903, with serum, Horse 147 (compare Experiment 94). This animal showed similar symptoms as those described in Horse 298. It was better the next day (7–7–03), but would not eat. On July 8th, 1903, hæmoglobinuria was noticed for the first time. The mucous membranes were yellow. There was loss of condition. The animal laid down almost the whole of the 9th July, 1903.

Horse 297 died on July 9th, 1903. *Post-mortem* was made one hour and a half after death. The condition of the animal was good. Rigor mortis was present, There was a general jaundiced condition of the flesh and the serous membranes. The blood was coagulated and separated into black and white clots; the plasmatic clot had a somewhat brownish hue. The heart-bag was filled with dark yellow liquid. The heart muscle was pale. There were no echymosæ on the endocard. The liver was dark green in colour. The spleen was enormously enlarged, its weight being 12 pounds; the pulpa was soft. The kidneys were uniformly dark red, with hæmorrhagic infarcts in the cortex. The urine was black. The mucous membrane of the stomach was covered with blackish mucous (coagulated blood). The intestines were normal.

Horse 298.—Injected on 6th July, 1903, with serum, Horse 147 (compare, Experiment 96). The injection was made into the lower part of the jugular vein. Soon after the injection the horse lost control of the hind quarters, staggered for a few minutes, and dropped suddenly; neighing repeatedly. The pulse was rapid; the respiration was accelerated. After a lapse of a few minutes, the horse rose again, it now kept the head stretched straight out; it breathed quickly with wide-opened nostrils. In the afternoon of the same day, the horse went down again. It showed tremor of the muscles; it moaned and had a rapid respiration. The next day (7–7–03) Horse 298 seemed to be better. On the 8th July, 1903, Horse 298 lay down again. The mucous membranes had a yellowish colour. The horse tried to stale repeatedly. Red urine was voided. The animal lost condition from day to day until death.

Horse 298 died at one o'clock p.m., on July 10th, 1903. *Post-mortem* was made one hour later. The condition of the cadaver was fair. There was a general jaundiced condition of the flesh, the serous membranes and all organs. The lungs were œdematous and a few hemorrhagic infarcts were present. The blood of the heart ventricles was not completely coagulated. A few hemorrhagic spots were noticed on the endocard of the left ventricle. The heart muscle was pale. There was but little liquid in the heart-bag. The spleen was slightly enlarged, there were several hæmatoms in the tissue of the spleen. The pulpa was black. A white thrombus was found in the splenic vein. The liver was enormously enlarged. The tissue was friable, of a green-yellow-reddish colour; it was studded with white spots, which on close examination, proved to be small abscesses; minute hæmorrhagic spots were also present. The kidneys were enormously enlarged, almost spherical. The capsula was infiltrated. The tissue of the kidneys was black, œdematous and friable. The mucosa of the stomach was uniformly reddened and swollen; that of the duodenum was also thickened, and red stripes across the mucosa were present. The mucous membrane of the cœcum and colon were slate-coloured. The bladder was empty. The lymphatic glands were not enlarged.

Microscopical examination of smears of the different organs proved the absence of endoglobular parasites. The small abscesses in the liver contained a bipolar bacterium.

Horse 317.—Inoculated on 6th July, 1903, with serum, Horse 147 (compare Experiment 93). The injection was followed by the same alarming symptoms as in Horses 297 and 298. The animal was found on the ground on the following day. It tried to rise, but was unable to stand up. On the 8th July, 1903, the horse was able to get up, but its gait was staggering; tremor of the muscles was noticed. Yellow mucous membranes and red urine were observed, which were present up to death. There was a daily increasing loss of condition.

Horse 317 died at 3.30 p.m., on the 12th July, 1903. *Post-mortem* was made one hour later. The condition was fair. Rigor mortis was not yet present. The blood was not well coagulated and had a brownish hue. There was a general jaundiced condition of the whole body. The lungs were slightly œdematous. The pericard contained a brownish-reddish liquid. The myocard was very pale. The ventricles contained but a little badly coagulated blood. There were no hemorrhagic spots on either endocard. The spleen was enormously enlarged; its weight was 13½ pounds. Its surface was very irregular, due to several hæmatoma in the pulpa. The lymphatic glands of this organ were but slightly enlarged. The liver was a little congested and jaundiced. The kidneys were much enlarged, rather friable; and the cortex showed several red stripes (red infarcts). The urine was black.

The mucous membrane of the stomach showed superficial hemorrhages. The mucosa of the duodenum and jejunum was bile-stained, slightly thickened, and the serosa was spotted with punctiform hæmorrhages. The mucosa of the cœcum and colon was slightly reddened.

Horse 349.—Injected on the 10th October, 1903, with a mixture of different sera (compare Experiment 107). On the 14th October, 1903, it was noticed that the horse had difficulty in passing urine, which was of a red colour. The mucous membrane of the eyes had a yellow colour, and there were superficial hæmorrhagic spots. On the 15th October, 1903, the animal refused food. A blood count was now made and the number of red corpuscles amounted to 3,645,000 per m.m. ^{3}. Two days previous to death, there was almost complete anuria, only a little bloody urine was passed at a time.

Horse 349 died during the night of the 15–16th October, 1903. *Post-mortem* was made in the morning. Rigor mortis was present. The condition was poor. The flesh had a brownish-yellow colour. The blood was coagulated in red and white clots. The serum had a red colour. The lungs were in a stage of œdema, of a brown-red colour, and contained several superficial infarcts; some of which reached the size of a small apple. The pericard contained a small quantity of red liquid. The heart was in diastyle; the heart muscle had a colour resembling sepia. Both ventricles were filled with coagulated blood; there was a black clot in the left ventricle, and a mixed black and white layer alternating in the right ventricle. The liver was greenish-yellow, very friable; the bile ducts contained bile. The spleen was normal in size, its pulpa was firm, its colour somewhat darker than normal. One of the kidneys weighed 4 lbs. 3½ ozs. Both kidneys were much enlarged; the capsula could not easily be stripped off. The section was uniformly dark red, intermixed with still darker stripes, and a reddish mucous could be scraped off with the knife. The urinary bladder was empty. The mucous membranes of all parts of the intestines were discoloured and black, and were uniformly swollen.

Horse 331.—Injected on 5th November, 1903, with a mixture of different sera (compare Experiment 109). The horse was found on the ground the following day. The mucous membrane of the eye had a yellowish tinge. A blood count was made, and the number of red corpuscles was found to be 2.430,000 per m.m.^{3}. On the 7th November, 1903, the horse was noticed to

pass red urine, which contained red blood corpuscles. The animal did not eat well. The count of red blood cells amounted to 2.437,000. On the 8th November, 1903, the horse had symptoms of colic. The blood count resulted in 2.660,000 per m.m.^{3}.

Horse 331 died during the night of the 8th–9th November, 1903. There was a blood-coloured liquid in the heart-bag. The flesh had a reddish-brown colour. The heart muscle was pale; both ventricles contained blackish clots, which were covered with a fine layer of a bright florescent green colour. The kidneys were black, and a bloody liquid exudated from sections of that organ. The liver was green. The spleen was normal. The mucous membrane of the stomach was normal; that of the smaller intestines was slate-coloured.

Horse 378.—Injected on the 13th November, 1903, with a mixture of several sera (compare Experiment 111). The urine of this horse was clear the following day; it contained 0·5 per cent. albumen, tested according to Esbach. The blood count resulted in 5,620,000 corpuscles per m.m.^{3}. On the 15th November, 1903, red urine was present. Blood count, 2.066,000 red corpuscles per m.m.^{3}. On the 17th November, 1903, the blood count was 2,240,000 per m.m.^{3}. The urine was somewhat clearer on this date.

Horse 378 died during the night of the 17th–18th November, 1903. Rigor mortis was present. Condition poor. The flesh had a brownish-yellow colour. The blood was not completely coagulated. The epeueurosæ had a greenish-yellow colour. The lymphatic glands were enlarged and infiltrated with blood. The lungs were œdematous. The heart muscle was pale. The right ventricle was blood-stained; there was a small clot in the left one, the endocard of which was pale. The pericard was empty. The liver showed the character of a nutmeg liver and was jaundiced; the bile ducts contained much bile and were friable. The spleen weighed 7 lbs., 10 oz.; the pulpa had the consistency of black currant jam; its lymphatic glands were enlarged. The kidneys were of normal size, uniformly reddened. The urinary bladder was empty. The mucosa of the stomach was normal; that of the duodenum and jejunum was bile-stained, reddened in patches and covered with a viscid mucous. The cœcum and colon were uniformly slate coloured.

Mule 356.—Injected on the 13th November, 1903, with a mixture of different sera (compare Experiment 112).

14–11–03,	Blood count,	6·100,000.	
15–11–03,	,,	2·640,000	Red urine.
16–11–03,	,,	2·560,000	,,
17–11–03,	,,	3·366,000	,,
18–11–03,	,,	4·560,000	,,
19–11–03,	,,	4 860,000	,,
20–11–03,	,,	4 246,000	,,
21–11–03,	,,	4 100,000	,,

Mule 356 died on the 21st November, 1903. This animal suddenly showed an alarming dyspnœ, accompanied by a whistling and trumpeting sound. It died within an hour after these symptoms had started. *Post-mortem* was made about half an hour after death. The condition was good. There was a general jaundice present. The flesh had a brownish colour. The lungs were retracted and normal. The heart was in systole, containing little blood, which was not yet coagulated. There were no hemorrhages on the endocard. The liver was slightly jaundiced. The spleen was normal. The kidneys were uniformly reddened with numerous wedge-shaped red infarcts. The urine was red. The mucous membrane of the stomach was normal; the mucosa of the duodenum and jejunum was bile-stained and swollen. That of the cœcum and colon was reddened and also swollen.

Mule 355.—Injected on the 10th October, 1903, with a mixture of several sera (compare Experiment 108). On the 14th October, 1903, red urine and yellow mucous membranes were noticed. On the 15th, a blood count was made and the red corpuscles amounted to 2·437,000. 16–10–03; the urine was clearing up in the morning; blood count, 2·502,000.

17–10–03, Urine again red. Animal feeding.

18–10–03, Blood count, 2·870,000.

19–10–03, No urine was passed since the previous day. A little urine was collected in the morning which contained numerous red corpuscles; Blood count, 2·130,000.

20–10–03, Blood count, 2·157,000.

21–10–03, Blood count, 2·157,000. Poikylocytosis was pronounced.

22–10–03, Urine was still bloody. Count, 1·940,000.

23–10–03, Urine began to clear up from the morning of the previous day; it was now clear, but had a greenish colour. Blood count, 2.097,000.

24–10–03,	Blood count,	2·047,000.
25–10–03,	,,	2·335,000.
26–10–03,	,,	2·470,000.
27–10–03,	,,	2·517,000.
28–10–03,	,,	3·207,000.
29–10–03,	,,	3·180,000.
30–10–03,	,,	3·370,000.
31–10–03,	,,	3·350,000.
1–11–03,	,,	4·150,000.
2–11–03,	,,	4·050,000.
3–11–03,	,,	4·853,000.
4–11–03,	,,	4·870,000.
5–11–03,	,,	5·042,000.
6–11–03,	,,	5·604,000.
7–11–03,	,,	5·390,000.
8–11–03,	,,	4·640,000.
9 11–03,	,,	4·794,000.
10–11–03,	,,	4·082,000.

Mule 386 (may serve as a control; did not show red urine). Injected with a mixture of different sera (compare Experiment 110). Hæmolysis had to be anticipated, but did not take place. Blood count was as follows:—

6–11–03,	Blood count,	6·246,000	per m.m.^{3}
7–11–03,	,,	4·340,000	,,
8–11–03,	,,	5·460,000	,,
9–11–03,	,,	5·512,000	,,
10–11–03,	,,	6·223,000	,,
11–11–03,	,,	4·800,000	,,
12–11–03,	,,	5·400,000	,,
13–11–03,	,,	5·612,000	,,
14–11–03,	,,	5·620,000	,,

The serum had, undoubtedly, a slight effect on the blood corpuscles of the mule, since the number of corpuscles decreased after the injection.

Horse 348.—Injected on the 20th November, 1903, with a mixture of different sera (compare Experiment 116). The count of red blood corpuscles was as follows :—

19–11–03, Blood count, 6·020,000 per m.m.^{3}
20–11–03, ,, 6·332,000.
21–11–03, ,, 4·706,000.
22–11–03, ,, 3·820,000.

There was a distinct jaundiced condition of the mucous membrane of the eyes, on which several blood spots were also noticed. Red urine was voided.

23–11–03, Blood count, 3·006,000 per m.m.^{3}
24–11–03, ,, 3·506,000.
25–11–03, ,, 3·966,000.
26–11–03, ,, 3·392,000.
27–11–03, ,, 4·632,000.

Urine was again clear; Esbach's albumen test gave a strong precipitation.

28–11–03, Blood count, 4·126,000.
29–11–03, ,, 4·612,000.

Albumen was less in the urine than on 27–11–03.

30–11–03, Blood count, 5·552,000.
1–12–03, ,, 4·320,000.

The urine was again red and contained a large amount of albumen. The horse had symptoms of colic; it lay down at intervals.

2–12–03, Blood count, 3·906,000 per m.m.^{3}
3–12–03, ,, 4·846,000.
4–12–03, ,, 5·066,000.
5–12–03, ,, 5·152,000.

Urine was again clear from the 3rd December, 1903.

6–12–03, Blood count, 5·050,000. This animal recovered.

Horse 360.—Injected on the 20th November, 1903, with a mixture of different sera (compare Experiment 115).

19–11–03, Count of red corpuscles, 7·650,000 per m.m.^{3}
20–11–03, ,, ,, 7·416,000.
21–11–03, ,, ,, 7·320,000.
22–11–03, ,, ,, 6·826,000.
23–11–03, ,, ,, 6·612,000.

A slight jaundiced condition of the mucous membrane of the eyes was noticeable.

24–11–03, Count of red corpuscles, 6·494,000.
25–11–03, ,, ,, 6·850,000.

Red urine was noticed since the day previous.

26–11–03, Count of red corpuscles, 7.080,000.

Urine had cleared up since previous evening, but still contained albumen.

27–11–03, Count of red corpuscles, 5·646,000.

Urine still clear.

28–11–03, ,, ,, 5·906,000.

The urine of the previous evening was red; of this date (morning) it was dark brown, containing much albumen.

29–11–03, Count of red corpuscles, 7·060,000.

The urine of the previous evening was again red.

30–11–03, Count of red corpuscles, 7·080,000.

The urine was clear and contained but little albumen.

1–12–03,	Count of red corpuscles,		6·352,000.
3–12–03,	,,	,,	6·494,000.
4–12–03,	,,	,,	6.180,000.
5–12–03,	,,	,,	5.086,000.

The horse died on the 5th December, 1903.

Post-mortem.—The animal was in a poor condition. The lungs were strongly œdematous (pulmonary form of horse-sickness). There were petechiæ on the endocard of the left ventricle. The liver was slightly enlarged, but fairly firm. The capsula of the kidneys was slightly adherent; the kidneys were dark, congested, and numerous dark infarcts were seen running deep into the cortex. There was a patchy congestion of the pyloric mucous membrane. The mucosa of the small intestines was slightly thickened and bile-stained. A patchy congestion was marked on the mucosa of the large intestines. The urine was of darkish amber colour; no blood was present.

Horse 391.—Injected on the 2nd December, 1903, with a mixture of different sera (compare Experiment 117).

2–12–03,	Count of red corpuscles,		5.703,000.
3–12–03,	,,	,,	3.672,000.
4–12–03,	,,	,,	3.406,000.
5–12–03,	,,	,,	3.460,000.
6–12–03,	,,	,,	3.390,000.

Horse 391 died of hæmolysis. No *post-mortem* recorded.

I have the honour to be,

Sir,

Your obedient servant,

A. THEILER,

Government Veterinary Bacteriologist.