

D.—THE INOCULATION OF MULES WITH POLYVALENT VIRUS—
(continued).

The experience of the year 1905-06, when a monovalent virus of the Ordinary strain was utilised for the inoculation of mules in practice, resulted in breakdowns in immunity to the extent of 0·6 per cent. After it had been found that the Tzaneen virus broke the immunity obtained from the Ordinary virus, and it was naturally thought that the virus Tzaneen would protect the animals better in practice, this virus was introduced into practice, with the result that 1·5 per cent. of these animals died after discharge. At the same time it was found that the immunity obtained by either of the above vira could be broken reciprocally and also by a number of other vira which in the meantime we obtained in practice from mules suffering from relapses. Then the idea occurred to unite all these various vira and to obtain in this way a polyvalent virus which would protect against any of the vira of which it was composed, and by this means I hoped to reduce the mortality in practice. In my last annual report I quoted a number of experiments carried out with a trivalent and octovalent mixture of vira, these experiments being conducted with either adequate or inadequate serum. I also showed last year that by immunising with a polyvalent virus and serum a protection was obtained against any of the constituents, which prevented any deaths from the tests, although reactions occurred, and these experiments induced me to make the polyvalency of the virus even stronger by including all the vira which had been used up to that time.

The virus was introduced, and is referred to as CD (composite district); it consists of Ordinary, Tzaneen, Bulawayo, and mixtures of these three, plus a horse which had previously been injected with all three vira, together with relapse and spontaneous cases, the blood being collected in Zoutpansberg, Waterberg, Middelburg, Lydenburg, Rustenburg, Natal, Rhodesia, Lourenco Marques, Onderstepoort, Pretoria, Potchefstroom, and the high veld; all these various vira were injected into one horse (2884), and the resulting virus is called CD.

(Experiments Nos. 1 to 9, see Annual Report, 1906-07, pages 192-213.)

EXPERIMENT NO. 10.

With polyvalent serum and virus CD.

Serum: Polyvalent.

Inadequate to virus CD 2884.

Injection of 300 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 9th August, 1907.

“A.” Mule 2929.—Injected as above.

Result.—No reaction.

Tested on the 5th September, 1907, by intrajugular injection of 5 c.c. virus 2884.

Result.—No reaction.

Retested on the 19th September, 1907, by intrajugular injection of 5 c.c. virus 2769 (high veld).

Result.—Reaction from 4th to 11th days.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2808 (Rustenburg II).

Result.—Reaction from 7th to 19th days.

“ B.” *Mule* 2930.—Injected as above.

Result.—Reaction from 4th day, but not typical for horse-sickness.

Tested on the 5th September, 1907, by intrajugular injection of 5 c.c. virus 2884.

Result.—No reaction.

Retested on the 19th September, 1907, by intrajugular injection of 5 c.c. virus 2770 (Zoutpansberg and Waterberg).

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2785 (Natal, Rhodesia, and Lourenco Marques).

Result.—Reaction from 4th to 9th days.

“ C.” *Mule* 2931.—Injected as above.

Result.—Mild reaction from 6th to 14th days.

Tested on the 5th September, 1907, by intrajugular injection of 5 c.c. virus 2884.

Result.—No reaction.

Retested on the 19th September, 1907, by intrajugular injection of 5 c.c. virus 2773 (Pretoria and Potchefstroom).

Result.—Slight reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2780 (Middelburg and Lydenburg).

Result.—Reaction and dikkop on 12th day.

“ D.” *Mule* 2932.—Injected as above.

Result.—Slight reaction from 6th to 16th days.

Tested on the 5th September, 1907, by intrajugular injection of 5 c.c. virus 2884.

Result.—No reaction.

Retested on the 19th September, 1907, by intrajugular injection of 5 c.c. virus 2780 (Middelburg and Lydenburg).

Result.—Slight reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2773 (Pretoria and Potchefstroom).

Result.—No reaction.

“ E.” *Mule* 2933.—Injected as above.

Result.—Irregular reaction from 7th to 19th days.

Tested on the 5th September, 1907, by intrajugular injection of 5 c.c. virus 2884.

Result.—No reaction.

Retested on the 19th September, 1907, by intrajugular injection of 5 c.c. virus 2785 (Natal, Rhodesia, and Lourenco Marques).

Result.—Reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2770 (Zoutpansberg and Waterberg).

Result.—No reaction.

“ F.” *Mule* 2934.—Injected as above.

Result.—Doubtful reaction.

Tested on the 5th September, 1907, by intrajugular injection of 5 c.c. virus 2884.

Result.—No reaction.

Retested on the 19th September, 1907, by intrajugular injection of 5 c.c. virus 2808 (Rustenburg II).

Result.—Reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2769 (high veld).

Result.—Reaction.

Results.—Of six mules inoculated with polyvalent serum inadequate to virus CD, one gave no reaction, two gave doubtful reactions, and three gave slight reactions. When tested with constituents of virus CD reactions were again noted.

EXPERIMENT NO. 11.

With polyvalent serum of horses and mules and virus CD.

Serum: Mixture of 1906 of horses and mules hyperimmunised with various strains.

Inadequate to virus 2884, CD (third generation).

Injection of 300 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 15th August, 1907.

“ A.” *Mule 2992.*—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 5 c.c. virus 2952, CD (fourth generation).

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2769 (high veld).

Result.—Reaction from 4th to 9th days, but not quite typical for horse-sickness.

“ B.” *Mule 2993.*—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 5 c.c. virus 2952, CD (fourth generation).

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2770 (Zoutpansberg and Waterberg).

Result.—No reaction.

“ C.” *Mule 2994.*—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 5 c.c. virus 2952, CD.

Result.—No reaction.

Retested on the 4th October by intrajugular injection of 5 c.c. virus 2773 (Pretoria and Potchefstroom).

Result.—No reaction.

“ D.” *Mule 2995.*—Injected as above.

Result.—Reaction.

Tested on the 12th September, 1907, by intrajugular injection of 5 c.c. virus 2952, CD

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2780 (Middelburg and Lydenburg).

Result.—Reaction from 4th to 9th days.

“ E.” *Mule 2996.*—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 5 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2785 (Natal, Rhodesia, and Lourenco Marques).

Result.—Reaction from 4th to 10th days.

“F.” *Mule* 2997.—Injected as above.

Result.—Reaction.

Tested on the 12th September, 1907, by intrajugular injection of 5 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2808 (Rustenburg II).

Result.—Reaction from 3rd to 8th days.

Results.—Of six mules injected with polyvalent serum of horses and mules inadequate to virus 2884 CD, four gave slight reactions and two strong reactions. When tested later with constituents of virus 2884 some reactions were again noted.

EXPERIMENT No. 12.

With polyvalent serum of horses and virus CD.

Serum: Polyvalent mixture of horses.

Inadequate to virus 2884 CD.

Injection of 300 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 15th August, 1907.

“A.” *Mule* 2998.—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2804 (Rustenburg I).

Result.—Reaction from 7th to 15th days with slight dikkop on 15th day.

“B.” *Mule* 2999.—Injected as above.

Result.—Reaction.

Tested on the 12th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Reaction from 2nd to 14th days, but not typical for horse-sickness.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2809 (Potchefstroom I).

Result.—Reaction from 4th to 10th days.

“C.” *Mule* 3000.—Injected as above.

Result.—Reaction.

Tested on the 12th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2810 (Potchefstroom II).

Result.—Reaction from 3rd to 10th days.

“D.” *Mule* 3001.—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2790 (Onderstepoort).

Result.—Reaction from 4th to 10th days.

“ E.” *Mule* 3002.—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2870 (District).

Result.—No reaction.

“ F.” *Mule* 3003.—Injected as above.

Result.—Slight reaction.

Tested on the 12th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2599 (spontaneous cases).

Result.—No reaction.

Results.—Of six mules injected with a polyvalent serum mixture of horses inadequate to virus CD, four showed slight reactions and two strong reactions. When tested later with constituents of virus 2884, reactions—and in one case a dikkop—were noted.

EXPERIMENT No. 13.

With polyvalent serum in varying doses and virus CD.

Serum: Polyvalent.

Inadequate to virus CD 2884.

(a) Injection of 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 22nd August, 1907.

“ A.” *Mule* 3026.—Injected as above.

Result.—Slight reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952 CD.

Result.—Doubtful reaction, not typical for horse-sickness.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2709 Ordinary strain.

Result.—Irregular reaction.

“ B.” *Mule* 3025.—Injected as above.

Result.—Irregular reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2870 (District).

Result.—Irregular reaction.

“ C.” *Mule* 3018.—Injected as above.

Result.—Slight irregular reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Irregular reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2810 (Potchefstroom II).

Result.—Reaction from 3rd to 8th days.

(b) Dose of serum: 200 c.c.

“D.” *Mule* 3009.—Injected as above.

Result.—Reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—No reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2804 (Rustenburg I).

Result.—Reaction with a dikkop on the 15th day.

“E.” *Mule* 3023.—Injected as above.

Result.—Reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Doubtful reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2790 (Onderstepoort).

Result.—Reaction from 2nd to 16th days.

“F.” *Mule* 3012.—Injected as above.

Result.—Reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Irregular reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2809 (Potchefstroom I).

Result.—Reaction from 3rd to 14th days.

(c) Dose of serum: 150 c.c.

“G.” *Mule* 2894.—Injected as above. (Had been previously injected on 27th July, 1907, and gave a doubtful reaction.)

Result.—Slight irregular reaction.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Slight atypical reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2172 (Bulawayo, ninth generation).

Result.—Reaction.

“H.” *Mule* 2893.—Injected as above. (Had been previously injected on 27th July, 1907, and gave a slight reaction.)

Result.—Reaction from 2nd to 16th days.

Tested on the 19th September, 1907, by intrajugular injection of 10 c.c. virus 2952.

Result.—Slight reaction.

Retested on the 4th October, 1907, by intrajugular injection of 5 c.c. virus 2199 (Tzaneen, twelfth generation).

Result.—Reaction from 4th to 10th days.

Results.—Of three mules injected with 250 c.c. polyvalent serum inadequate to virus 2884 C.D., one gave a reaction and two slight reactions. When tested later with constituents of the virus mixture, reactions were again noted.

Of three mules injected with 200 c.c. polyvalent serum inadequate to virus 2884, all gave reactions; when tested later with constituents of the virus CD, reactions were again noticed in every case, and in one instance accompanied with a dikkop.

Of two mules injected with 150 c.c. polyvalent serum inadequate to virus 2884, one gave a doubtful reaction and the other a slight reaction; when tested later with constituents of virus 2884, reactions were again noted.

EXPERIMENT No. 14.

With serum and virus CD.

Serum: CD of two horses, dated 28th August, 1907.

Adequate to virus 2884 CD.

Injection of 300 c.c. serum and 2 c.c. virus simultaneously and subcutaneously.

N.B.—Serum CD was obtained from two horses, already hyperimmunised, one with virus OTBLPW, and the other with O, T, and O virus alternately, and finally both hyperimmunised with virus CD.

“A.” *Mule 3005.*—Injected as above.

Result.—Slight reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Irregular and doubtful reaction.

“B.” *Mule 3006.*—Injected as above.

Result.—Slight reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Irregular doubtful reaction.

“C.” *Mule 3007.*—Injected as above.

Result.—Reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Irregular, doubtful reaction.

Results.—Of three mules injected with serum CD, adequate to virus 2884 CD, two gave slight reactions, and one a strong reaction. When tested later with constituents of virus 2884, all three animals gave doubtful reactions.

EXPERIMENT No. 15.

With serum and virus CD.

Serum: CD of horses.

Adequate to virus CD 2884.

Injection of 300 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 28th August, 1907.

N.B.—Serum obtained from horses hyperimmunised with CD virus exclusively.

“A.” *Mule 3008.*—Injected as above.

Result.—Reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).*

Killed on 30th October, 1907, owing to debility.

* Virus “Total CD” is a virus obtained by mixing the constituents of CD.

“ B.” *Mule* 3010.—Injected as above.

Result.—Slight irregular reaction of a doubtful character.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—Slight reaction.

“ C.” *Mule* 3011.—Injected as above.

Result.—Hardly any reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—Slight reaction from 6th to 12th day.

Results.—Of three mules injected with polyvalent serum adequate to virus 2884, one gave a reaction, and two doubtful reactions. When tested later with a mixture of the constituents of virus CD, two slight reactions were noted.

EXPERIMENT No. 16.

With polyvalent serum and virus CD.

Serum: Polyvalent.

Inadequate to virus 2884 CD.

(a) Injection of 150 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 5th September, 1907.

“ A.” *Mule* 3013.—Injected as above.

Result.—Irregular reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—Irregular reaction.

“ B.” *Mule* 3014.—Injected as above.

Result.—Reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“ C.” *Mule* 3015.—Injected as above.

Result.—Reaction and dikkop.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“ D.” *Mule* 3016.—Injected as above.

Result.—Reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—Doubtful reaction.

“ E.” *Mule* 3021.—Injected as above.

Result.—Reaction and dikkop.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—Doubtful reaction.

“ F.” *Mule* 3024.—Injected as above.

Result.—Reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

Results.—Of six mules injected with 150 c.c. polyvalent serum inadequate to virus 2884, reactions were noted in every case. When tested later no reactions were given against virus CD, but doubtful reactions were noted against a mixture of the constituents of virus CD.

EXPERIMENT No. 17.

With virus and serum CD.

Serum: CD.

Adequate to virus 2884 CD.

Injection of 200 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 19th October, 1907.

“A.” *Mule* 3017.—Injected as above.

Result.—Reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“B.” *Mule* 3019.—Injected as above.

Result.—Slight reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“C.” *Mule* 3020.—Injected as above.

Result.—Irregular reaction.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—Reaction.

“D.” *Mule* 3022.—Injected as above.

Result.—Slight reaction of a doubtful character.

Tested on the 24th October, 1907, by subcutaneous injection of 5 c.c. virus (Total CD).

Result.—No reaction.

Results.—Of four mules injected with 200 c.c. serum CD, adequate to virus CD, reactions and doubtful reactions were given. When tested later, none showed reactions against virus CD, but one out of two gave a reaction against a mixture of the constituents of virus CD.

EXPERIMENT No. 18.

With serum and virus CD.

Serum: CD of four horses, dated 2nd September, 1907.

Adequate to virus CD 2884.

(a) Injection of 150 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 10th October, 1907.

“A.” *Mule* 3113.—Injected as above.

Result.—Atypical reaction.

Tested on the 14th November, 1907, by subcutaneous injection of 5 c.c. virus 2936 (CD, fourth generation).

Result.—Atypical reaction.

“B.” *Mule* 3103.—Injected as above.

Result.—Irregular reaction.

Tested on the 14th November, 1907, by subcutaneous injection of 5 c.c. virus 2936.

Result.—No reaction.

“C.” *Mule* 3114.—Injected as above.

Result.—Reaction.

Tested on the 14th November, 1907, by subcutaneous injection of 5 c.c. virus 2936.

Result.—No reaction.

(b) Dose of serum 200 c.c.

“ D.” *Mule* 3112.—Injected as above.

Result.—Irregular reaction.

Tested on the 14th November, 1907, by subcutaneous injection of 5 c.c. virus 2936.

Result.—No reaction.

“ E.” *Mule* 3102.—Injected as above.

Result.—Reaction.

Tested on the 14th November, 1907, by subcutaneous injection of 5 c.c. virus 2936.

Result.—No reaction.

“ F.” *Mule* 3107.—Injected as above.

Result.—Reaction.

Tested on the 14th November, 1907, by subcutaneous injection of 5 c.c. virus 2936.

Result.—No reaction.

Results.—Of six mules injected with 150 c.c. and 200 c.c. serum CD, adequate to virus CD, three showed irregular reactions and three strong reactions; when tested later with the next generation of virus CD, five gave negative results and one showed an atypical reaction.

EXPERIMENT NO. 19.

With virus and serum CD.

Serum: No. 196 and 197 CD of forty-two horses.

Adequate to virus 2884 CD.

(a) Injection of 100 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 8th November, 1907.

“ A.” *Mule* 3138.—Injected as above.

Result.—Reaction.

Exposed from the 11th January, 1908, to 8th March, 1908, at Potgietersrust.

Result.—No reaction.

“ B.” *Mule* 3139.—Injected as above.

Result.—Contracted horse-sickness and died on the 11th day.

“ C.” *Mule* 3106.—Injected as above.

Result.—Severe reaction with symptoms of horse-sickness.

Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.

Result.—No reaction.

(b) Dose of serum 150 c.c.

“ D.” *Mule* 3137.—Injected as above.

Result.—Reaction.

Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.

Result.—No reaction.

“ E.” *Mule* 3140.—Injected as above.

Result.—Reaction.

Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.

Result.—No reaction.

- “ F.” *Mule 3111.*—Injected as above.
Result.—Reaction.
 Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.
Result.—No reaction.
- “ G.” *Mule 3110.*—Injected as above.
Result.—Reaction.
 Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.
Result.—No reaction.
- (c) Dose of serum 200 c.c. .
- “ H.” *Mule 3109.*—Injected as above.
Result.—Reaction.
 Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.
Result.—No reaction.
- “ I.” *Mule 3108.*—Injected as above.
Result.—Reaction.
 Exposed on the 11th January, 1908, in Potgietersrust.
Result.—Died of spontaneous horse-sickness on 2nd March, 1908.
- “ J.” *Mule 3099.*—Injected as above.
Result.—Severe reaction. Died on 1st January, 1908, with symptoms of anaemia.
- “ K.” *Mule 3105.*—Injected as above.
Result.—Contracted horse-sickness and died on the 10th day.
- “ L.” *Mule 3104.*—Injected as above.
Result.—Died of horse-sickness on the 12th day.
- “ M.” *Mule 3101.*—Injected as above.
Result.—Reaction.
 Exposed from the 11th January, 1908, to 8th March, 1908, in Potgietersrust.
Result.—No reaction.
- “ N.” *Mule 3100.*—Injected as above.
Result.—Reaction. Died on the 17th day from peritonitis and pneumonia necrotica.

Results.—Of three animals injected with 100 c.c. horse serum Nos. 196 and 197, adequate to virus CD, one died of horse-sickness on the 11th day, and two showed reactions. Two months later the surviving animals were exposed in Potgietersrust without any relapses occurring.

Of four mules injected with 150 c.c. horse serum Nos. 196 and 197, adequate to virus CD, all showed reaction. Two months later when exposed in Potgietersrust no relapses occurred.

Of seven mules injected with 200 c.c. horse serum Nos. 196 and 197, adequate to virus CD, all gave reactions, and two died of horse-sickness; when exposed two months later in Potgietersrust one of the mules contracted horse-sickness spontaneously and died.

EXPERIMENT NO. 20.

With polyvalent serum and virus CD 2884.

- (a) Serum: Polyvalent No. 198 (rest of sample).
 Inadequate to virus 2884 CD.

Injection of 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 7th December, 1907.

N.B.—Serum 198=Serum OTB of 1906, consisting of two-thirds horse serum and one-third mule serum of animals hyperimmunised with virus O+T+B.

“A.” *Mule* 3185.—Injected as above.

Result.—Slight reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2884.

Result.—Indefinite reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Indefinite reaction.

“B.” *Mule* 3179.—Injected as above.

Result.—Doubtful reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2415 (Tzaneen).

Result.—No reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Indefinite reaction.

(b) Serum 201 (OTB, as before).

Inadequate to virus 2884 CD.

“C.” *Mule* 3187.—Injected as above.

Result.—Slight reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2884.

Result.—Indefinite reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Indefinite reaction.

“D.” *Mule* 3183.—Injected as above.

Result—Reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2415.

Result.—Reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Reaction; probably due to test with virus 2415.

(c) Serum 202 (OTB, as before).

Inadequate to virus 2884 CD.

“E.” *Mule* 3182.—Injected as above.

Result.—Doubtful reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2884.

Result.—No reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

" F." Mule 3186.—Injected as above.

Result.—Doubtful reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2415.

Result.—Indefinite reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Indefinite reaction.

(d) Serum 203 (OTB, as before).

Inadequate to virus 2884, CD.

" G." Mule 3188.—Injected as above.

Result.—Slight reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2884.

Result.—Indefinite reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Indefinite reaction.

" H." Mule 3181.—Injected as above.

Result.—Reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2415.

Result.—Indefinite reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Indefinite reaction.

(e) Serum 204 (OTB, as before).

Inadequate to virus 2884 CD.

" I." Mule 3180.—Injected as above.

Result.—Doubtful reaction.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2884.

Result.—No reaction.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

" J." Mule 3184.—Injected as above.

Result.—Reaction and dikkop.

Tested on the 7th January, 1908, by subcutaneous injection of 2 c.c. virus 2415.

Result.—Reaction with dikkop on 14th day.

Retested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

Results.—Of ten mules injected with polyvalent serum, twelve months old, inadequate to virus 2884 CD, three gave strong reactions, accompanied in one case with dikkop; three gave slight reactions and four gave doubtful reactions. Five mules were tested with CD virus, and three gave doubtful reactions and two negative results. The other five mules were tested with virus 2415, with the result that one gave a reaction and dikkop, one a reaction, two doubtful reactions, and one negative.

All ten mules were retested with virus 2884, resulting in one reaction, six doubtful reactions, and three negative.

EXPERIMENT No. 21.

With serum and virus CD.

(a) Serum 188, dated 23rd September, 1907.

Adequate to virus 2884 CD.

Injection of 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 23rd December, 1907.

N.B.—Serum 188 was obtained from the fifteen horses which were the constituents of serum 197.

“A.” *Mule* 3223.—Injected as above.

Result.—Irregular reaction.

Tested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Reaction.

“B.” *Mule* 3224.—Injected as above, but virus 2884 given intrajugularly.

Result.—Reaction.

Tested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—Irregular reaction.

(b) Serum 192, dated 7th October, 1907.

Adequate to virus CD 2884.

Injection of 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously.

N.B.—Serum 192 was obtained from seventeen horses, the constituents of serum 197.

“C.” *Mule* 3221.—Injected as above.

Result.—Slight reaction.

Tested on the 5th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“D.” *Mule* 3222.—Injected as above, but virus given intrajugularly.

Result.—Reaction and dikkop on 10th day. Died of horse-sickness on 14th day.

(c) Serum 197.

Adequate to virus CD 2884.

Injection of 200 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 23rd December, 1907.

“E.” *Mule* 3217.—Injected as above.

Result.—Very severe reaction with dikkop.

Tested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

F.” *Mule* 3218.—Injected as above, but dose of serum 250 c.c.

Result.—Reaction.

Tested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“G.” *Mule* 3219.—Injected as above. (Dose of serum, 200 c.c.)

Result.—Severe reaction.

Tested on the 15th January, 1909, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“H.” Mule 3220.—Injected as above, but virus intrajugularly.

Result.—Doubtful reaction.

Tested on the 15th January, 1908, by injection of 5 c.c. virus 2884.

Result.—Doubtful reaction.

(d) Serum 215 A, dated 21st December, 1907.

Adequate to virus 2884 CD.

Injection of 200 and 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 23rd December, 1907.

N.B.—Serum 215 A obtained from twenty-two horses, constituents of serum 197.

“I.” Mule 3213.—Injected as above.

Result.—Severe reaction.

Tested on the 15th January, 1907, by injection of 5 c.c. virus 2884.

Result.—No reaction.

“J.” Mule 3214.—Injected as above, but dose of serum 250 c.c.

Result.—Severe reaction with dikkop.

Not tested.

“K.” Mule 3215.—Injected as above.

Result.—Died of horse-sickness on 11th day.

“L.” Mule 3216.—Injected as above, but virus intrajugularly.

Result.—Severe reaction with piroplasmosis.

Tested on the 15th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

Results.—Of twelve mules injected with serum CD, adequate to virus CD, two gave reactions and dikkop, five strong reactions, two slight reactions, one doubtful, and two died of horse-sickness. When tested later with the same virus—one strong, one slight, and one doubtful—reactions were noted, and six gave negative results; the remaining mule was not tested.

EXPERIMENT No. 22.

With serum and virus CD.

(A) Serum 218.

Adequate to virus 2884.

Injection of 300 and 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 10th January, 1908.

N.B.—Serum 218, obtained from eleven horses, constituents of serum 197.

(a) Dose, 300 c.c. serum.

“A.” Mule 3296.—Injected as above.

Result.—Reaction from 8th to 16th days.

(b) Dose of serum, 250 c.c.

“B.” Mule 3291.—Injected as above.

Result.—Reaction from 10th day, and death of horse-sickness on 14th day.

(B) Serum 219.

Adequate to virus 2884 CD.

Injection of 300 and 250 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 10th January, 1908.

N.B.—Serum 219, obtained from twelve horses, constituents of serum 197.

(a) Dose of serum, 300 c.c.

“C.” *Mule 3297*.—Injected as above.

Result.—Hardly any reaction.

Tested on the 24th January, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

(b) Dose of serum, 250 c.c.

“D.” *Mule 3292*.—Injected as above.

Result.—Reaction from 7th to 17th days.

Results.—Of four mules injected with serum CD, adequate to virus 2884 CD, one died of horse-sickness, three gave strong reactions, and one a very slight reaction. The latter animal was tested with 5 c.c. same virus, but did not give a reaction.

EXPERIMENT No. 22.

With serum OTB and virus CD.

Serum 201 OTB

Inadequate to virus 2884 CD.

Injection of 300 and 350 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 7th February, 1908, followed by a second injection of 200 or 100 c.c. serum seven days later.

(a) Dose of serum 300 c.c. and second injection of 100 c.c.

“A.” *Mule 3346*.—Injected as above.

Result.—Indefinite reaction.

Tested on the 24th February, 1908, by subcutaneous injection of 2 c.c. virus 3330 (Ordinary, seventy-first generation).

Result.—No reaction.

“B.” *Mule 3347*.—Injected as above.

Result.—Reaction.

Tested on the 24th February, 1908, by subcutaneous injection of 2 c.c. virus 3330.

Result.—No reaction.

“C.” *Mule 3349*.—Injected as above.

Result.—Incubation time of five days; reaction for seven days; dikkop on 11th day, and death on following night.

“D.” *Mule 3366*.—Injected as above.

Result.—Reaction.

Tested on the 21st February, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

“E.” *Mule 3367*.—Injected as above.

Result.—Reaction.

Tested on the 21st February, 1908, by subcutaneous injection of 5 c.c. virus 2884.

Result.—No reaction.

(b) Dose of serum 350 c.c. followed by second injection of 200 c.c.

“F.” *Mule 3348*.—Injected as above.

Result.—Reaction.

Tested on the 24th February, 1908, by subcutaneous injection of 2 c.c. virus 3330.

Result.—No reaction.

(c) Dose of serum 300 c.c., followed by second injection of 200 c.c.

“G.” *Mule* 3350.—Injected as above.

Result.—Reaction.

Tested on the 24th February, 1908, by subcutaneous injection of 2 c.c. virus 3330.

Result.—No reaction.

Results.—Of seven mules injected with OTB serum, inadequate to virus 2884, and which received a second injection of the same serum on the 7th day, one died of horse-sickness, five showed strong reactions, and one gave a doubtful reaction. When tested later with either a constituent or the virus CD 2884, no reactions were noted.

EXPERIMENT No. 23.

With serum mixture 230 and virus CD.

Serum 230.

Inadequate to virus 2884 CD.

Injection of 300 c.c. serum and 2 c.c. virus simultaneously and subcutaneously on the 10th March, 1908.

N.B.—Serum mixture 230 consisted of serum known as Dikkop, Tzaneen, Spontaneous cases, and of mules Tzaneen and Bulawayo, together with serum CD.

“A.” *Mule* 3418.—Injected as above.

Result.—Incubation three days; died from horse-sickness on the 11th day.

“B.” *Mule* 3419.—Injected as above.

Result.—Incubation time three days; dikkop on 12th day, and death from horse-sickness during night of 13th–14th day.

“C.” *Mule* 3420.—Injected as above.

Result.—Slight reaction.

Tested on the 15th April, 1908, by subcutaneous injection of 2 c.c. virus 3275 (Simpson).

Result.—No reaction.

Retested on the 7th May, 1908, by subcutaneous injection of 2 c.c. virus 3494 (Potgietersrust).

Result.—No reaction.

“D.” *Mule* 3421.—Injected as above.

Result.—Doubtful reaction.

Tested on the 15th April, 1908, by subcutaneous injection of 2 c.c. virus 3287 (Simpson).

Result.—No reaction.

Retested on the 7th May, 1908, by subcutaneous injection of 2 c.c. virus 3494 (Potgietersrust).

Result.—No reaction.

“E.” *Mule* 3422.—Injected as above.

Result.—Reaction.

Tested on the 15th April, 1908, by subcutaneous injection of 2 c.c. virus 3308 (Simpson).

Result.—No reaction.

Retested on the 7th May, 1908, by subcutaneous injection of 2 c.c. virus 3494 (Potgietersrust).

Result.—No reaction.

“ F.” *Mule 3423*.—Injected as above.

Result.—Reaction.

Tested on the 15th April, 1908, by subcutaneous injection of 2 c.c. virus 3415 (Simpson).

Result.—No reaction.

Retested on the 7th May, 1908, by subcutaneous injection of 2 c.c. virus 3494 (Potgietersrust).

Result.—No reaction.

Conclusions.—Of six mules injected with polyvalent serum inadequate to virus 2884, two died of horse-sickness, three gave reactions, and one a doubtful reaction.

When tested later with a constituent of virus 2884, no reactions occurred.

TABULATED RÉSUMÉ.

MULE No.	IMMUNISATION.					FIRST TEST.					
	Date.	Poly- valent. Serum.	Virus.			RE- SULT.	Date.	Virus.			RE- SULT.
			No.	Orig.	Gen.			No.	Orig.	Gen.	
	1907.						1907.				
2929	Aug. 9	Inadeq.	2884	CD	3	—	Sept. 5	2884	CD	3	—
2930	"	"	"	"	"	?	"	"	"	"	—
2931	"	"	"	"	"	r	"	"	"	"	—
2932	"	"	"	"	"	r	"	"	"	"	—
2933	"	"	"	"	"	r	"	"	"	"	—
2934	"	"	"	"	"	?	"	"	"	"	—
2992	Aug. 15	"	"	"	"	r	Sept. 12	2952	"	4	—
2993	"	"	"	"	"	r	"	"	"	"	—
2994	"	"	"	"	"	r	"	"	"	"	—
2995	"	"	"	"	"	R	"	"	"	"	—
2996	"	"	"	"	"	r	"	"	"	"	—
2997	"	"	"	"	"	R	"	"	"	"	—
2998	"	"	"	"	"	r	"	"	"	"	—
2999	"	"	"	"	"	R	"	"	"	"	?
3000	"	"	"	"	"	R	"	"	"	"	—
3001	"	"	"	"	"	r	"	"	"	"	—
3002	"	"	"	"	"	r	"	"	"	"	—
3003	"	"	"	"	"	r	"	"	"	"	—

?—Doubtful reaction.
r—Slight reaction.
R—Typical reaction.
—No reaction.

TABULATED RÉSUMÉ—(continued).

SECOND TEST.					THIRD TEST.				
Date.	Virus.			RE-SULT.	Date.	Virus.			RE-SULT.
	No.	Orig.	Gen.			No.	Orig.	Gen.	
1907.					1907.				
Sept. 19	2769	H. Veld	—	R	Oct. 4	2808	R. II	—	R
"	2770	Z. and W.	—	—	"	2785	N., Rh., L.M.	—	R
"	2773	P. and P.	—	r	"	2780	M. and L.	—	R & D
"	2780	M. and L.	—	r	"	2773	P. and P.	—	—
"	2785	N., Rh., L.M.	—	R	"	2770	Z. and W.	—	—
"	2808	R. II	—	R	"	2769	H. Veld	—	R
Oct. 4	2769	H. Veld	—	?					
"	2770	Z. and W.	—	—					
"	2773	P. and P.	—	—					
"	2780	M. and L.	—	R					
"	2785	N., Rh., J.M.	—	R					
"	2808	R. II	—	R					
"	2804	R. I	—	R & D					
"	2809	P. I	—	R					
"	2810	P. II	—	R					
"	2790	O. Poort	—	R					
"	2870	District	—	—					
"	2599	Spont.	—	—					

R—Typical horse-sickness reaction.

r—Slight reaction.

R & D—Typical reaction accompanied with dikkop.

—No reaction.

TABULATED RESUME—(continued).

MULE No.	IMMUNISATION						FIRST TEST.					SECOND TEST.				
	Date.	Poly-valent Serum.	Virus.			RESULT.	Date.	Virus.			RESULT.	Date.	Virus.			RESULT.
			No.	Orig.	Gen.			No.	Orig.	Gen.			No.	Orig.	Gen.	
3026	1907. Aug. 22	Inadeq.	2884	CD	3	r	1907. Sept. 19	2952	CD	4	?	1907. Oct. 4	2709	O.	70	r
3025	"	"	"	"	"	r	"	"	"	"	R	"	2870	District	—	r
3018	"	"	"	"	"	r	"	"	"	"	—	"	2810	P. II	—	R
3009	"	"	"	"	"	R	"	"	"	"	—	"	2804	R. I	—	R & D
3023	"	"	"	"	"	R	"	"	"	"	?	"	2790	O. Poort	—	R
3012	"	"	"	"	"	R	"	"	"	"	r	"	2809	P. I	—	R
2894	"	"	"	"	"	?	"	"	"	"	?	"	2172	Bul.	9	R
2893	"	"	"	"	"	R	"	"	"	"	r	"	2199	Tz.	12	R
3005	Aug. 28	Adeq.	2884	CD	3	r	Oct. 24	2884	CD	3	?	"				
3006	"	"	"	"	"	r	"	"	"	"	?	"				
3007	"	"	"	"	"	R	"	"	"	"	?	"				
3008	"	"	"	"	"	R	"	—	Total CD	—	Killed on account of debility.	"				
3010	"	"	"	"	"	?	"	—	"	—	r	"				
3011	"	"	"	"	"	?	"	—	"	—	r	"				
3013	Sept. 5	Inadeq.	2884	CD	3	r	Oct. 24	—	"	—	r	"				
3014	"	"	"	"	"	R	"	2884	CD	3	—	"				
3015	"	"	"	"	"	R & D	"	"	"	"	—	"				
3016	"	"	"	"	"	R	"	—	Total CD	—	?	"				
3021	"	"	"	"	"	R & D	"	—	"	—	?	"				
3024	"	"	"	"	"	R	"	2884	CD	3	—	"				
3017	Sept. 19	Adeq.	2884	CD	3	R	Oct. 24	"	"	"	—	"				
3019	"	"	"	"	"	r	"	"	"	"	—	"				
3020	"	"	"	"	"	r	"	—	Total CD	—	R	"				
3022	"	"	"	"	"	?	"	—	"	—	—	"				
3113	Oct. 10	"	2884	CD	3	?	Nov. 14	2936	CD	4	?	"				
3103	"	"	"	"	"	r	"	"	"	"	—	"				
3114	"	"	"	"	"	R	"	"	"	"	—	"				
3112	"	"	"	"	"	r	"	"	"	"	—	"				

TABULATED RESUME—(continued).

MULE No.	IMMUNISATION.						FIRST TEST.				SECOND TEST.					
	Date.	Poly- valent Serum.	Virus.			RESULT.	Date.	Virus.			RESULT.	Date.	Virus.			RESULT.
			No.	Orig.	Gen.			No.	Orig.	Gen.			No.	Orig.	Gen.	
3224	1907. Dec. 23	Adeq. CD	2884	CD	3	R	1908. Jan. 15	2884	CD	3	—					
3221	"	"	"	"	"	r	"	"	"	"	—					
3222	"	"	"	"	"	Died of horse- sickness	"	"	"	"	—					
3217	"	"	"	"	"	R & D	Jan. 15	2884	CD	3	—					
3218	"	"	"	"	"	R	"	"	"	"	—					
3219	"	"	"	"	"	R	"	"	"	"	—					
3220	"	"	"	"	"	?	"	"	"	"	—					
3213	"	"	"	"	"	R	"	"	"	"	—					
3214	"	"	"	"	"	R & D	—	—	—	—	Not tested.					
3215	"	"	"	"	"	Died of horse- sickness										
3216	"	"	"	"	"	R	Jan. 15	2884	CD	3	—					
3296	Jan. 10	"	2884	CD	3	R	—	—	—	—	Not tested.					
3291	"	"	"	"	"	Died of horse- sickness										
3297	"	"	"	"	"	r	Jan. 24	2884	CD	3	—					
3292	"	"	"	"	"	R	—	—	—	—	Not tested.					
3346	Feb. 7	Inadeq. OTB	2884	CD	3	?	Feb. 24	3330	O	71	—					
3347	"	"	"	"	"	R	"	"	"	"	—					
3348	"	"	"	"	"	R	"	"	"	"	—					
3349	"	"	"	"	"	Died of horse- sickness										

3350	"	"	"	"	"	R	Feb. 24	3330	O	71	—					
3366	Feb. 8	"	"	"	"	R	Feb. 21	2884	CD	3	—					
3367	"	"	"	"	"	R	"	"	"	"	—					
3393	Feb. 21	"	"	"	"	R	March 9	"	"	"	—					
3394	"	"	"	"	"	R	"	"	"	"	—					
3395	Feb. 28	"	"	"	"	R	"	"	"	"	—					
3418	March 10	Inadeq. Polyv.	"	"	"	Died of horse- sickness do.										
3419	"	"	"	"	"	r	April 15	3275	Simpson	—	—	1908.				
3420	"	"	"	"	"	r	"	3287	"	2	—	May 7	3494	P.P.R.	—	—
3421	"	"	"	"	"	?	"	3308	"	2	—	"	"	"	1	—
3422	"	"	"	"	"	R	"	3308	"	2	—	"	"	"	1	—
3423	"	"	"	"	"	R	"	3415	"	2	—	"	"	"	1	—

r—Slight reaction.
 ?—Doubtful reaction.
 R—Typical reaction.
 R & D—Typical reaction accompanied with dikkop.
 ——No reaction.

ANALYSIS OF RESULTS FROM TABULATED RÉSUMÉ.

1. The immunisation of mules was introduced with two kinds of sera—(a) inadequate to the virus being composed of various monovalent sera, and also some trivalent and octovalent sera used in previous years for experimental purposes (see Annual Report, 1906–07), and (b) adequate to the virus 2884, that is of horses hyperimmunised with this virus.

2. Of 60 mules immunised with polyvalent serum inadequate to virus CD, 3 died of horse-sickness, or 5 per cent., the dose of serum being 300 c.c.

3. Of 46 mules immunised with polyvalent serum adequate to virus CD, 6 died, or 13 per cent.,

1	having	received	300	c.c.	serum.
2	„	„	250	c.c.	„
1	„	„	200	c.c.	„
2	„	„	100	c.c.	„

4. Based on these results the dose of serum was fixed at 300 c.c.

5. The immunity obtained by either an inadequate or an adequate serum was tested by subsequent inoculations of the CD virus, with the result that (a) of 44 mules immunised with serum inadequate to the virus CD, and tested with the same virus, 5 reacted—12 per cent.; (b) of 25 mules immunised with serum adequate to the virus CD, and tested with the same virus, 5 reacted—20 per cent.

6. The total result of 69 animals immunised with CD, and tested with the same virus, is therefore that 10 reacted—15 per cent.

7. Twenty-two animals were immunised with CD, and tested, as follows:—

	RESULT.			
	Reaction.	Reaction and Dikkop.	Reaction and Died.	No Reaction.
4 were tested with Ordinary virus ..	%	%	%	%
4 „ „ „ Simpson virus ..	—	—	—	100
5 „ „ „ Tzaneen, fourth generation ..	20	20	—	60
9 Were exposed in Potgietersrust ..	—	—	11	—

8. Twenty-eight animals, immunised with CD virus and tested for the first time with the same virus, were tested again with constituents of virus 2884, with the following result :—

	RESULT.		
	Reaction.	Reaction and Dikkop.	No Reaction.
2 With High Veld virus	1	—	1
2 „ Zoutpansberg and Waterberg	—	—	2
2 „ Potgietersrust	1	—	1
2 „ Middelburg and Lydenburg	2	—	—
2 „ Natal, Rhodesia, and Lourenco Marques	2	—	—
2 „ Rustenburg (2)	2	—	—
3 „ Rustenburg (1)	1	2	—
2 „ Pretoria (1)	2	—	—
2 „ Pretoria (2)	2	—	—
2 „ Onderstepoort	2	—	—
2 „ Total District virus (all above)	2	—	—
1 „ Virus of Spontaneous cases	—	—	1
1 „ Tzaneen, fourth generation	1	—	—
1 „ Tzaneen, twelfth generation.	1	—	—
1 „ Ordinary virus	1	—	—
1 „ Bulawayo virus	1	—	—
28	21	2	5

The percentage being : Reactions, 75 per cent.
Reactions and dikkop, 7 per cent.
No reactions, 18 per cent.

9. Eight animals immunised with CD virus were tested (1) with CD, (2) with constituents of CD, and (3) as under :—

	RESULT.
1 With Rustenburg (2).. .. .	Distinct reaction.
1 „ Natal, Rhodesia, and Lourenco Marques	„ „
1 „ Middelburg and Lydenburg	Reaction and dikkop.
1 „ Pretoria and Potchefstroom	No reaction.
1 „ Zoutpansberg and Waterberg	„
1 „ High Veld	Distinct reaction.
1 „ Tzaneen	Slight reaction.
1 „ OTB	No reaction.

The total percentage of reactions = 62 per cent.

10. Of 10 animals which were immunised with CD and tested with CD or Tzaneen for the first time, and secondly with CD, 1 showed a distinct reaction to this second test; [this animal (3183) had shown a reaction to the immunisation, and a reaction to the first test with Tzaneen, and a third reaction when tested with the virus with which it was originally immunised].

11. Four animals immunised with CD, and tested for the first time with a non-constituent (Simpson) did not react, neither did they when tested with a second non-constituent (Potgietersrust).

RÉSUMÉ.

1. The immunisation of mules with a polyvalent virus and an adequate or inadequate serum resulted in an immunity which was not complete. When tested later with the corresponding virus the immunity was broken to the extent of 14 per cent. reactions.
2. The immunity obtained with an inadequate serum and virus was broken by the same virus to the extent of 12 per cent. reactions.
3. The immunity obtained with an adequate serum and virus was broken by the same virus to the extent of 20 per cent. reactions.
4. The immunity obtained by the CD virus was broken by constituents to the extent of 22 per cent. reactions, and no deaths.
5. The immunity obtained by CD virus was broken by non-constituents to the extent of 7 per cent. deaths.
6. The immunity obtained by two injections of CD virus was broken by constituents of this virus to the extent of 82 per cent. reactions.
7. The immunity obtained by two injections of CD virus and one injection of a constituent was broken by constituents to the extent of 62 per cent. reactions.

CONCLUSIONS.

1. Animals immunised with a polyvalent virus and tested with the same virus show reactions when subsequently retested with the identical virus.
2. When the immunity was tested with constituents of this virus, break-downs occurred to a large extent in the first test, and even in the second test, showing that the virus, considered to be polyvalent, did not contain all the constituents which were originally mixed. The fact remains, however, that none of the tested animals died, thus showing that the immunity resulting from the CD virus protected against any of the constituents.
3. For the purposes of the practice, where such severe tests are hardly encountered, the immunity given by this CD virus should prove sufficient.

E.—THE LOSS OF VIRULENCY OF HORSE-SICKNESS VIRUS IN PRACTICE.

On page 85 of my Annual Report for 1906-07 I quoted an instance where a virus (Tzaneen) which had proved virulent on the station became inert after it was introduced into practice. The virus was recalled and again tested on the station, when the virulency was proved beyond doubt, and since another virus, which had formerly given typical horse-sickness reactions, subsequently failed to do so on the station, a thorough investigation was made.

EXPERIMENT No 1.

With virus which became inert in vitro.

Virus 2199 (horse, origin Tzaneen, twelfth generation) had proved to be so virulent in the experiments with serum that it caused reactions in animals previously immunised against the first generation of the same strain.