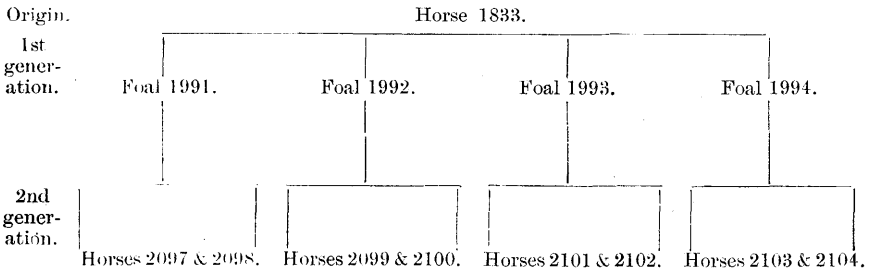


“H.”—CONTINUATION OF EXPERIMENTS FOR INOCULATION AGAINST EQUINE PIROPLASMOSIS.

In my last Annual Report a number of experiments were enumerated showing (1) that the inoculation of mules with immune mule blood can be performed with a large prospect of success; (2) that a certain amount of risk is attached to the inoculation of donkeys with immune mule blood; and (3) that the inoculation of horses with immune donkey blood may be followed by disastrous results. During the past year these experiments have been continued on a somewhat different line, and based on the observation made in connection with redwater, namely, that the inoculation of cattle with blood of a calf immune against this disease is not so frequently followed by strong reactions and mortality as when the blood is derived from a full-grown beast.

Accordingly, I decided to utilise the blood (*a*) from young immune weaned horse foals, and (*b*) from immune donkey foals which are still suckling. The experiments were classified according to the origin of the blood, namely, (1) from an immune horse, (2) from an immune mule, and (3) from an immune donkey. At the commencement of each of the three classes of experiments I attach the genealogical table of the animals whose blood was utilised for the inoculation.

I.—INOCULATION AGAINST EQUINE PIROPLASMOSIS BY MEANS OF HORSE FOAL BLOOD—ORIGIN HORSE BLOOD.



1.—INOCULATION BY MEANS OF HORSE FOAL BLOOD—ORIGIN HORSE BLOOD.

*Horse 1833.*—Argentine, aged (compare Annual Report, 1905-6 page 94). This horse had been injected with blood of mule 589—immune against *piroplasma equi*—and showed two reactions accompanied by *piroplasma equi*, but in rare numbers.

*Transvaal Foals* injected with immune horse blood.—Transvaal foals Nos. 1991, 1992, 1993 and 1994, all six months, and obtained from the S.A.C., were injected on the 25th May, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of horse 1833.

EXPERIMENT I.

1. *Foal 1991.*— *First Generation.*

*Injected* as above.

Irregular reaction, commencing on the 8th day and continuing for 18 days. On the 27th day—the morning temperature being 101.8—a sharp reaction was noticed for five days, the

maximum temperature being 105 F. on the evening of the 28th, and falling to 101 at the conclusion. The ring form of piroplasma equi was noticed in rare numbers for the first time on the 12th day; temperature 99.6 in the morning and rising to 101 in the evening. Piroplasma equi—chiefly spherical form—present in fairly large numbers the following day, causing the temperature to rise from 101.6 in the morning to 103 in the evening. One rosette also present on this date. Further rise to 103.6 in the evening of the next day, but no piroplasms observed. Spherical ring and irregular forms in very small numbers noticed at intervals until the 31st day, the evening temperature reading 103 F. Mucous membrane injected on the 22nd day, but were clearer next morning. No further piroplasms observed; the temperature remained normal, and, accordingly, the examinations were discontinued.

The animal recovered.

2. *Foal 1992.*—

*Injected as above.*

Temperature reaction commenced on the 9th day and reached 106 F. in the morning of the 20th day; it now slowly fell and recorded 98.4 F. on the morning of the 33rd day. A second reaction ensued, reaching the maximum of 104 F. on the mornings of the 38th and 39th days, but gradually fell and remained normal from the 50th day.

No piroplasms observed until the 20th day, and these were in extremely rare numbers—the temperature at that time recording the maximum of 106. The ring forms were noticed in fair numbers on the 22nd day, with a temperature record of 105 F.; no further piroplasms were observed until the commencement of the second reaction, when the ring forms were found in rare numbers on the 39th day—the temperature showed 101 and 102.8 in the morning and evening of this date respectively.

Mucous membranes were slightly injected on the 19th day, and a slight coryza was present; the mucous membranes were icteric and echymosed on the 22nd day, but attained a normal state the following morning, although the animal's appearance was not good.

No further piroplasms were observed, and the examinations were accordingly discontinued.

3. *Foal 1993.*—

*Injected as above.*

A short reaction on the 7th, 8th, 9th and 10th days, rising from 99 F. The curve now assumed a regular reaction, lasting until the 31st day, and touching the maximum of 104.2 F. on the 20th day. This was followed by a second rise of a fairly regular nature until the morning of the 46th day, when the temperature recorded 99.4 F., but was immediately followed by a sharp rise to 106 in the evening of the 48th day. Temperature now returned to normal.

Piroplasms in very rare numbers noted for the first time on the 8th and 9th days, coinciding with the rise of temperature to 103. Examinations gave negative results until four days later, when one piroplasma equi and one rosette were present. Another piroplasm seen on the 16th day accompanied with the

lesions of a slight poikilocytosis. Spherical ring, leaf, and irregular forms occasionally noted in very rare numbers from the 19th to 33rd days, but further examinations gave negative results. Mucous membranes injected and icteric on the 22nd days. On the 26th day a rather profuse coryza was present, but no further lesions observed.

*Examination* discontinued.

Recovered.

4. *Foal* 1994.—

*Injected* as above.

*Reaction* of a regular nature lasted from the 6th day, until the evening of the 16th day, when a sharp rise occurred to 104.8 on the following evening, and 105 F. on the evenings of the 19th and 20th days. It decreased from this date, and assumed a regular but slight reaction between 100 and 102 F. for the next 25 days.

The ring form in rare numbers observed for the first time on the 9th day, accompanied with the appearance of a few megaloblasts. All examinations negative until the 18th day, when a slight coryza was present, and the mucous membranes were injected. Piroplasms very scarce the following day, and on the 20th day the mucous membranes were observed to be slightly yellow. Mucous membranes slightly jaundiced on the 22nd day, but clearer the following morning. Piroplasms were still absent. The lesions of a slight poikilocytosis observed on the 27th day, and two days later the ring form was present in very rare numbers. These ring forms increased in number the following day, but coinciding with the fall in temperature from 102 F. to 99.4, the latter being obtained on the 33rd day. The number of piroplasms decreased, and on the 34th day only one rosette was present.

Further examinations proved negative, and consequently were discontinued from the 37th day.

Recovered.

*Results* of four Transvaal horse foals injected with blood of a horse immune against piroplasma equi all showed a reaction due to this inoculation, accompanied with piroplasms, and none died.

*Injections of Argentine Horses with Blood of Transvaal Horse Foals.*

EXPERIMENT No. 2.

*Second Generation.*

(a) *Injections with blood of horse foal* 1991.

1. *Horse* 2097.—Mare about four years old, directly imported from the Argentine.

*Injected* on the 9th July with 5 c.c. fresh defibrinated blood from foal 1991.

*Reaction* commenced on the 10th day, rising to 106 in the morning of the 12th day. A fairly sharp drop from this reading to 99.4 on the morning of the 17th day, was followed by a regular reaction between 101 and 105 F., which lasted until death on the 48th day. Coinciding with the rise to 106 F. numerous ring forms were present, and remained in fair numbers for the next

six days, but two days later—19th day after injection—they were recorded as very scarce.

On the 14th day—two days after the maximum of 106 F.—the mucous membranes were yellow, and hindquarters weak. On the 16th day the mucous membranes were still yellow, and also echymosed, remaining yellow until the 25th day. Piroplasms again present in rare numbers on the 26th day, and a few points were observed three days later, increasing on the 31st day, and remaining in fair numbers until the 46th day.

Piroplasms in scarce numbers noted for the last time on the 36th day, and twelve days later the animal died from bronchopneumonia, complicated with pregnancy.

*Post-mortem Examination.*—

*Condition:*—Good. Uterus pregnant; vulva swollen; blood of a normal colour.

*Lungs:*—Right middle lobe contained a patch of hepatisation with mucus in the bronchi; in the left middle lobe was an abscess the size of a walnut; right posterior lobe shows white thrombi in the veins.

*Heart:*—Ventricles normal but rather pale.

*Spleen:*—Normal.

*Liver:*—Normal.

*Stomach:*—Normal.

*Kidneys:*—Pale; capsula not easily stripped off.

*Intestines:*—Mucous membranes pale.

2. *Horse 2098.*—Three-year-old mare, directly imported from Argentine.

*Injected* on the 9th July, 1906, with 5 c.c. fresh defibrinated blood of foal 1991.

Irregular reaction from date of injection. A typical reaction commenced on the 13th day until the 24th day, and reaching the maximum of 105.8 on the 19th day. On the 28th day another sharp rise ensued, lasting for four days, and followed by a slight reaction between 99 and 103 until the 56th day.

*Examinations.*—Two piroplasms seen for the first time on the 12th day, and two days later piroplasma equi, and rosettes were present in fair numbers. The hindquarters of the animal were noticed to be slightly weak during the first rise, but no piroplasma seen at the time of the maximum temperature. On the 21st day—temperature being 103 in the morning and 104.8 F. in the evening—the mucous membranes were pale, and two points were present.

On the 25th day—at the conclusion of the primary rise—the animal's legs were weak, but no piroplasms were observed until the day after the second rise. Piroplasms and points were present in fair numbers for the next eight days, but on the 40th day only a few points were visible. These points were present in rare numbers on the 45th, 47th, 49th and 51st days, and on this latter date the urine was tested, but no traces of albumen found. One piroplasm was found on the 56th day, and two days later another piroplasm, also a few points. All further examinations proved negative, and were accordingly discontinued.

The animal recovered.

(b) *Injections with blood of foal 1992.*

3. *Horse 2099.*—Mare, 3½ years old, and directly imported from the Argentine.

*Injected* on the 9th July, 1906, subcutaneously with 5 c.c. fresh blood of foal 1992.

*Reaction* commenced on the 14th day, rising to a maximum of 105.4 on the 21st day, when the mucous membranes were noticed to be pale. Temperature gradually fell for the next six days, but suddenly dropped to a sub-normal record of 96.4 on the 28th day. The animal, however, rallied, and the temperature fluctuated between normal and 103.4 for the next eight days. Death occurred on the 15th August—37 days after injection—from piroplasmosis, complicated with pregnancy.

*Blood Examinations.*—Piroplasms noticed in very rare numbers for the first time on the 12th day. A few rosette forms noticed a few days later; piroplasms increased on the 15th and 16th days, but only two were observed on the 17th day. A slight poikilocytosis recorded on the 19th day. Examinations proved negative during the rise of temperature to 105.4, and were not observed until the 29th day, just after the animal had rallied from 96.4 F., when piroplasms and points were observed in fair numbers, and on the day of death were very numerous.

*Post-mortem Examination.*—

*Condition:*—Fair; mucous membranes blanched; uterus pregnant.

*Lungs:*—Slightly œdematous.

*Heart:*—About an ounce of clear fluid in pericard.

*Spleen:*—Slightly swollen.

*Liver:*—Slightly swollen, but not icteric.

*Stomach:*—A few hæmorrhages on mucous membranes.

*Kidneys:*—Swollen; capsulas adherent.

*Intestines:*—A few punctate hæmorrhages in cæcum.

4. *Horse 2100.*—Mare, two years old, and directly imported from the Argentine.

*Injected* on the 9th July subcutaneously with 5 c.c. fresh defibrinated blood of foal 1992.

*Temperature* fluctuated directly after injection, from 98 F. in the morning to 102 in the evening. A regular reaction commenced on the 11th day, rising to 105 F. three days later, and accompanied on this date with a yellowish appearance of mucous membranes. Slight fall noticed for the next three days, and, in the morning of the 17th day—temperature being 101—the mucous membranes were slightly yellow; the hindquarters were weak. In the afternoon of the same day the animal died, the cause of death being recorded as gangrenous pneumonia.

*Examinations.*—Piroplasms were present for the first time on the 12th day; noticed in rare numbers the following morning, and very numerous at the time of the maximum temperature of 105 F. No piroplasms observed at death.

*Post-mortem Examination.*—

*Condition:*—Fair; mucous membranes pale; blood-stained discharge from nostrils.

*Lungs*:—Anterior lobe and lower edge of left lung gangrenous; almost the whole of the remainder of the left lung hepatised; ecchymoses on pleura.

*Heart*:—Normal.

*Spleen*:—Enlarged.

*Stomach*:—Normal.

*Kidneys*:—Swollen and pale.

*Liver*:—Normal.

*Intestines*:—Normal.

(c) *Injections with blood of horse foal 1993.*

5. *Horse 2101.*—Mare, three years old, and directly imported from the Argentine.

*Injected* subcutaneously on the 9th July, 1906, with 5 c.c. fresh blood of foal 1993.

*Reaction* started immediately, rising to 104 on the 7th day, but the animal died the following morning from gastro-enteritis (probably horse-sickness).

All examinations negative.

*Post-mortem.*—

*Condition*:—Poor.

*Lungs*:—Normal.

*Heart*:—Normal.

*Spleen*:—Normal.

*Liver*:—Normal.

*Kidneys*:—Normal.

*Stomach*:—Intense patchy congestion.

*Intestines*:—Duodenum as far as end of large colon congested; remainder normal.

6. *Horse 2102.*—Mare, three years old, directly imported from the Argentine.

*Injected* on the 9th July, 1906, subcutaneously with 5 c.c. fresh blood of foal 1993.

*Temperature.*—Reaction from the 6th day, rising to 104.6 on the 14th day, but falling to 99.8 two days later. For the next sixteen days it fluctuated between 99.6 and 105.6, on the 34th day recording 99.4, which marked the commencement of a secondary reaction, lasting for about 18 days. This secondary reaction was of a very irregular character, and fluctuated between 98.8 in the morning to 104 in the evening.

*Examinations.*—The ring form of piroplasma noticed for the first time on the 10th day in fair numbers, and irregular forms present the following day. Occasional points and piroplasms observed on the 15th, 16th, 17th, 18th, and 20th days, but none were seen at the time of the maximum temperature on the 23rd to 26th days. The lesions of a slight poikilocytosis were present on the 26th day, and, four days later, piroplasms and points again appeared. These were present almost daily until the 67th day, this being the last time any were present. Future examinations gave negative results, and were accordingly discontinued from the 75th day. The lesions of a slight poikilocytosis were again noticed on the 50th and 58th day after injection.

Recovered.

(d) *Injections with blood of horse foal 1994.*

7. *Horse 2103.*—Four-year-old mare, directly imported from the Argentine.

*Injected* on the 9th July, 1906, with 5 c.c. defibrinated fresh blood of foal 1994.

*Reaction* commenced on the 11th day, reaching the maximum of 105.4 on the 26th day, and dropping to 97 on the morning of the 32nd day. Recovered to 101.6 in the evening and now remained normal.

*Examinations.*—One piroplasm seen for the first time on the 11th day coinciding with the commencement of the reaction. Piroplasms and rosettes noticed for the next six days, and, on this latter date—the 17th day after injection—the mucous membranes were slightly yellow and the hindquarters weak. Piroplasms in rare numbers were present on the 21st day—the morning temperature being 101.4, and, in the evening, 104.1. No piroplasms seen during the time of the maximum temperature and remained absent until two days previous to the sub-normal record of 97, on which day—the 30th after the injection—two piroplasms were present and several points. A few points were present on the 37th and 40th days, and one piroplasm on the 44th day.

The animal recovered.

8. *Horse 2104.*—Three-year-old mare, and directly imported from the Argentine.

*Injected* on the 9th July, 1906, with 5 c.c. defibrinated blood of foal 1994.

*Temperature Records.*—Typical reaction started on the 11th day, rising to 103.2 in the morning of the 19th, and 106 in the evening of the 21st, but steadily dropped, and recorded 99.2 on the morning of the 29th day. It remained normal for five days, but, on the 35th day, started to rise, reaching 103.2 on the evening of the 37th day, and suddenly fell next morning.

The animal died the same afternoon.

*Examinations.*—Piroplasms seen for the first time on the 15th day, remaining present for four days. Mucous membranes slightly yellow on the 17th day, and in the same condition on the day of the maximum temperature of 106—the 21st day after injection—on which date one piroplasm and a few points were noted. No further piroplasms observed until the temperature dropped to normal, when, on the 30th, 31st and 32nd days, points were present in fair numbers. Coinciding with the second rise, the piroplasms increased, and, on the three days previous to death, were present in extremely large numbers.

The animal died on the 17th August from piroplasmosis, complicated with pregnancy.

*Post-mortem.*—

*Condition:*—Good; uterus pregnant. Subcutaneous tissue very icteric.

*Lungs*:—Pale.

*Heart*:—One ecchymose in left ventricle; a few petechiae (in one patch) in right ventricle.

*Spleen*:—Slightly swollen.

*Liver*:—Very slightly icteric.

*Stomach*:—Normal.

*Intestines*:—Cæcum slightly congested; fæces yellow: numerous filaria papillosa in peritoneal cavity.

*Results* of eight Argentine mare horses injected with blood of Transvaal horse foal (1st generation) immune against piroplasma equi. They all showed a reaction accompanied with the appearance of piroplasma equi, and five died. In two of these cases, however, the mares were in foal, and no doubt this factor had some bearing on the mortality, but, at the same time, it will be noted from the post-mortem reports that two deaths were complicated with pneumonia—probably contracted on board ship, one of these animals also being a mare heavy in foal.

## 2.—INOCULATION AGAINST EQUINE PIROPLASMOSIS BY MEANS OF HORSE FOAL BLOOD—ORIGIN MULE BLOOD.

	Mule 589	...	...	Origin.
	Horse foal 1766	...	...	1st generation.
	├── Horse foal 2053.			
	└── Horse foal 2054	...	...	2nd generation.
	Horse foal 2314	...	...	3rd generation.
	Horse foal 2273	...	...	4th generation.
	Horse foal 2274	...	...	5th generation.
	Horse foal 2621	...	...	6th generation.
	Horse foal 2734	...	...	7th generation.
	Horse foal 2786	...	...	8th generation.

## 2.—INOCULATION BY MEANS OF HORSE FOAL BLOOD—ORIGIN MULE BLOOD.

*Mule 589*.—(Compare "Further Notes on Piroplasmosis of the Horse, Mule and Donkey," Annual Report, 1904-5, Experiment "B," No. 1, page 98).

This was an Argentine mule, and had been injected with blood of three horses immune against piroplasma equi. Two reactions due to the injection and piroplasma equi present during primary reaction on the 7th day for the first time; unfortunately no microscopical examinations were made during secondary reaction.



## EXPERIMENT NO. 3.

*First Generation.*

1. *Horse Foal 1766.*—Transvaal foal, about six months old.

*Injected* on the 19th April, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of mule 589.

Very slight reaction from date of injection, the temperature consistently remaining between 99 and 103 for 63 days.

*Examinations.*—No piroplasms seen until the 22nd day, but then only in very rare numbers. They were never numerous, but were present on the 23rd, and from the 26th to 30th days. All further examinations negative, and accordingly discontinued from the 57th day.

*Result.*—One Transvaal horse foal injected with blood of a mule immune against *piroplasma equi*, passed through a slight reaction, accompanied with piroplasms.

## EXPERIMENT NO. 4.

- 1.—*Transvaal horse foals injected with blood of Transvaal horse foal 1766, immune against piroplasma equi.*

*Second Generation.*

Horse foals 2053 and 2054, both obtained from S.A.C., were injected on the 4th July, 1906, subcutaneously with 5 c.c. defibrinated blood of Transvaal horse foal 1766.

1. *Horse Foal 2053.*—Nine-month-old gelding obtained from S.A.C.

*Injected* as above.

*Temperature.*—Sharp rise from 99.4 in the morning of the 6th day to 104.2 in the evening. The regular reaction commenced on the 9th day and reached the maximum of 104 in the evening of the 20th. A second reaction now ensued, the temperature remaining between 100 and 103.6 for the next 16 days, after which it resumed a normal aspect.

*Examinations.*—Piroplasms in scarce numbers observed for the first time on the 15th day, and remained present for two days, one ring also being noted on the 16th day. On the day of the maximum temperature—the 20th—one piroplasm was present, and the following day points were noticed. Points again present on the 27th and 37th days; one piroplasm noted on the 25th day and two on the 38th day. All further examinations proved negative and were accordingly discontinued.

The animal recovered.

2. *Horse Foal 2054.*—A six-month-old gelding.

*Injected* as above.

*Temperature* reached 104 on the 3rd day, but dropped to normal six days later, when a regular reaction ensued, lasting for 20 days, and reaching the maximum of 103.4 on the 21st day after injection. A secondary rise noted from the 27th day, rising to 104 two days later, and remaining between 99 and 103 for 18 days.

*Examinations.*—*Piroplasma equi* in rare numbers present for the first time on the 16th and 17th days. Coinciding with the

commencement of the second reaction, one piroplasm was present on the 27th and following day. All further examinations were discontinued from the 41st day.

The animal recovered.

*Results* of two Transvaal horse foals injected with blood of foals immune against *Piroplasma equi* (first generation), both showed a reaction, accompanied with piroplasms, and recovered.

2.—*Argentine horses injected with blood of Transvaal horse foal 1766, immune against Piroplasma equi.*

3. *Horse 2067.*—Two-year-old mare, directly imported from the Argentine.

*Injected* on the 16th June, 1906, with 5 c.c. defibrinated fresh blood of foal 1766.

*Reaction* commenced on the 9th day, reaching the maximum of 105.4 in the evening of the 13th day, and returning to normal on the 23rd day. Five days later a second reaction ensued, rising from 99.8 in the morning to 106 in the evening. It gradually fell for the next two days, and remained between 101.4 and 104.2 until the 42nd day. The animal died two days later from the sequel of piroplasmosis.

*Examinations.*—Coinciding with the rise to 105.6 on the 13th day, piroplasms were present in large numbers, but very scarce the following day. All other examinations negative. Mucous membranes pale and yellowish, and number of corpuscles on the 14th dropped from 3,900,000 per c.m.m. to 2,100,000.

4. *Horse 2075.*—Argentine, two years old, mare, and directly imported.

*Injected* on the 9th July, 1906, with 5 c.c. fresh defibrinated blood of foal 1766.

*Reaction* commenced on the 5th day, rising to 104 in the evening of the 8th day, but fell to 99.6 in the morning of the 11th day. The animal died the same evening from syncope.

*Result.*—Of two Argentine mare horses injected with blood of a Transvaal foal immune against *Piroplasma equi* (first generation), both showed a reaction, and subsequently died, one of the sequel of piroplasmosis and the other from syncope.

3.—*Argentine donkeys, injected with blood of Transvaal horse foal 1766, immune against Piroplasma equi.*

*Second Generation.*

*Note.*—The following donkeys were all injected on the 16th June, 1906, with 5 c.c. fresh defibrinated blood of foal 1766.

5. *Donkey 841.*—Argentine, six-year-old mare.

*Injected* as above.

*Temperature.*—No distinct reaction.

6. *Donkey 842.*—Five-year-old stallion, imported from the Argentine.

*Injected* as above.

*Temperature.*—No distinct reaction.

Recovered.

7. *Donkey 843.*—Six years old; Argentine mare.

*Injected* as above.

*Temperature.*—No distinct reaction.

Recovered.

8. *Donkey* 844.—Four-year-old Argentine mare.

*Injected* as above.

*Temperature*.—No distinct reaction

9. *Donkey* 845.—Four-year-old mare, Argentine.

*Injected* as above.

*Temperature*.—No distinct reaction.

*Result*.—Of five Argentine donkeys injected with blood of a Transvaal foal (first generation), immune against *piroplasma equi*, none showed a distinct reaction, neither did any die.

#### EXPERIMENT No. 5.

##### *Third Generation.*

*Argentine mules injected with blood of Transvaal horse foal 2053, immune against piroplasma equi.*

Argentine mules 2315, 2320, 2321, 2322, 2324 and 2325 were injected on the 5th April, 1906, with 5 c.c. fresh defibrinated blood of Transvaal horse foal 2053.

1. *Mule* 2315.—Aged gelding.

*Injected* as above.

*Temperature*.—Reaction commenced on the 8th day, and recorded 104.8, 105 and 105 in the evenings of the 13th, 14th and 15th days respectively. It now dropped and remained normal for 11 days. On the 28th day, a second reaction ensued, reaching the maximum of 105 five days later. The temperature fell sharply and remained normal from the 39th day.

*Examinations*.—Piroplasms present for the first time on the 11th day, and remained in fair numbers during the height of the first reaction. A single piroplasm noticed on the 19th day, and coinciding with the second reaction; the lesions of poikilocytosis and piroplasms were present on the 33rd and 34th days. All further examinations negative.

2. *Mule* 2320.—Three-year-old Argentine gelding.

*Injected* as above.

*Temperature*.—Reaction commenced on the 11th day, rising to 103.4 in the evening, and 103.8 twenty-four hours later, but fell and remained normal from the 15th day.

*Examinations*.—The lesions of poikilocytosis were noticed on the 12th and 13th days, and on this latter date one point was present. Piroplasma equi noted for the first time on the 15th day and again four days later.

Recovered.

3. *Mule* 2321.—Three-and-a-half-year-old Argentine.

*Injected* as above.

*Temperature*.—Very slight reaction from the 10th day, reaching 102.4 in the evening two days later.

*Examination*.—Two points and the lesions of a slight poikilocytosis present on the 13th day, and two days later piroplasms in fair numbers seen for the first time. Poikilocytosis also noted on the 18th and following day.

Recovered.

4. *Mule 2322*.—Argentine gelding, two and a half years old.  
*Injected* as above.  
*Temperature*.—Slight reaction from the 8th day, recording the maximum temperature of 102.4 on the 16th day, but slowly fell from that date, and remained normal.  
*Examinations*.—The lesions of poikilocytosis noted on the 12th day, and again 10 days later. On the 24th day two points were present, but no piroplasms were noticed.  
 The animal recovered.
5. *Mule 2324*.—Aged Argentine gelding.  
*Injected* as above.  
*Temperature*.—Very slight reaction from the 8th day and lasting for 10 days.  
*Examinations*.—On the day of the maximum temperature of 102—12 days after injection—three points were present; three days later one piroplasm was noted for the first time. Another piroplasm was present the following day, and two noted on the 19th day. All further examinations negative, and were discontinued from the 27th day.
6. *Mule 2325*.—Aged Argentine gelding.  
*Injected* as above.  
*Temperature*.—Reaction of a very slight nature from the 8th to 19th days. On the 24th day the temperature commenced to rise and indicated a second reaction, reaching 103.2 four days later.  
*Examinations*.—The lesions of a slight poikilocytosis observed during the height of the first reaction, and on the 15th and 16th days points were present in fair numbers. Piroplasma equi not noted during either reaction.  
 Recovered.  
*Results*.—Of six Argentine mules injected with blood of a Transvaal horse foal, 2nd generation, immune against piroplasma equi, all showed a reaction, and accompanied in four cases with piroplasms, and recovered.

#### *Third Generation.*

*Transvaal horse foal injected with blood of Transvaal horse foal 2054*  
 —immune against piroplasma equi.

7. *Horse foal 2314*.—Obtained from the S.A.C.  
*Injected* on the 16th October, 1906, with 5 c.c. defibrinated fresh blood of foal 2054.  
*Temperature*.—Fluctuated between 100 and 103 for the first 10 days, and on the evening of the 11th day reached 105, dropped to 99.6 on the morning of the 14th day, but returned to 105.6 in the evening of the 16th day. The 20th day marked the commencement of a second reaction lasting for 11 days, although not of as strong a nature as the first.  
*Examinations*.—Piroplasms and rosettes present on the 10th day for the first time; the former increased in numbers the following day, but were only noted in rare numbers each day until the 19th. Piroplasms and points were occasionally noted during the next 20 days, and on the 39th day after injection the lesions of poikilocytosis were observed. All further examinations negative.

*Argentine horses injected with blood of Transvaal foal 2054—immune against piroplasma equi.*

Argentine horses 2231, 2242 were injected on the 16th October, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 2054.

8. *Horse 2231.*—Four-year-old Argentine mare.

*Injected as above.*

*Temperature.*—Very slight reaction between 100 and 102.4, lasting for 19 days. On the 22nd day after injection a strong curve was noted, the temperature recording 106.4 in the evenings of the 27th and 28th days, after which it slowly fell and regained normal on the 35th day after injection.

*Examinations.*—On the 7th day points were noted, the eyes were yellow, and the animal was generally weak, but the following day an improvement was shown. Points were again present on the 9th, 10th, and 11th days, and on the 15th day the lesions of poikilocytosis appeared. Piroplasms noted for the first time on the 16th day, and were present in rare numbers until the 24th day. Coinciding with the time of the maximum temperature during the second reaction, the animal was noticed to be weak; piroplasms were noted on the 28th and 29th days, but in rare numbers. Piroplasms, points, and rosettes were occasionally present during the next 14 days.

9. *Horse 2242.*—Six-year-old Argentine gelding.

*Injected as above.*

*Temperature.*—On the 3rd day a rise to 104 was noted, but on the 7th day it recorded 100, from when a reaction commenced, reaching 105 on the 12th day, and keeping high for a considerable time. 105.2 was recorded in the evenings of the 20th, 21st, 22nd, and 23rd days. The temperature now remained normal until the 36th day, when a rise from 100.4 in the morning to 105 in the evening was noted (on account of relativity of the anus, the temperature record was probably not accurate). The animal died the following morning from broncho-pneumonia, probably contracted on board ship.

*Examinations.*—One piroplasm seen for the first time on the 7th day, when the eyes were noted to be slightly yellow, and the animal was generally weak. This weakness was not so pronounced the following day, and on the 10th day from injection piroplasms were again noted accompanied with points. Rings, piroplasms, and points were noted almost daily for the next 20 days. The animal remained in poor condition during the whole of the reaction, with petechial spots in the eyes and pallid mucous membranes. The lungs were also affected.

*Post-mortem.*—

*Condition:*—Good.

*Lungs:*—Apex and lower edge of right lung and also lower edge of left lung shows broncho-pneumonia (red and grey hepatisation). A few patches of yellow fibrinous lymph on surface of right lung.

*Heart:*—Normal.

*Liver:*—Congested.

*Kidneys:*—Congested.

All other organs normal.

*Result.*—Of one Transvaal horse foal and two Argentine horses injected with blood of a Transvaal foal, immune against *piroplasma equi*, 2nd generation, all showed a reaction accompanied with *piroplasma equi*, and one Argentine horse died from broncho-pneumonia—probably ship's pneumonia and contracted on board ship.

*Transvaal mules injected with blood of Transvaal horse foal 2054—immune against piroplasma equi.*

10. *Mule 2216.*—Three-year-old gelding.

*Injected* on the 16th October, 1906, with 5 c.c. defibrinated fresh blood of foal 2054.

*Temperature.*—Very slight reaction, only recording a maximum of 101.4 on the 14th day.

*Examinations.*—Points present on the 10th day, and the following morning piroplasms, in rare numbers, noted for the first time. Piroplasms were also present in rare numbers on the 15th, 16th, 17th, and 18th days. The lesions of poikilocytosis noticed on the 21st and 22nd days, but all further examinations proved negative.

Transvaal mules 2218, 2219, 2221 and 2232 were injected subcutaneously on 21st September, 1906, with 5 c.c. fresh defibrinated blood of Transvaal horse foal 2054.

11. *Mule 2218.*—Three-year-old Transvaal gelding.

*Injected* as above.

*Temperature.*—A slight reaction from the 18th day, continuing until discharge.

*Examinations.*—A few rings noted on the 14th day, remaining for the following two days. The lesions of a slight poikilocytosis noticed on the 18th day and again on the 23rd day. Poikilocytosis and points were occasionally noted until the 32nd day, but no piroplasms were seen.

12. *Mule 2219.*—Three-year-old Transvaal mare.

*Injected* as above.

*Temperature.*—Reaction commenced on the 7th day, reaching 103 in the evening of the same day. A second reaction followed from the 15th day lasting for 16 days, and reaching the maximum temperature of 103 on the 21st day after injection.

*Examinations.*—Rings noted on the 14th, 15th, and 16th days and a few points on the 26th day. The lesions of poikilocytosis observed on the 35th and 27th days, but no piroplasms.

13. *Mule 2221.*—Three-year-old Transvaal gelding.

*Injected* as above.

*Temperature.*—Reaction from the 9th day, but of a very indistinct nature, and lasting until the 26th day after the injection.

*Examinations.*—One piroplasm noted for the first time on the 12th day; they increased and became fairly frequent on the 14th and 15th days; on the former date—the 14th—rods were also present. Piroplasms were again noted on the 18th and 22nd days, and on the 25th day the lesions of poikilocytosis were

observed. Poikilocytosis also present on the 26th day, accompanied with points, and the latter were again noted on the following day.

14. *Mule 2232.*—Three-year-old Transvaal mare.

*Injected* as above.

*Temperature.*—A sharp rise noted from the date of injection, reaching 104 in the evening of the 4th day, but recovered to normal, and from the 10th day a reaction of a rather indistinct nature ensued, reaching 103 on the 14th day, and remaining between this figure and 100 for 18 days.

*Examinations.*—Rings noted on the 14th day in fair numbers, and were also present on the 15th day, when they were accompanied with rods. One piroplasm noted for the first time on the 18th day, together with the lesions of a slight poikilocytosis. Poikilocytosis was also noted on the 22nd day, and two days later another piroplasm was present. Points were noticed the following day, and on the 26th day piroplasms and points were present in fair numbers.

*Inoculation of Argentine mules with blood of Transvaal foal 2054—  
immune against piroplasma equi.*

Argentine mules 2368, 2369, 2370, 2371 and 2372 were all injected subcutaneously on the 26th November, 1906, with 5 c.c. defibrinated fresh blood of Transvaal horse foal 2054.

15. *Mule 2368.*—Aged Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction of a distinct character, lasting from the 7th to the 18th day, and reaching the maximum temperature of 102.4 on the evening of the 7th day.

*Examinations.*—All negative.

16. *Mule 2369.*—Eighteen-month-old stallion imported from the Argentine.

*Injected* as above.

*Temperature.*—Reaction of an indistinct nature, reaching 102.4 on the 7th day, and returning to normal on the 15th day.

*Examinations.*—Piroplasms present for the first time in rare numbers on the 14th day, accompanied with the lesions of poikilocytosis. Slight poikilocytosis observed three days later. All further examinations negative.

17. *Mule 2370.*—Six-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction from the 9th to the 19th day, recording the maximum of 102.8 on the 14th day.

*Examinations.*—The lesions of poikilocytosis noticed on the 20th day. All other examinations negative.

18. *Mule 2371.*—Four-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—No distinct reaction.

*Examination.*—Slight poikilocytosis noted on the 14th day, and three days later piroplasms seen for the first time. All further examinations negative.

19. *Mule 2372*.—Eighteen-month-old Argentine mare.

*Injected* as above.

*Temperature*.—Sharp reaction from the 4th day, lasting for four days, and recording between 103 and 104. A slight reaction noted from the 10th to the 18th days, but only reaching 102.8.

*Examinations*.—Piroplasms seen for the first time on the 18th day, and in rare numbers three days later. Rosette forms noted on the 20th day.

- Argentine mules 2514, 2518, 2519, 2520, 2522, 2523, 2524, 2525, 2531, 2532, 2548 were all injected subcutaneously on the 30th January, 1907, with 5 c.c. fresh defibrinated blood of foal 2054.

20. *Mule 2514*.—Argentine gelding.

*Injected* as above.

*Temperature*.—Reaction from the 11th day, lasting for 10 days, and recording the maximum temperatures of 104 and 104.4 on the 17th and 18th days respectively.

*Examinations*.—All negative.

21. *Mule 2518*.—Argentine mare.

*Injected* as above.

*Temperature*.—Reaction from the 5th day, reaching a maximum of 104.2 in the evening of the 11th day, and remaining about this record for the next seven days.

*Examinations*.—All negative.

22. *Mule 2519*.—Argentine mare.

*Injected* as above.

*Temperature*.—Sharp rise from the 4th day, reaching 103.6 24 hours later. Reaction of an indistinct nature from the 10th to the 21st days, the evening record on this latter date being 103.6.

*Examinations*.—All negative.

23. *Mule 2520*.—Argentine gelding.

*Injected* as above.

*Temperature*.—At the date of injection the temperature recorded 105, but slowly fell to 101 on the 4th day, rising again to 104 on the 6th day. On the morning of the 11th day the temperature was 100.6, rising in the evening to 104. It now fell and remained normal.

*Examinations*.—All examinations negative.

24. *Mule 2522*.—Argentine mare.

*Injected* as above.

*Temperature*.—Very slight reaction, the temperature recording the maximum of 103 on the 13th day.

*Examinations* negative.

25. *Mule 2523*.—Argentine stallion.

*Injected* as above.

*Temperature*.—Slight reaction from the 7th day, reaching 102.8 six days later.

*Examinations*.—Slight poikilocytosis noted on the 13th day. All other examinations negative.

26. *Mule 2524*.—Argentine gelding.

*Injected* as above.

*Temperature*.—Very slight reaction.

*Examinations*.—All negative.



27. *Mule 2525*.—Argentine mare.  
*Injected* as above.  
*Temperature*.—Very slight reaction.  
*Examinations*.—All negative.
28. *Mule 2531*.—Argentine gelding.  
*Injected* as above.  
*Temperature*.—Reaction from the 7th day, reaching 103 on the 11th and 12th days.  
*Examinations*.—All negative.
29. *Mule 2532*.—Argentine gelding.  
*Injected* as above.  
*Temperature*.—Reaction from the 8th day, lasting 15 days, and reaching a maximum of 104.4 fourteen days after injection.  
*Examinations*.—The lesions of poikilocytosis noted on the 15th day. All other examinations negative.
30. *Mule 2548*.—Argentine mare.  
*Injected* as above.  
*Temperature*.—Sharp rise to 104 on the 1st day after injection, and from the 8th day a slight reaction was noticed lasting for 12 days. A second slight reaction to 104.2 was noted on the 23rd day, but only lasted for six days.  
*Examinations*.—All negative.

*Argentine donkeys injected with blood of Transvaal foal 2054—  
immune against piroplasma equi.*

Argentine donkeys 2248, 2249, 2250 and 2251 were injected on the 26th November, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 2054.

31. *Donkey 2248*.—Five-year-old Argentine mare.  
*Injected* as above.  
*Temperature*.—Slight reaction, the temperature remaining between 98 and 102 for 21 days after injection.  
*Examinations*.—All negative.
32. *Donkey 2249*.—Four-year-old Argentine gelding.  
*Injected* as above.  
*Temperature*.—Very slight reaction.  
*Examinations*.—One point noted on the 15th day. All other examinations negative.
33. *Donkey 2250*.—Three-year-old Argentine mare.  
*Injected* as above.  
*Temperature*.—Slight reaction.  
*Examinations*.—One piroplasm seen on the 18th day. All other examinations negative.
34. *Donkey 2251*.—Four-year-old Argentine gelding.  
*Injected* as above.  
*Temperature*.—Slight reaction.  
*Examinations*.—Piroplasms seen on the 15th day only, but in very rare numbers.

Argentine donkeys 2256, 2257 and 2258 were all injected subcutaneously on the 16th October, 1906, with 5 c.c. defibrinated fresh blood of Transvaal horse foal 2054.

35. *Donkey 2256*.—Five-year-old Argentine mare.

*Injected* as above.

*Temperature*.—On the 4th day after injection the temperature rose from 99.8 in the morning to 103.4 in the evening. It remained high for the next three days, and the donkey died of debility. On *post-mortem*, anaemia, fatty degeneration and hydronephrosis was found.

*Note*.—Slipped her foal during the reaction.

36. *Donkey 2257*.—Three-year-old Argentine mare.

*Injected* as above.

*Temperature*.—Slight reaction, from the 7th day, lasting for 11 days, and recording as a maximum 102.8 in the evening of the 10th day after injection.

*Examinations*.—Piroplasms in rare numbers noted for the first time on the 10th day, points having been noticed two days previously. Piroplasms and the lesions of poikilocytosis were noted almost daily from the 10th to 24th days.

37. *Donkey 2258*.—Two-year-old Argentine mare.

*Injected* as above.

*Temperature*.—A slight reaction from the 9th to the 19th day. A second slight reaction from the 26th to the 35th days, touching 103 on the 30th day after injection.

*Examinations*.—One piroplasm seen for the first time on the 10th day, and piroplasms again noted two days later, accompanied with points. The lesions of poikilocytosis appeared on the 14th day, and piroplasms were noted daily from the 15th to 18th days. Piroplasms present in very rare numbers on the 21st and 23rd days, and on the 24th day from injection the lesions of poikilocytosis were again noticed.

*Results of 5 Transvaal mules, 16 Argentine mules and 7 Argentine donkeys injected with blood of a Transvaal horse foal—immune against piroplasma equi (2nd generation).*

All passed through a piroplasmosis reaction, and, with the exception of 1 donkey which died of debility, all recovered.

#### EXPERIMENT No. 6.

##### *Fourth Generation.*

*Transvaal Horse Foal 2314*.—Transvaal horse foal 2314 had been injected with blood of foal 2054 on the 16th October, 1906 [compare Experiment No. 5 (7)].

*Injection of Transvaal horse foal with blood of Transvaal horse foal 2314—immune against piroplasma equi.*

1. *Horse Foal 2273*.—Four-month-old foal, and born on the station.

*Injected* on the 10th December, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2314.

*Temperature*.—Reaction from the 3rd day, reaching 104.2 on the 6th day, and regaining normal 16 days after injection. A second reaction noted from the 22nd day, reaching 105 in the evening five days later, when the temperature fell and remained from the 35th day after injection.

*Examinations.*—Piroplasms noted for the first time on the 11th day, and were present on the following two days. Again noted on the 15th day, accompanied with a slight poikilocytosis, the latter being also noticed on the 19th day. Points were present the following day, and, together with piroplasmus, were frequently noted during the next 10 days. One rosette also seen on the 45th day.

*Argentine mules injected with blood of Transvaal foal 2314, immune against piroplasma equi.*

2. *Mule 2448.*—Argentine gelding. (Had been in a horse-sickness experiment previously.)

*Injected* subcutaneously on the 14th February, 1907, with 3 c.c. fresh defibrinated blood of foal 2314.

*Temperature.*—Slight reaction from the 6th day, reaching 102.8 on the 10th day, and remaining normal from the 14th day after injection.

*Examinations.*—All negative.

*Note.*—The following Argentine mules—Nos. 2484, 2486, 2487, 2488, 2498, 2490, 2491, 2492, 2493, 2500, 2501, 2502—had been running on a farm infected with piroplasmosis, and, in addition, had been utilised for horse-sickness experiments before the injection of 3 c.c. defibrinated fresh blood of Transvaal horse foal 2314 on the 7th February, 1907.

3. *Mule 2484.*—Four-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—Slight reaction.

4. *Mule 2486.*—Three-and-a-half-year-old mare, imported from the Argentine.

*Injected* as above.

*Temperature.*—Reaction from the 4th day, recording 105 three days later, and for the next four days the evening temperature remaining about 104. A second reaction noted from the 21st day, lasting for 10 days, and reaching the maximum temperature of 104 on the 25th day after injection.

5. *Mule 2487.*—Three-and-a-half-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction from the 10th day, reaching 103.8 three days later. A sharp rise was noted from the 19th day, rising to 103.2 the following day, falling to 100 in the morning of the 22nd day, and rising in the evening to 104.2.

6. *Mule 2488.*—Argentine mare, about three and a half years old.

*Injected* as above.

*Temperature.*—No distinct reaction until the 24th day after injection, when the temperature rose from 99.6 in the morning to 103.6 in the evening, and remaining between 103 and 104 for the next seven days.

*Examinations.*—All negative.

7. *Mule 2489.*—Two-and-a-half-year-old Argentine mare.

*Injected* as above.

*Temperature.*—No distinct reaction until the 20th day after injection, when the temperature rose from 100 and recorded 105.4 in the evening of the 22nd day. It now fell, and 100.2 was noted in the morning of the 26th day, and 106 in the evening of the 29th day.

*Examinations.*—All negative.

8. *Mule* 2490.—Seven-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—Slight reaction.

9. *Mule* 2491.—Five-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—Very slight reaction, recording 103.4 on the 10th day from injection.

10. *Mule* 2492.—Aged Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction from the 9th day, recording 104 on the 17th and 18th days. A second reaction from the 25th day, and reaching the same figure on the 27th and 28th days.

11. *Mule* 2493.—A three-and-a-half-year-old Argentine mare.

*Injected* on the 7th February, 1907, subcutaneously with 3 c.c. defibrinated fresh blood of foal 2314.

*Temperature.*—Slight reaction.

12. *Mule* 2500.—Argentine gelding.

*Injected* as above.

*Temperature.*—Slight reaction, reaching 103 on the 18th and 19th days.

13. *Mule* 2501.—Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction, reaching 104.8 on the 21st and 22nd days.

14. *Mule* 2502.—Argentine mare.

*Injected* as above.

*Temperature.*—Slight reaction, reaching the maximum of 104.4 on the 24th day.

Mules 2503, 2504, 2505, 2506, 2507 injected subcutaneously with 3 c.c. defibrinated fresh blood of Transvaal horse foal 2314 on the 14th February, 1907.

15. *Mule* 2503.—Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction, recording 104.8 on the 20th day.

16. *Mule* 2504.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction, reaching 104 on the 4th day after injection. A second reaction noted from the 27th to 32nd days.

17. *Mule* 2506.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 14th day, and recording the maximum of 104.2 on the 24th day after injection.

18. *Mule* 2507.—Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction from the 7th day, reaching 103.4 on the 11th day. Second reaction from the 14th day, lasting for 11 days.

*Note.*—The following mules, all directly imported from the Argentine, were injected on the 12th February, 1907, subcutaneously with 3 c.c. defibrinated fresh blood of foal 2314:—Mules Nos. 2581, 2588, 2537, 2589, 2590, 2591, 2592, 2594, 2595, and 2596. Further, they had been previously utilised for horse-sickness experiments.

19. *Mule* 2581.—Three-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—Reaction from the 7th day.
20. *Mule* 2588.—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—No distinct reaction.
21. *Mule* 2537.—Argentine stallion.  
*Injected* as above.  
*Temperature.*—Reaction from the 9th day.
22. *Mule* 2589.—Two-and-a-half-year old mare.  
*Injected* as above.  
*Temperature.*—Reaction from the 13th to 23rd days, and reaching the maximum of 105.2 on the 20th day.  
*Examinations.*—All negative.
23. *Mule* 2590.—Four-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—No reaction.
24. *Mule* 2591.—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—Slight reaction from the 15th day, and recording the maximum of 103.8 in the 26th day after injection.
25. *Mule* 2592.—Three-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—Reaction from the 10th day, and recording 104.6 on the 22nd day. A secondary reaction from the 30th day.
26. *Mule* 2594.—Argentine gelding.  
*Injected* as above.  
*Temperature.*—Reaction from the 5th day; second from the 10th day.
27. *Mule* 2595.—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—Doubtful reaction.
28. *Mule* 2596.—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—Slight reaction.

The following Argentine mules were all injected on the 29th January, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2314:—Nos. 2526, 2527, 2528, 2529, 2573, 2574, 2575, 2576, 2577.

29. *Mule* 2526.—Argentine stallion.  
*Injected* as above.  
*Temperature.*—Reaction from the 6th day.

30. *Mule 2527*.—Argentine mare.  
*Injected as above.*  
*Temperature*.—Slight reaction from the 8th day.
31. *Mule 2528*.—Argentine gelding.  
*Injected as above.*  
*Temperature*.—Reaction from the 8th day.  
*Examinations*.—All negative.
32. *Mule 2529*.—Argentine gelding.  
*Injected as above.*  
*Temperature*.—Reaction from the 10th day.  
*Examinations*.—All negative.
33. *Mule 2573*.—Argentine mare.  
*Injected as above.*  
*Temperature*.—Doubtful reaction.  
*Examinations*.—*Piroplasma equi* noted in rare numbers on the 14th and 15th days.
34. *Mule 2574*.—Argentine mare.  
*Injected as above.*  
*Temperature*.—Reaction from the 8th day.  
*Examinations*.—*Piroplasma equi* noted for the first time, but in rare numbers on the 19th day after injection.
35. *Mule 2575*.—Argentine mare.  
*Injected as above.*  
*Temperature*.—Reaction from the 7th day.  
*Examinations*.—Slight poikilocytosis noticed on the 14th and 15th days.
36. *Mule 2576*.—Argentine mare.  
*Injected as above.*  
*Temperature*.—Slight reaction from the 6th day.  
*Examinations*.—*Piroplasms* noted on the 13th day only.
37. *Mule 2577*.—Argentine mare.  
*Injected as above.*  
*Temperature*.—Distinct reaction from the 7th day, recording 103.6 on the 14th and 15th days.  
*Examinations*.—*Piroplasms* in rare numbers noted for the first time on the 13th day, and the following two days were again present, accompanied with the lesions of poikilocytosis.
- Result of 1 Transvaal horse foal and 36 Argentine mules injected with blood of a Transvaal horse foal, immune against piroplasma equi (third generation).*  
 All passed through a reaction and recovered.

## EXPERIMENT No. 7.

*Fifth Generation.*

*Transvaal horse foal injected with blood of a Transvaal horse foal, immune against piroplasma equi.*

1. *Horse Foal 2274*.—Filly born on the station in September, 1906.  
*Injected on the 28th January, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2273,*

*Temperature.*—Reaction from the 6th day, reaching 103.6 on the 10th day. A second reaction from the 17th day, lasting for 10 days.

*Examinations.*—All negative.

*Note.*—The following Argentine mules were all injected on the 29th January, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2273, now immune against *piroplasma equi*:—Nos. 2530, 2533, 2566, 2567, 2568, 2569, 2570, 2571 and 1572.

2. *Mule* 2530.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 6th day.

3. *Mule* 2533.—Argentine mare.

*Injected* as above.

*Temperature.*—The morning record on the 7th day was 97.8, rising to 101.4 in the evening of the 9th day, but dropped during the night, and the animal died the following morning from pneumonia.

4. *Mule* 2566.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 7th day, lasting for 14 days.

*Examinations.*—Slight poikilocytosis noted on the 12th and 13th days. *Piroplasma equi* present in rare numbers on the 14th day.

5. *Mule* 2567.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 6th day, recording 105.6 in the evening of the 14th day.

*Examinations.*—Slight poikilocytosis noted on the 11th day, and the following day *piroplasma equi* present. Piroplasms again present in rare numbers on the 15th day.

6. *Mule* 2568.—Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction from the 7th day.

*Examinations.*—*Piroplasma equi* noted on the 14th day, and the lesions of poikilocytosis present the following day.

7. *Mule* 2569.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 5th day, recording 103.6 four days later.

*Examinations.*—All negative.

8. *Mule* 2570.—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 4th day. Second rise noted from the 14th to 22nd days.

*Examinations.*—*Piroplasma equi* noted on the 12th day, and the lesions of poikilocytosis the following day. *Piroplasma equi* again noted during the second reaction, and followed on the 19th day by the lesions of a slight poikilocytosis.

9. *Mule 2571.*—Argentine gelding.

*Injected* as above.

*Temperature.*—Reaction from the 4th day, lasting for 25 days, and recording between 100 and 105.2, this latter record being noted on the 19th day.

*Examinations.*—*Piroplasma equi* noted on the 13th, 14th and 15th days; on this latter date the lesions of poikilocytosis also being present. On the 14th day the urine was coloured red, but became clear two days later. On the 15th day the red corpuscles numbered 3,700,000 per c.m.m., and 24 hours later decreased to 2,500,000. Slight poikilocytosis noted on the 18th and 19th days, and again on the 24th day.

10. *Mule 2572.*—Argentine mare.

*Injected* as above.

*Temperature.*—Reaction from the 5th day, reaching 104.6 in the evening four days later, and regaining normal on the 11th day. A second reaction from the 12th day, and lasting for six days.

*Examinations.*—*Piroplasms* present in fair numbers on the 13th day, and again noted on the following two days.

*Result of 1 Transvaal horse foal and 9 Argentine mules injected with blood of a Transvaal horse foal, immune against piroplasma equi.*

All passed through a reaction, and, with the exception of one Argentine mule, which died of pneumonia, recovered.

*Note.*—In the majority of cases these reactions were of a severe character; secondary reactions were also noted, and *piroplasma equi* was frequently present.

## EXPERIMENT NO. 8.

*Sixth Generation.*

*Injection* with blood of Transvaal foal 2274, immune against *piroplasma equi*. (Compare Experiment 7, 1.)

*Transvaal Horse Foal 2621.*—Colt obtained from the S.A.C.

*Injected* on the 26th March, 1907, subcutaneously with 5 c.c. defibrinated blood of foal 2274.

*Temperature.*—Reaction from the 6th day.

*Examinations.*—The lesions of poikilocytosis noted on the 11th, 14th and 15th days, and on the 16th day *piroplasma equi* appeared.

*Result.*—One Transvaal horse foal inoculated with blood of a horse foal, immune against *piroplasma equi* (fifth generation), passed through a *piroplasmosis* reaction and recovered.

## EXPERIMENT NO. 8A.

*Seventh Generation.*

*Transvaal horse foal injected with blood of Transvaal foal 2621, immune against piroplasma equi.*

*Transvaal Horse Foal 2734.*—

*Injected* subcutaneously on the 25th April, 1907, with 10 c.c. blood of foal 2621.



*Temperature.*—Reaction from the 8th day. Second reaction from the 26th day, recording 105.6 on the 39th day.

*Examinations.*—*Piroplasma equi* noted on the 9th day, and again on the 40th day.

*Argentine horse injected with blood of Transvaal foal 2621, immune against piroplasma equi.*

*Horse 2684.*—Four-year-old Argentine mare.

*Injected* on the 8th May, 1907, with 5 c.c. defibrinated fresh blood of foal 2621.

*Temperature.*—Reaction from the 7th day, rising to 104.2 in the evening of the 10th day, falling to sub-normal 24 hours later, and recording 97. On the 13th day temperature reached 105.6 in the evening, but the animal died the following morning from debility, complicated with piroplasmosis.

*Post-mortem Examination.*—

*Condition:*—Poor; blood-stained foam in nostrils; flesh of a brick-red colour.

*Lungs:*—Slight œdema; liquid in peritoneal cavity.

*Heart:*—Inhibition of left and right endocardis; myocard soft; abnormal amount of blood-stained liquid in pericard.

*Spleen:*—Slightly enlarged; pulpa soft.

*Liver:*—Decomposed.

*Kidneys:*—Pale and yellowish.

*Stomach:*—Mucosa pale.

*Intestines:*—Cæcum pale and slate coloured; colon slate coloured; *strongylus armatus* and *tetracanthus* present.

*Result.*—One Transvaal horse foal and one Argentine mare injected with blood of a Transvaal horse foal (sixth generation); both passed through a typical piroplasmosis reaction, the Argentine mare dying of debility, complicated with piroplasmosis.

#### EXPERIMENT No. 8B.

##### *Eighth Generation.*

*Transvaal horse foal injected with blood of Transvaal foal 2734, now immune against piroplasma equi.*

*Horse Foal 2786.*—Transvaal gelding.

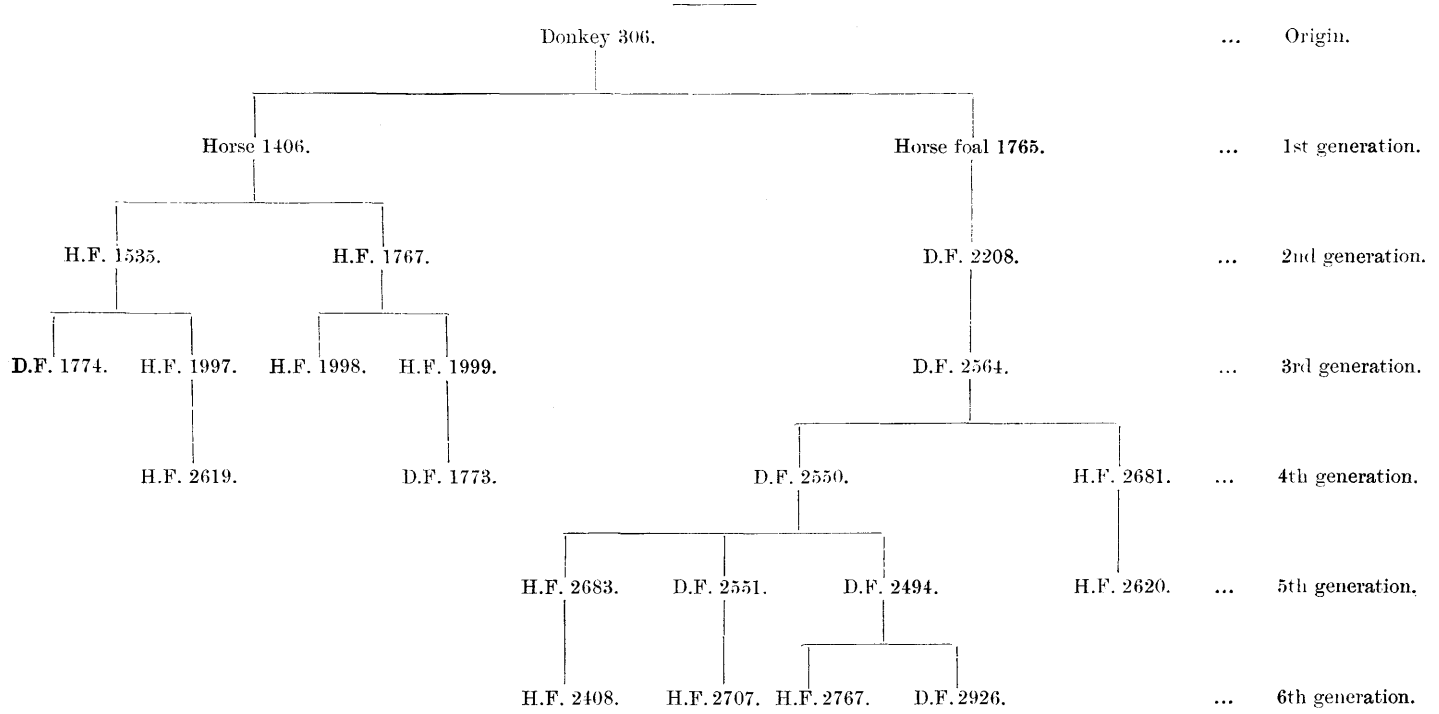
*Injected* subcutaneously on the 30th January, 1907, with 5 c.c. blood of foal 2734.

*Temperature.*—Reaction from the 4th day. Second reaction from the 23rd day, and lasting for 11 days.

*Examinations.*—Poikilocytosis noted on the 5th, 6th, 11th and 12th days, and *piroplasma equi* on the 7th, 8th and 9th days.

*Result.*—One Transvaal horse foal injected with blood of a Transvaal horse foal, immune against *piroplasma equi* (seventh generation), passed through a piroplasmosis reaction and recovered. A second reaction was also noticed.

3. INOCULATION AGAINST EQUINE PIROPLASMOSIS BY MEANS OF HORSE FOAL AND DONKEY FOAL BLOOD.  
 ORIGIN DONKEY BLOOD.



H.F.—Horse foal.      D.F.—Donkey foal.

3.—INOCULATION BY MEANS OF DONKEY FOAL AND HORSE FOAL BLOOD.  
ORIGIN DONKEY BLOOD.

*Donkey 306.* (Compare Annual Report 1904-5, page 101.)

EXPERIMENT No. 9.

*First Generation.*

1. *Horse 406.*—About three years old, injected with 10 c.c. blood of donkey 306. Compare Annual Report 1905-6, page 90.

2. *Horse foal 1765.*—

*Injected* on the 21st April, 1906, subcutaneously with 5 c.c. blood of donkey 306.

*Result.*—Reaction from the 9th day, reaching 103 five days later and remaining normal from the 19th day. A slight rise noted from the 44th day, recording 103.4 the following day. Piroplasms noted for the first time, but in exceedingly rare numbers, on the 18th day. Again present on the 21st day, and together with rosettes, rings and points were occasionally noted during the next three weeks. At the time of the sharp rise on the 55th day, the mucous membranes were noted to be dirty and slightly injected.

*Red Corpuscles.*—

Count on 7th day ...	...	8,900,000	per c.m.m.
„ 10th „ ...	...	9,190,000	„
„ 12th „ ...	...	7,776,000	„
„ 14th „ ...	...	9,040,000	„
„ 20th „ ...	...	8,496,000	„
„ 22nd „ ...	...	7,520,000	„
„ 25th „ ...	...	7,240,000	„
„ 28th „ ...	...	7,904,000	„

*Argentine horses injected with blood of donkey 306—immune against piroplasma equi.*

Argentine horses 2229, 2231, 2238, 2239, 2243, 2244 and 2245.—These animals had previously been inoculated with blood of a foal immune against piroplasma equi, and were also utilised for horse-sickness experiments. They were now tested on their immunity against piroplasma equi.

Argentine horses 2229 and 2245 previously inoculated with blood of Transvaal foal 1535—compare Experiment 11, Nos. 7 and 8—and now injected on the 21st December, 1906, subcutaneously with 5 c.c. blood of donkey 306.

3. *Horse 2229.*—Three-year-old Argentine gelding.

*Injected* as above.

*Result.*—Reaction from the 7th day. All examinations negative.

4. *Horse 2245.*—Five-year-old Argentine gelding.

*Injected* as above.

*Result.*—Reaction from the 14th day, recording 105 in the following evening. A few points noted on the 5th day.

Argentine horses 2231, 2238, and 2239, previously inoculated with blood of Transvaal horse foals [compare Experiments 5 (8) and 10 (27

and 28)] now tested on their immunity. Injected on the 19th December, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of donkey 306.

5. *Horse 2231*.—Four-year-old Argentine mare.

*Injected* as above.

*Result*.—Slight reaction from the 13th day. All blood examinations negative.

6. *Horse 2238*.—Argentine three-year-old gelding.

*Injected* as above.

*Result*.—Slight reaction. One point present on the 16th day.

7. *Horse 2239*.—Seven-year-old gelding.

*Injected* as above.

*Result*.—Slight reaction. One point present on the 16th day.

Argentine horses 2243 and 2244 previously injected with blood of foal 2208 (compare Experiment 10, Nos. 30 and 29) and now injected on the 12th December, 1906, subcutaneously with 5 c.c. fresh defibrinated blood of donkey 306.

8. *Horse 2243*.—Four-year-old Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction from the 12th day. All examinations negative.

9. *Horse 2244*.—Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction. Poikilocytosis and a few points noted.

*Argentine mules injected with blood of donkey 306—immune against piroplasma equi.*

Argentine mules Nos. 2322 and 2325 were previously injected on the 5th November, 1906, with blood of foal 2053, immune against piroplasma equi (compare Experiment 5, Nos. 4 and 6), and mule 2326 was injected on the same date with blood of foal 1999, immune against piroplasma equi (compare Experiment 12, No. 46), now tested on their immunity; three were injected on the 19th December, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of donkey 306.

10. *Mule 2322*.—Two-and-a-half-year-old Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction from the 15th day, piroplasms and the lesions of a slight poikilocytosis noted on the 11th and 18th days. Slight poikilocytosis again present on the 21st day.

11. *Mule 2325*.—Aged Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction. Poikilocytosis and points noted on the 13th day; the former again present on the 23rd day. Piroplasma equi not present.

*Argentine donkeys, injected with blood of donkey 306—immune against piroplasma equi.*

Donkeys 2248 and 2249 were injected on the 26th November, 1906, with blood of foal 2054, immune against piroplasma equi, and donkey 2254 was injected on the same date with blood of foal 1997,

immune against piroplasma equi. All three donkeys were now injected on the 19th December, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of donkey 306.

13. *Donkey 2248*.—Four-year-old Argentine mare.

*Injected* as above.

*Result*.—Reaction from the 11th day. Piroplasms present on the 20th and 22nd days.

14. *Donkey 2249*.—Four-year-old Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction from the 15th day, reaching 103 twenty-four hours later. All microscopical examinations negative.

15. *Donkey 2254*.—Three-year-old Argentine mare.

*Injected* as above.

*Result*.—Reaction from the 15th day. Piroplasms present on the 16th day.

*Results*.—One Transvaal horse foal injected with blood of a Transvaal donkey, immune against piroplasma equi, passed through a reaction and recovered. Seven Argentine horses, three Argentine mules, and three Argentine donkeys were tested on their immunity, and all showed slight reactions and recovered.

#### EXPERIMENT No. 10.

##### *Second Generation.*

*Transvaal horse foal injected with blood of Argentine horse—immune against piroplasma equi.*

1. *Transvaal Foal 1535*.—About seven months old. (Note.—This foal was utilised in a horse-sickness experiment in January, 1906.)

*Injected* subcutaneously on the 19th April, 1906, with 5 c.c. blood of Argentine horse 1406.

*Temperature*.—Reaction from the 8th day, rising to 105.6 sixteen days after injection, and remaining high for the next four days. The temperature remained normal from the 28th day until the 37th day, when a rise from 100 in the morning to 104.8 in the evening was noted.

*Examinations*.—Piroplasms noted for the first time, but in rare numbers, on the 18th day, and again on the 22nd day. Piroplasms, rosettes, marginal points, and the leaf form were occasionally noted until the 54th day after injection. The mucous membranes were yellow on the 28th day, and slightly pale on the 45th day. On the 37th day, the occasion of the sharp rise referred to above, the animal was affected with a nasal catarrh.

*Red Corpuscles*.—The blood count on the 9th day recorded 8,943,000, falling to 7,232,000 four days later, and on the 21st day recorded 5,568,000 per c.m.m.

2. *Foal 1767*. (Note.—This animal had been utilised in a horse-sickness experiment in January, 1906.)

*Injected* on the 19th June, 1906, subcutaneously with 5 c.c. fresh defibrinated blood of horse foal 1406.

*Temperature*.—Reaction from the 7th day, reaching 104 in the evening four days later, but falling to 100 on the 18th day. A short rise noted from the 23rd to 27th days, but only reaching 103.

*Examinations.*—The pear and ring form of *piroplasma equi* noted on the 13th day, and a few more seen seven days later. Piroplasms were frequently noted during the next ten days, but they appeared for the last time on the 34th day. Points were noted on the 34th day, and again on the 50th day.

*Red Corpuscles.*—

Count of 9th day	...	...	9,600,000	per c.m.m.
„ 13th	„	...	6,320,000	„
„ 15th	„	...	5,600,000	„
„ 19th	„	...	6,816,000	„
„ 21st	„	...	6,260,000	„
„ 23rd	„	...	6,400,000	„
„ 26th	„	...	5,610,000	„
„ 29th	„	...	7,260,000	„

*Transvaal donkey foal injected with blood of horse foal 1765—  
immune against piroplasma equi.*

3. *Donkey Foal 2208.*—Six months old and born on the station.

*Injected* on the 14th September, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of horse foal 1765.

*Temperature.*—Reaction from the 10th day, but only recording as a maximum 103.2 on the 17th day. Second reaction noted on the 32nd day, recording 103 six days later.

*Examinations.*—All negative.

*Argentine horses injected with blood of Transvaal horse foal 1765—  
immune against piroplasma equi.*

4. *Horse 2073.*—Two-year-old Argentine mare.

*Injected* subcutaneously on the 9th July with 5 c.c. defibrinated fresh blood of foal 1765.

*Temperature.*—Slight reaction from the 11th to 20th days, the temperature recording 104.6 on this latter date. A second reaction from the 24th day, recording 105 six days later, and regaining normal on the 34th day.

*Examinations.*—Piroplasms noted, but in very rare numbers on the 12th day, accompanied with rings. Piroplasms present the following day, and again on the 17th day. Points noted on the 16th and 18th days; the mucous membranes pale on the 18th day. Piroplasms again noted during the second reaction, on the 27th day, and one point noted on the 31st day.

5. *Horse 2074.*—Two-year-old Argentine mare.

*Injected* subcutaneously on the 9th July, 1906, with 5 c.c. blood of foal 1765.

*Temperature.*—Very slight reaction. The animal died on the 31st day from rupture of spleen.

*Examination.*—Piroplasms noted for the first time on the 12th day, accompanied with rings and points; the latter were also present on the 16th and 17th days, and the following day piroplasms were again noted. One piroplasm and one point were present on the 21st day, and on the following day, when the animal died, smears were made of the spleen, but not piroplasms were seen.

*Post-mortem.*—

*Condition*:—Good; body tympanitic.

*Lungs*:—Several gallons of blood in peritoneal cavity.

*Spleen*:—Anterior surface shows a ragged rupture about three inches long; spleen enlarged about three times normal; pulpa dark brown, but firm.

All other organs normal in appearance, but pale.

*Transvaal horse injected with blood of Transvaal horse foal 1765—immune against piroplasma equi.*

6. *Horse 2205.*—Aged gelding. (Note.—This animal had previously been utilised in horse-sickness experiments.)

*Injected* on the 14th September subcutaneously with 5 c.c. defibrinated fresh blood of foal 1765.

*Result.*—No distinct reaction.

*Results of animals inoculated with blood of a Transvaal horse foal—immune against piroplasma equi (first generation).*

1 Transvaal donkey foal showed a reaction and recovered.

Of 2 Argentine horses one showed a reaction and recovered, and one died from rupture of spleen.

1 Transvaal horse—no distinct reaction.

2 Transvaal horse foals injected with blood of a Transvaal horse (first generation) showed reactions and recovered.

## EXPERIMENT NO. 11.

*Third Generation.*

*Transvaal donkey foal injected with blood of Transvaal horse foal 1535—immune against piroplasma equi.*

1. *Donkey Foal 1774.*—Eighteen-month-old mare, and born on station. (Note.—This donkey foal had been utilised in horse-sickness experiments in August, 1906.)

*Injected* on the 14th September, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 1535.

*Result.*—Reaction from the 12th day.

All examinations negative.

*Transvaal horse foals injected with blood of Transvaal horse foal—immune against piroplasma equi.*

Transvaal horse foals 1997, 1998 and 1999—all six-month-old mares—were injected on the 9th July, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 1535.

2. *Horse Foal 1997.*—

*Injected* as above.

*Result.*—Reaction from the 26th to the 41st day. Piroplasms noted on the 15th, 16th, 19th, 20th and 27th days, and circular points noted on the 22nd day.

3. *Horse Foal 1998.*—

*Injected* as above.

*Result.*—Reaction from the 19th day. Piroplasms noted on the 13th, 15th, 16th, 21st, 22nd, 28th, 36th and 37th days.

4. *Horse Foal 1999.*—

*Injected as above.*

*Result.*—High temperature at date of injection. Reaction from the 13th day. Piroplasms present on the 15th, 18th and 21st days, and on this latter date were accompanied with rings and points.

*Argentine horses injected with blood of Transvaal horse foal—immune against piroplasma equi.*

Horses 2061 and 2062, both two-year-old Argentine mares, were injected on the 16th June, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 1535.

5. *Horse 2061.*—

*Injected as above.*

*Result.*—Reaction from the 14th day, reaching 104 on the 17th day and 105 on the 30th day. Piroplasms, rings and the lesions of poikilocytosis noted on the 18th day.

6. *Horse 2072.*—

*Injected as above.*

*Result.*—Reaction from the 11th day, recording 105 on the 11th, 13th and 14th days. The temperature now remained fairly high until the 22nd day, and two days later a sharp drop to sub-normal of 97.6 was recorded. The animal now rallied, but after the temperature reached 105 on the 29th day, the animal was killed on account of pleuro-pneumonia. Piroplasms, accompanied with rings, noted on the 12th, 13th, 15th, 20th and 22nd days; mucous membranes pale; the animal very weak on the 18th day.

*Note.*—Horses 2229 and 2245 were inoculated with horse-sickness blood on the 3rd September, 1906, and now injected subcutaneously with 5 c.c. defibrinated fresh blood of foal 1535 on the 16th October, 1906.

7. *Horse 2229.*—Three-year-old gelding.

*Injected as above.*

*Result.*—Reaction from the 16th day, recording 105 on the 23rd, 24th and 25th days, and regaining normal on the 33rd day. Slight yellowish appearance of the eyes and general weakness noted on the 6th day, but followed by an improvement within 24 hours. Ring forms present on the 11th day, and piroplasms noted on the 12th, 14th and 15th (accompanied with the lesions of poikilocytosis on these latter two days), 16th, 17th, 18th, 19th (on which date one rosette was also noted), 22nd, 23rd and 24th. Points present on the 28th day, and eight days later piroplasms again observed.

The lowest records of the red corpuscle were noted on the 30th day—4,700,000, and on the 39th day 4,600,000 per c.m.m.

8. *Horse 2245.*—Three-year-old Argentine gelding.

*Injected as above.*

*Result.*—Reaction from the 10th day, and lasting for 20 days. Short reaction noted from the 32nd to 37th days. Points noted on the 7th day, accompanied with the lesions of weakness and yellowish eyes. Piroplasms noted on the 15th, 16th, 17th and 18th days, but on the latter two days in very rare numbers. The lesions of poikilocytosis, points and piroplasms occasionally noted from the 21st to 38th days. The red corpuscles reached the minimum record of 4,200,000 per cmm. on the 38th day.



*Argentine donkeys injected with blood of Transvaal horse foal—  
immune against piroplasma equi.*

Argentine donkeys 1846, 1847, 1848, 1849 and 1850, injected on the 16th June, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of foal 1535.

9. *Donkey* 1846.—Three-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Slight irregular reaction.
10. *Donkey* 1847.—Three-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Slight reaction.
11. *Donkey* 1848.—Four-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Slight reaction.
12. *Donkey* 1849.—Two-year-old Argentine gelding  
*Injected* as above.  
*Result.*—Reaction from the 8th day.
13. *Donkey* 1850.—Three-year-old Argentine mare.  
*Injected* as above.  
*Result.*—(No record kept).

Argentine donkeys 2259, 2260, 2261, all three-year-old Argentine mares, and previously utilised for horse-sickness experiments, were injected on the 16th October, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 1535.

14. *Argentine Donkey* 2259.—  
*Injected* as above.  
*Result.*—Reaction from the 11th day. Chromatic points noted on the 5th and 7th days, and piroplasms—but in rare numbers—on the 15th, 16th and 17th days. The lesions of poikilocytosis present on the 16th, 21st and 26th days.
15. *Argentine Donkey* 2260.—  
*Injected* as above.  
*Results.*—Reaction from the 3rd day. Points noted on the 5th, 7th, 10th and 11th days; on this latter date piroplasms were also present. Piroplasms and the lesions of poikilocytosis occasionally noted until the 26th day.
16. *Argentine Donkey* 2661.—  
*Injected* as above.  
*Result.*—Irregular reaction. Piroplasms noted on the 10th, 11th, 14th, 15th, 17th and 21st days. Points present on the 16th day and the lesions of poikilocytosis appeared on the 22nd and 24th days.

*Transvaal mules injected with blood of Transvaal horse foal—  
immune against piroplasma equi.*

17. *Transvaal Mule* 2211.—Three-year-old mare, and previously utilised for horse-sickness experiments.

*Injected* on the 16th October, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of foal 1535.

*Result.*—Irregular reaction. Piroplasms and flagellated forms noted on the 10th day; the former again present on the 11th, 14th, 15th, 18th, 22nd and 23rd days.

Transvaal mules 2213, 2214, 2215 and 2217—previously utilised for horse-sickness experiments—were injected on the 21st September, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 1535.

18. *Mule 2213.*—Three-year-old Transvaal mare.

*Injected as above.*

*Result.*—Reaction from the 15th day, recording 106 nine days later. A second reaction commenced on the 32nd day, and terminated by the death of the animal seven days later from piroplasmosis (29th October, 1906). Piroplasms noted on the 13th, 15th and 16th days. Rings, points and the lesions of poikilocytosis occasionally noted. The pear-shaped piroplasm was observed on the 23rd day.

*Post-mortem.*—

*Condition:*—Fair; blood watery, brownish in colour.

*Lungs:*—Very pale; mediastinal gland swollen and congested.

*Heart:*—Normal.

*Spleen:*—Slightly swollen; splenic lymphatic glands swollen and deeply congested.

*Liver:*—Swollen slightly.

*Kidneys:*—One infarct size of a threepenny piece in left kidney.

*Stomach:*—Some erosions on mucous membranes.

*Intestines:*—Nil.

*Bladder:*—Distended with blood-coloured urine.

19. *Mule 2214.*—Four-year-old gelding.

*Injected as above.*

*Result.*—Reaction from the 18th day. Piroplasms present on the 15th, 16th and 18th days. Rings, rods and the lesions of poikilocytosis also noted.

20. *Mule 2215.*—Four-year-old gelding.

*Result.*—Reaction from the 13th to 34th days. Piroplasms noted on the 13th, 15th and 16th days. Rods, rings, points and the lesions of poikilocytosis also present.

21.—*Mule 2217.*—Three-year-old mare.

*Injected as above.*

*Result.*—Slight reaction from the 16th day. Piroplasms noted on the 13th, 16th, 22nd, 23rd and 26th days. Rings, rods, points and the lesions of poikilocytosis also present.

*Argentine horse injected with blood of Transvaal horse foal—immune against piroplasma equi.*

22. *Horse 2095.*—Four-year-old Argentine mare.

*Injected on the 9th July, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of foal 1767.*

*Result.*—Rise to 102 noted in the evening of the 11th day, and followed by a drop to sub-normal of 96.4. The temperature now recovered, recorded 106 in the evening of the 23rd day, and remained normal from the 31st day. Piroplasms noted on the 11th and 12th days, and again daily from the 14th to 17th days. From the 31st day to the 56th day piroplasma equi was frequently noted, occasionally accompanied with points and the lesions of poikilocytosis,

23. *Horse* 2096.—Three-year-old Argentine mare.

*Injected* on the 9th July, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 1767.

*Result*.—The 14th day after injection marked the commencement of a reaction, the temperature reached 102.6 two days later, but the animal died the following evening from rupture of spleen. Piroplasms noted on the 12th and 15th days. The examination of the animal about 12 hours previous to death showed the hind-quarters to be slightly weak and the mucous membranes slightly yellow.

*Post-mortem*.—

*Condition*:—Good; uterus pregnant.

*Lungs*:—Normal.

*Heart*:—Normal.

*Stomach*:—Normal.

*Kidneys*:—Pale.

*Liver*.—Pale.

*Spleen*.—A rupture about four inches long on anterior surface.

*Abdominal Cavity*:—Full of blood.

*Argentine mules injected with blood of Transvaal horse foal—immune against piroplasma equi.*

Mules 2513 and 2516 were previously utilised for horse-sickness experiments on the 12th January, 1907, and injected on the 29th January, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal horse foal 1767.

24. *Mule* 2513.—Argentine gelding.

*Injected* as above.

*Result*.—No distinct reaction.

25.—*Mule* 2516.—Argentine mare.

*Injected* as above.

*Result*.—Sharp rise from the 4th day, and regaining normal three days later. Reaction from the 16th day. All microscopical examinations negative.

*Transvaal donkey foal injected with blood of Transvaal donkey foal—immune against piroplasma equi.*

26. *Donkey Foal* 2564.—Three months old; born on the station.

*Injected* on the 4th January, 1907, subcutaneously with 5 c.c. blood of donkey foal 2208.

*Result*.—No distinct reaction until the 23rd day. All examinations negative with the exception of the presence of a slight poikilocytosis on the 18th day.

*Argentine horses injected with blood of Transvaal donkey foal—immune against piroplasma equi.*

*Note*.—The following three horses, Nos. 2238, 2239 and 2241, were all injected subcutaneously on the 5th November, 1906, with 5 c.c. defibrinated fresh blood of donkey foal 2208.

27. *Horse* 2238.—Three-year-old Argentine gelding.

*Injected* as above.

*Temperature*.—Slight reaction from the 4th day, reaching 103 on the 9th day, and regaining normal on the 15th day,

*Examinations.*—Points noted on the 11th and 14th days, and 25th day piroplasms, accompanied with the lesions of a slight poikilocytosis, present on the 33rd day, the latter also being present the following day.

28. *Horse 2239.*—Seven-year-old Argentine gelding.

*Injected* as above.

*Temperature.*—No distinct reaction, the temperature only reaching the maximum of 102.2 on the 16th day.

*Examinations.*—One point noted on the 15th day, and two days later piroplasma equi was present. The lesions of a slight poikilocytosis were present on the 22nd day, and nine days later piroplasma equi again appeared.

29. *Horse 2244.*—Argentine gelding.

*Injected* as above.

*Temperature.*—No distinct reaction.

*Examinations.*—One point noted on the 10th day, and piroplasma equi present on the 13th day. A few points were again noted the following day and on the 24th day. Piroplasma equi again noticed on the 26th and 27th days.

*Argentine horse injected with blood of Transvaal donkey—immune against piroplasma equi.*

30. *Horse 2243.*—Four-year-old Argentine gelding.

*Injected* on the 16th October, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of donkey foal 2208.

*Results.*—Slight reaction from the 6th day, and second reaction from the 21st day, recording the maximum of 103.2 three days later. Points noted on 5th day; the animal was noted to be generally weak on the 7th day, with slightly yellow eyes, but an improvement was noted the following day. Points again noted on the 10th day, and piroplasms present in fair numbers the following day. Piroplasms now noted daily from the 14th to the 18th day, one rosette being present on the 16th day. Flagellated forms, points, forms, piroplasms, rosettes and the lesions of poikilocytosis frequently noted from the 19th to 33rd days. Piroplasms again seen on the 39th and 45th days. The lowest record of red corpuscles was 5,900,000 per c.m.m. on the 35th day.

*Transvaal mule injected with blood of Transvaal donkey foal—immune against piroplasma equi.*

31. *Mule 2223.*—Three-year-old mare. (Note.—Had previously been utilised in horse-sickness experiments.)

*Injected* on the 16th October, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal donkey foal 2208.

*Result.*—Irregular reaction, recording, as the maximum temperature, 102.4 on the 21st day. Points noted on the 7th, 10th and 11th days. The lesions of poikilocytosis appeared on the 14th day, and from the 15th to the 18th days piroplasms were noted daily.

*Argentine donkeys injected with blood of Transvaal donkey foal—immune against piroplasma equi.*

Donkeys 2262 and 2263, both Argentine mares about two years old, had been utilised in horse-sickness experiments in September,

1906, and were injected on the 16th October, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal donkey foal 2208.

32. *Donkey 2262.*—

*Injected* as above.

*Result.*—Irregular reaction. The maximum temperature of 104 obtained on the 7th day. Rings noted on the 10th day, and following by the appearance of piroplasms for two days. Piroplasms and poikilocytosis noted from the 14th to the 18th days.

33. *Donkey 2263.*—

*Injected* as above.

*Result.*—Irregular reaction. Piroplasms present on the 11th, 12th, 13th, 15th and 16th days. The lesions of poikilocytosis noted on the 15th, 19th and 26th days.

*Results of animals inoculated with blood of Transvaal horse foals—immune against piroplasma equi (second generation).*

3 Transvaal horse foals passed through the disease and recovered.

1 Transvaal donkey foal passed through the disease and recovered.

5 Transvaal mules passed through the disease, and one died of piroplasmosis.

6 Argentine horses passed through the disease, one being subsequently killed on account of pleuro-pneumonia, and one dying from rupture of spleen.

8 Argentine donkeys passed through the disease and recovered.

2 Argentine mules passed through the disease and recovered.

*Results of animals inoculated with blood of a Transvaal donkey foal—immune against piroplasma equi (second generation).*

1 Transvaal mule passed through a reaction and recovered.

1 Transvaal donkey foal passed through a reaction and recovered.

4 Argentine horses passed through a reaction and recovered.

2 Argentine donkeys passed through a reaction and recovered.

#### EXPERIMENT No. 12.

##### *Fourth Generation.*

*Transvaal horse foal injected with blood of foal 1997—immune against piroplasma equi.*

1. *Transvaal Horse Foal 2619.*—About four months old.

*Injected* subcutaneously on the 2nd February, 1907, with 5 c.c. defibrinated fresh blood of foal 1997.

*Result.*—Reaction from the 2nd day, reaching 106.6 on the 12th day, the evening temperature remaining high for the next four days. A second sharp rise was noted from the 19th day, the temperature recording 106.6 twenty-four hours later. Piroplasma equi noted on the 9th day. All other examinations negative.

*Argentine mules injected with blood of foal 1997—immune against piroplasma equi.*

*Note.*—The following mules (Nos. 2363, 2364, 2365, 2366 and 2367) were all injected subcutaneously on the 26th November, 1906, with 5 c.c. defibrinated fresh blood of foal 1997.

2. *Mule 2363.*—Four-year-old Argentine mare.

*Injected* as above.

*Result.*—Reaction from the 10th day. All microscopical examinations negative,

3. *Mule* 2364.—Five-year-old Argentine gelding.  
*Injected* as above.  
*Result.*—Reaction from the 10th day. The lesions of a slight poikilocytosis noted on the 16th day, and again on the 19th day, accompanied with points.
  4. *Mule* 2365.—Four-year-old Argentine gelding.  
*Injected* as above.  
*Result.*—No distinct reaction. Poikilocytosis and points observed; piroplasma equi not present.
  5. *Mule* 2366.—Three-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from date of injection. Piroplasms noted on the 13th and 16th days, and poikilocytosis the 19th day.
  6. *Mule* 2367.—Eighteen-month-old Argentine gelding.  
*Injected* as above.  
*Result.*—Very slight reaction from the 14th day. Piroplasms noted, but in very rare numbers, on the 16th day.
- Note.*—The following mules (Nos. 2508, 2509, 2511, 2512, 2534, 2535, 2536, 2538, 2540, 2541, 2542, 2543, 2544, 2545, 2546) were all injected subcutaneously on the 7th February, 1907, with 3 c.c. fresh defibrinated blood of foal 1997. These animals had previously been utilised for horse-sickness experiments.
7. *Mule* 2508.—Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction.
  8. *Mule* 2509.—Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction.
  9. *Mule* 2511.—Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction.
  10. *Mule* 2512.—Argentine gelding.  
*Injected* as above.  
*Result.*—Slight reaction from the 9th day.
  11. *Mule* 2534.—Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction.
  12. *Mule* 2535.—Argentine gelding.  
*Injected* as above.  
*Result.*—No reaction.
  13. *Mule* 2536.—Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction.
  14. *Mule* 2538.—Argentine mare.  
*Injected* as above.  
*Result.*—Slight reaction from the 10th day.
  15. *Mule* 2540.—Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction.
  16. *Mule* 2541.—Argentine mare.  
*Injected* as above.  
*Result.*—Slight reaction from the 15th day.

17. *Mule 2542*.—Argentine mare.  
*Injected* as above.  
*Result*.—No reaction.
18. *Mule 2543*.—Argentine mare.  
*Injected* as above.  
*Result*.—No distinct reaction.
19. *Mule 2544*.—Argentine mare.  
*Injected* as above.  
*Result*.—No reaction.
20. *Mule 2545*.—Argentine mare.  
*Injected* as above.  
*Result*.—Slight reaction.
21. *Mule 2546*.—Argentine mare.  
*Injected* as above.  
*Result*.—Slight reaction.

*Argentine donkeys injected with blood of foal 1997—immune against piroplasma equi.*

*Note*.—The following donkeys, Nos. 2264, 2265, 2254, 2255, were all injected subcutaneously on the 26th November, 1906, with 5 c.c. defibrinated fresh blood of foal 1997.

22. *Donkey 2264*.—Three-year-old Argentine mare.  
*Injected* as above.  
*Result*.—A slight reaction. All microscopical examinations negative.
23. *Donkey 2265*.—Two-year-old Argentine mare.  
*Injected* as above.  
*Result*.—Slight reaction. All microscopical examinations negative.
24. *Donkey 2254*.—Three-year-old Argentine mare.  
*Injected* as above.  
*Result*.—Slight reaction.
25. *Donkey 2255*.—Three-year-old Argentine mare.  
*Injected* as above.  
*Result*.—Slight reaction from the 15th day. All microscopical examinations negative.

*Argentine mules injected with blood of Transvaal foal 1998—immune against piroplasma equi.*

The following Argentine mules, Nos. 2446, 2447, 2449, 2450, 2451, 2453, 2454, 2457, 2547, 2579, 2580, 2583, 2584, 2585, 2586, 2587, had all been utilised in horse-sickness experiments before being injected on the 7th February, 1907, subcutaneously with 3 c.c. defibrinated fresh blood of foal 1998.

26. *Mule 2446*.—Argentine gelding.  
*Injected* as above.  
*Result*.—Reaction from the 16th day. All microscopical examinations negative.
- 27.—*Mule 2447*.—Argentine mare.  
*Injected* as above.  
*Result*.—Reaction from the 16th day. All microscopical examinations negative.

28. *Mule 2449.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 17th day.
29. *Mule 2450.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 11th to 23rd days.
30. *Mule 2451.*—Argentine mare.  
*Injected* as above.  
*Result.*—Slight reaction from the 20th day.
31. *Mule 2453.*—Argentine gelding.  
*Injected* as above.  
*Result.*—Reaction from the 19th day. All microscopical examinations negative.
32. *Mule 2454.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 16th day. All microscopical examinations negative.
33. *Mule 2457.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 13th day.
34. *Mule 2547.*—Argentine mare.  
*Injected* as above.  
*Result.*—Slight reaction from the 15th day.
35. *Mule 2579.*—Two-and-a-half-year-old Argentine gelding.  
*Injected* as above.  
*Result.*—Temperature remained high from the date of injection, but was probably due to the previous injection of horse-sickness blood. Slight reaction from the 6th day, recording 104.6 twenty-four hours later.
36. *Mule 2580.*—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Result.*—High temperature from date of injection, probably due to the previous inoculation of horse-sickness blood.
37. *Mule 2583.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 4th day.
38. *Mule 2584.*—Four-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 7th day.
39. *Mule 2585.*—Two-and-a-half-year-old Argentine gelding.  
*Injected* as above.  
*Result.*—Slight reaction from the 6th day.
40. *Mule 2586.*—Four-year-old Argentine mule.  
*Injected* as above.  
*Result.*—No distinct reaction.
41. *Mule 2587.*—Argentine gelding about 18 months old.  
*Injected* as above.  
*Result.*—Slight reaction.



*Transvaal donkey foal injected with blood of horse foal 1999—immune against piroplasma equi.*

42. *Donkey Foal 1773.*—Nine-month-old stallion, and born on the station.

*Injected* on the 14th September, 1906, subcutaneously with 5 c.c. defibrinated fresh blood of foal 1999.

*Result.*—Irregular reaction. All microscopical examinations negative.

*Argentine horses injected with blood of Transvaal horse foal 1999—immune against piroplasma equi.*

*Note.*—Horses 2098 and 2102 were previously inoculated on the 9th July, 1906, with blood of foal 1991 and 1993 respectively (compare Experiment 1, Nos. 2 and 6), and were now tested on their immunity.

43. *Horse 2098.*—Three-year-old Argentine mare.

*Injected* subcutaneously on the 5th November, 1906, with 5 c.c. defibrinated fresh blood of foal 1999.

*Result.*—Slight reaction from the 14th day. One point noted on the 10th day, and again present on the 12th and 13th days. *Piroplasma equi* present for the first time on the 16th day, but in rare numbers, and again noted three days later, accompanied with the rosette form. All further examinations negative. Red corpuscles did not drop below 6,000,000 per c.m.m.

44. *Horse 2102.*—

*Injected* subcutaneously on the 5th November with 5 c.c. defibrinated fresh blood of foal 1999.

*Result.*—No distinct reaction, the temperature consistently remaining between 97 and 101.8. All blood examinations negative.

*Transvaal mule injected with blood of Transvaal horse foal 1999—immune against piroplasma equi.*

45. *Mule 2212.*—Three-year-old gelding.

*Injected* on the 5th November, 1906, subcutaneously with 5 c.c. blood of foal 1999.

*Result.*—Slight reaction. One point noticed on the 12th day, followed by the lesions of a slight poikilocytosis. *Piroplasms* only present on the 16th and 31st days, but in very rare numbers.

*Argentine mule injected with blood of Transvaal horse foal 1999—immune against piroplasma equi.*

The following mules, 2326, 2327, 2328, 2329, 2330, were all injected subcutaneously on the 5th November, 1906, with 5 c.c. blood of foal 1999.

46. *Mule 2326.*—Two-year-old Argentine gelding.

*Injected* as above.

*Result.*—On the 11th day the temperature rose from 100 in the morning to 104.8 in the evening, but fell almost immediately, and remained normal from the 18th day. Points noted on the 15th day, and the lesions of poikilocytosis appeared on the 27th day. One point also noted on the 31st day, but *piroplasma equi* were not present.

47. *Mule 2327.*—Three-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Distinct reaction from the 9th day. Piroplasms noted on the 11th day, and together with points and the lesions of poikilocytosis, were fairly frequent during the next six days.
48. *Mule 2328.*—Two-year-old Argentine mare.  
*Injected* as above.  
*Result.*—Distinct reaction. Points noted on the 15th day, and the following day the lesions of a slight poikilocytosis present. One piroplasm, together with poikilocytosis, appeared on the 20th day, and this latter, together with points, were occasionally noted from the 26th to 31st days.
49. *Mule 2329.*—Eighteen-month-old Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction. Piroplasms only noted on the 27th day, but points and the lesions of poikilocytosis were of fairly frequent occurrence.
50. *Mule 2330.*—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Result.*—No distinct reaction. Piroplasms noted on the 25th and 27th days. The lesions of poikilocytosis also present.
- The following Argentine mules, Nos. 2458, 2459, 2460, 2461, 2462, 2464, 2465, 2467, 2468, 2469, 2470, 2471, 2472, 2473, were all injected subcutaneously on the 7th February, 1907, with 3 c.c. blood of foal 1999, and had previously been utilised for horse-sickness experiments.
51. *Mule 2458.*—Argentine gelding.  
*Injected* as above.  
*Result.*—Sharp reaction immediately after injection, recording 105 in the evenings of the 2nd and 4th days. Remained normal from the 5th day.
52. *Mule 2459.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 13th day, reaching 105 in the evening of the 15th day. Second reaction from the 24th day.
53. *Mule 2460.*—Argentine gelding.  
*Injected* as above.  
*Result.*—Reaction from the 9th to 22nd days, recording as a maximum 103.4 in the 17th day.
54. *Mule 2461.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 18th day, reaching 105 in the same evening, and again the following 24 hours later. All microscopical examinations negative.
55. *Mule 2462.*—Argentine mare.  
*Injected* as above.  
*Result.*—Distinct reaction from the 15th day.
- 56.—*Mule 2464.*—Argentine gelding.  
*Injected* as above.  
*Result.*—No distinct reaction until the 17th day, when a rise to 102.3 was noted two days later.

57. *Mule 2465*.—Argentine mare.

*Injected* as above.

*Result*.—Reaction commenced on the 8th day, but was terminated two days later by the death of the animal from the sequel of piroplasmosis.

*Post-mortem*.—

*Condition*:—Fair; rigor mortis not set in; yellow infiltration of subcutaneous issue on shoulder.

*Lungs*:—Slightly œdematous.

*Heart*:—Brownish liquid in heartbag; white coagula of plasma; no lesions on endocard; gelatinous infiltration of sulci-transversalis.

*Spleen*:—Slightly enlarged and slightly congested.

*Liver*:—Thicker than normal, and congested.

*Kidney*.—Capsula of left kidney diffusely infiltrated with blood; left capsula not easily removed, and broke whilst removing; blood infiltrations in capsula only; kidney pale; right kidney capsula not easily slipped off; a white spot, the size of a walnut, and resembling an infarct, at beginning of cortex; malpighis bodies distinctly enlarged.

*Stomach*:—Contained a few superficial hæmorrhages.

*Intestines*.—Pale; cæcum and colon pale.

58. *Mule 2467*.—Argentine mare.

*Injected* as above.

*Result*.—Slight reaction from the date of injection, the animal dying on the 14th day from piroplasmosis.

*Post-mortem*.—

*Condition*:—Fair; rigor mortis not completely set in; watery blood of a brownish colour ran from shoulder after cutting; fascies and flesh somewhat of a brownish colour; serous membranes and all organs pale.

*Lungs*:—Clear yellow liquid in peritoneal cavity; lungs normal in appearance, but pale.

*Heart*:—Abnormal amount of liquid in heartbag; myocard soft and of a sepia colour; left endocard distended and hæmorrhagic; right endocard distended with a few hæmorrhages.

*Spleen*:—Considerably enlarged; pulpa soft; urinary bladder distended and contained red urine.

*Liver*:—Of a lightish brown colour.

*Kidneys*:—Capsula firmly fixed to subcutaneous tissue.

*Stomach*:—Filled with food; mucosa normal.

*Intestines*:—Cæcum and colon normal.

59. *Mule 2468*.—Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction from the 17th day, recording 103 three days later, and remaining normal from the 24th day.

60. *Mule 2469*.—Argentine gelding.

*Injected* as above.

*Result*.—Slight reaction from the 9th day.

61. *Mule 2470.*—Argentine gelding.  
*Injected* as above.  
*Result.*—Reaction from the 5th to the 12th days.
62. *Mule 2471.*—Argentine gelding.  
*Injected* as above.  
*Result.*—Reaction from the 15th day, recording 104 on the 17th and 18th days.
63. *Mule 2472.*—Argentine gelding.  
*Injected* as above.  
*Result.*—Slight reaction from the 17th day.
64. *Mule 2473.*—Argentine mare.  
*Injected* as above.  
*Result.*—Reaction from the 5th day.
- Transvaal donkey foal injected with blood of a Transvaal donkey foal—  
 immune against piroplasma equi.*
65. *Donkey Foal 2550.*—Three months old, and born on the station.  
*Injected* on the 1st March, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2564.  
*Result.*—Reaction from the 7th day. *Piroplasma equi* noted on the 10th and 11th days, and two days later the lesions of poikilocytosis appeared.
- Argentine horse injected with blood of Transvaal donkey foal—  
 immune against piroplasma equi.*
66. *Horse 2681.*—Eight-year-old Argentine.  
*Injected* on the 8th May, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2564.  
*Result.*—Slight reaction from the 11th to 19th days.
- Results of animals inoculated with blood of Transvaal horse foals—  
 immune against piroplasma equi (third generation).*
- 1 Transvaal horse foal showed a reaction and recovered.  
 Of 55 Argentine mules the majority showed reactions, and two died, one from piroplasmosis and the other from the sequel of piroplasmosis.
- 4 Argentine donkeys showed reactions and recovered.  
 1 Transvaal donkey foal showed reactions and recovered.  
 1 Transvaal mule showed reactions and recovered.  
 Of two Argentine horses tested on their immunity, one showed a slight reaction; the other gave negative results.
- Results of animals inoculated with blood of a Transvaal donkey foal—  
 immune against piroplasma equi (third generation).*
- 1 Transvaal donkey foal and 1 Argentine horse passed through a reaction and recovered.

#### EXPERIMENT No. 13.

##### *Fifth Generation.*

*Argentine horse injected with blood of Transvaal donkey foal—  
 immune against piroplasma equi.*

1. *Horse 2683.*—Six-year-old Argentine mare.  
*Injected* on the 8th May, 1907, subcutaneously with 5 c.c. of donkey foal 2550.  
*Result.*—Slight reaction from the 7th day.

*Transvaal donkey foal injected with blood of a Transvaal donkey foal—  
immune against piroplasma equi.*

2. *Donkey Foal 2551.*—Three months old, and born on the station.  
*Injected* on the 26th March, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of Transvaal donkey foal 2550.  
*Temperature.*—Very slight reaction.  
*Examinations.*—The lesions of poikilocytosis noted on the 21st day.
3. *Donkey Foal 2494.*—Four-month-old foal, and born on the station.  
*Injected* on the 25th April, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2550.  
*Temperature.*—Reaction from the 13th day.  
*Examinations.*—*Piroplasma equi* noted on the 13th day, followed by the lesions of poikilocytosis on the 14th day.

*Argentine mules injected with blood of a Transvaal donkey foal—  
immune against piroplasma equi.*

4. *Mule 2539.*—Argentine gelding. (Note.—Mules 2539 and 2597 had previously been utilised for horse-sickness experiments.)  
*Injected* on the 26th March subcutaneously with 5 c.c. defibrinated fresh blood of foal 2550.  
*Temperature.*—Short reaction from the 9th to 13th days, recording as a maximum 103.8 on the 11th day after injection.  
*Examinations.*—*Piroplasma equi* only noted on the day of the maximum temperature.
5. *Mule 2597.*—Two-and-a-half-year-old Argentine mare.  
*Injected* on the 26th March subcutaneously with 5 c.c. defibrinated fresh blood of foal 2550.  
*Temperature.*—Slight reaction. *Piroplasma equi* noted on the 15th day, and the lesions of poikilocytosis appeared the next day.

*Argentine donkey injected with blood of a Transvaal donkey foal—  
immune against piroplasma equi.*

*Note.*—The following donkeys, Nos. 2432, 2433, 2435, 2437, 2439, 2441, 2444, 2445, were all injected subcutaneously on the 26th March, 1907, with 5 c.c. defibrinated fresh blood of foal 2550.

6. *Donkey 2432.*—Five-year-old Argentine mare.  
*Injected* as above.  
*Temperature.*—Reaction from the 7th day, recording 104 two days later.  
*Examinations.*—All negative.
7. *Donkey 2433.*—Four-year-old Argentine gelding.  
*Injected* as above.  
*Temperature.*—Slight reaction.  
*Examinations.*—All negative.
8. *Donkey 2435.*—Four-and-a-half-year-old Argentine gelding.  
*Injected* as above.  
*Temperature.*—Sharp rise noted from the 13th day, recording 104.2 on the evening of the 14th day, and regaining normal two days later.

9. *Donkey* 2437.—Two-and-a-half-year-old Argentine mare.  
*Injected* as above.  
*Temperature*.—Reaction from the 6th day, reaching 105 three days later, and 104 in the evening of the 13th day.  
*Examinations*.—Piroplasms noted on the 9th, 10th and 14th days.
10. *Donkey* 2439.—Two-and-a-half-year-old Argentine gelding.  
*Injected* as above.  
*Temperature*.—No distinct reaction.  
*Examinations*.—All negative.
11. *Donkey* 2441.—Three-year-old Argentine mare.  
*Injected* as above.  
*Temperature*.—Short reaction from the 14th day, lasting for three days.  
*Examinations*.—All negative.
12. *Donkey* 2444.—Three-and-a-half-year-old Argentine gelding.  
*Injected* as above.  
*Temperature*.—No distinct reaction.  
*Examinations*.—All negative.
13. *Donkey* 2445.—Six-year-old Argentine gelding.  
*Injected* as above.  
*Temperature*.—No reaction.  
*Examinations*.—All negative.

*Transvaal horse foal injected with blood of a Transvaal horse foal—immune against piroplasma equi.*

14. *Foal* 2620.—Filly obtained from S.A.C.  
*Injected* on the 30th May, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of foal 2681.  
*Temperature*.—Very slight reaction from the 8th day.  
*Examinations*.—The lesions of poikilocytosis noted on the 12th day.

*Results of animals injected with blood of a Transvaal donkey foal—immune against piroplasma equi (fourth generation).*

- 1 Argentine horse showed a reaction and recovered.  
 2 Transvaal donkey foals showed a reaction and recovered.  
 2 Argentine mules showed a reaction and recovered.  
 Of 8 Argentine donkeys, the majority showed a reaction, and all recovered.  
 1 Transvaal horse foal, injected with blood of an immune Transvaal horse foal (fourth generation), passed through a piroplasmosis reaction and recovered.

#### EXPERIMENT NO. 14.

##### *Sixth Generation.*

*Transvaal horse foal injected with blood of a Transvaal horse foal—immune against piroplasma equi.*

1. *Horse Foal* 2408.—About six months old.  
*Injected* on the 30th May, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of horse foal 2683.  
*Result*.—Slight reaction from the 5th to 21st day. Piroplasma noted on the 6th, 7th and 11th days. The lesions of poikilocytosis present on the 8th, 9th and 13th days.

*Transvaal horse foal injected with blood of Transvaal donkey foal—  
immune against piroplasma equi.*

2. *Foal 2707.*—Three-month-old colt, and obtained from S.A.C.

*Injected* on the 30th May subcutaneously with 5 c.c. defibrinated fresh blood of foal 2551.

*Temperature.*—Sharp rise from the 3rd day to 105 in the evenings of the 5th and 6th days, followed by a fall to 100 two days later, and a rise to 104.6 on the 10th day.

*Examinations.*—The lesions of poikilocytosis noted on the 5th, 7th, 9th and 11th days; piroplasma equi in rare numbers noted on the 8th day.

*Transvaal horse foal injected with blood of Transvaal donkey foal—  
immune against piroplasma equi.*

3. *Foal 2767.*—Six-month-old colt, and obtained from the S.A.C.

*Injected* on the 30th May, 1907, subcutaneously with 5 c.c. defibrinated fresh blood of donkey foal 2494.

*Temperature.*—Sharp reaction from the 8th day, reaching 104 in the evening two days later. The 21st day marked the commencement of a second reaction, but the animal died four days later of horse-sickness.

*Examinations.*—The lesions of poikilocytosis noted on the 9th, 11th and 14th days.

*Result* of two Transvaal horse foals injected with blood of Transvaal donkey foals (fifth generation), one died of horse-sickness.

One Transvaal horse foal injected with blood of a Transvaal horse foal (fifth generation), immune against piroplasma equi, passed through a reaction and recovered.

#### SUMMARY OF RESULTS.

##### (a) *Origin Horse Blood.*

###### *Injections with blood of horses and horse foals.*

Of 4 Transvaal horse foals, injected with immune blood (origin), none died.

Of 8 Argentine horse mares, injected with blood of 1st generation,  
1 died from gangrenous pneumonia (probably ship's pneumonia).

(This mare was heavy in foal.)

1 died from gastro-enteritis (probably horse-sickness.)

2 died from piroplasmosis. These mares were in foal, and this no doubt had some connection with the mortality.

##### (b) *Origin Mule Blood.*

###### *Injection with blood of horse foals.*

Of 9 Transvaal horse foals, injected with blood of 1st to 7th generations, none died.

Of 2 Argentine horses, injected with blood of 1st generation,

1 died of debility, complicated with piroplasmosis, and

1 died of syncope.

Of 2 Argentine horses, injected with blood of 2nd generation,

1 died of pneumonia (probably ship's pneumonia).

1 Argentine horse, injected with blood of 6th generation, died from debility, complicated with piroplasmosis.

- Of 12 Argentine donkeys, injected with blood of 1st and 2nd generations, 1 died of debility. This mare slipped her foal during the reaction.  
 Of 5 Transvaal mules, injected with blood of 2nd generation, none died.  
 Of 67 Argentine mules, injected with blood of 2nd, 3rd and 4th generations, 1 died of pneumonia (probably ship's pneumonia).

(c) *Origin Donkey Blood.*

1.—*Injections with donkey blood.*

- 1 Transvaal horse foal, injected with donkey blood (origin), recovered.  
 7 Argentine horses, 3 Argentine mules and 3 Argentine donkeys, tested on their immunity by injection of donkey foal blood (origin), recovered.

2.—*Injections with horse blood.*

- 2 Transvaal horse foals, injected with blood of a Transvaal horse (1st generation), recovered.

3.—*Injections with horse foal blood.*

- 1 Transvaal horse, injected with blood of Transvaal horse foal (1st generation), recovered.  
 7 Transvaal horse foals, injected with blood of Transvaal horse foal (2nd to 6th generations), recovered.  
 Of 6 Transvaal mules, injected with blood of Transvaal horse foals (2nd and 3rd generations), 1 died of piroplasmosis.  
 3 Transvaal donkey foals, injected with blood of Transvaal horse foals (1st, 2nd and 3rd generations), recovered.  
 Of 8 Argentine horses, injected with blood of a Transvaal horse foal (1st and 2nd generations), 2 died from rupture of the spleen, and 1 was killed on account of pleuro-pneumonia.  
 2 Argentine horses were tested on their immunity by injection of blood of Transvaal horse foal (3rd generation), and recovered.  
 Of 57 Argentine mules, injected with blood of Transvaal horse foal (1st, 2nd and 3rd generations), 1 died from piroplasmosis and 1 from sequel of piroplasmosis.  
 12 Argentine donkeys, injected with blood of Transvaal horse foal (2nd and 3rd generations), recovered.

4.—*Injections with donkey foal blood.*

- Of 2 Transvaal horse foals, injected with donkey foal blood (2nd and 5th generations), 1 died of horse-sickness, contracted spontaneously.  
 1 Transvaal mule, injected with blood of Transvaal donkey foal (2nd generation), recovered.  
 2 Transvaal donkey foals, injected with blood of a Transvaal donkey foal (2nd and 3rd generations), recovered.  
 6 Argentine horses, injected with blood of Transvaal donkey foals (2nd, 3rd and 4th generations), recovered.  
 2 Argentine mules, injected with blood of Transvaal donkey foals (4th generation), recovered.  
 Of 10 Argentine donkeys, injected with blood of Transvaal donkey foals (2nd and 4th generations), none died.

From this summary several points are noticeable, namely :—

- (a) 3 Argentine horses and 1 Argentine mule died from pneumonia, probably caused by an infection of "ship's pneumonia," one mare also being heavy in foal.



- (b) 2 Argentine mare horses, heavy in foal, died from piroplasmosis, and undoubtedly the pregnancy, together with the reaction, must be held responsible.
- (c) 2 Argentine horses died from piroplasmosis, complicated with debility.
- (d) 2 Argentine horses died from rupture of the spleen.

These points suggest certain precautionary measures which should be taken previous to inoculation:—(a) Animals imported from oversea should not be inoculated until all danger of an infection with ship's pneumonia has been removed; (b) mares heavy in foal should not be inoculated; (c) animals in poor condition should not be inoculated; (d) the contingency must always be expected that Argentine horses and mules may die of rupture of spleen, as they are very wild, and stabling often causes them to contract serious injuries.

*Deaths wholly or partially caused by the piroplasmosis reaction.*

(a) *Origin Horse Blood.*

3 Argentine horses (injected with horse foal blood of 1st generation).

(b) *Origin Mule Blood.*

1 Argentine horse (injected with horse foal blood of 1st generation).

1 Argentine horse (injected with horse foal blood of 6th generation).

(c) *Origin Donkey Blood.*

1 Transvaal mule (injected with horse foal blood of 2nd generation).

2 Argentine mules (injected with horse foal blood of 3rd generation).

In no instance did the injection of donkey foal blood cause the death of an animal.

*Note.*—All animals which survived the vaccination were exposed to natural infection soon after the conclusion of the reaction. These animals have been kept under close daily observation, and no deaths or relapses have been reported.

*Conclusions.*

(1) The inoculation of animals with horse foal blood of 1st, 2nd and 3rd generations caused a mortality of 7 out of 186, or 4 per cent.

(2) The inoculation of animals with horse foal blood of 4th, 5th, 6th, 7th and 8th generations caused a mortality of 1 in 16, or 6 per cent.

(3) The inoculation of animals with donkey foal blood of 2nd to 6th generation caused no mortality amongst 25 animals.

(4) The reactions caused by injection of horse foal blood were more severe than those given by injection of donkey foal blood.

(5) No cases of relapses after discharge have occurred, proving that the immunity given by the injection of donkey foal blood is as good as that afforded by horse foal blood.

(6) For further immunisation purposes, therefore, I recommend the passing of blood originating from a natural infection of a donkey with piroplasma equi through donkey foals, and to use 1 c.c. blood of that obtained from the 4th generation and upwards, bearing in mind the precautionary measures mentioned above.

(7) Finally, all foals kept for tapping purposes, and used in connection with the inoculation, must be kept free from ticks. This precaution has been carried out at this Laboratory with all foals used in the experiments, and it stands to reason that a reinfection by means of ticks would increase the virulency of the blood which is to be used as vaccine.