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THE AUSTRALIAN EPIDEMIC, 1914.

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IN November, 1914, the Director of the Lister Institute sent me a sealed tube containing dried crusts or scabs which were stated to have been removed from patients, subjects of the Australian epidemic earlier in the year. Dr Martin requested me to investigate any ascertainable facts as to their pathological nature, and what relation, if any, they bore to vaccinia on the one hand and to variola on the other.

The clinical aspect of the epidemic has been described by Dr Armstrong[1], while certain bacteriological investigations of the disease have been reported by Drs J. Burton-Cleland and E. W. Ferguson[2].

Although the majority of the medical authorities in Australia are stated to have regarded the epidemic as one of small-pox, probably of a slight nature, doubt appears to have existed in the minds of some of the observers as to whether this was actually the case, and the present investigation is an outcome of this uncertainty.

The crusts or scabs had been forwarded to England in cold storage from Australia, and there were in all some 40 gms. of them. Throughout the time of the investigation the crusts were stored in a desiccator at a temperature of about 4° C., small quantities being removed from time to time for use as required. The material for the inoculation of animals was invariably composed as follows, one part of crusts ground up in a pestle and mortar with four times its own bulk of 50 per cent. glycerine and water, the mixture being made immediately before use. The variolar material used as a control was from a case of confluent small-pox in a man, and the specific activity of this was ascertained by inoculation on monkeys; it was strongly active throughout. The vaccine lymph

[1] *Proceedings of the Royal Society of Medicine, Section of Epidemiology and State Medicine,* vol. VIII. No. 2, p. 1.
[2] *Ibid.* p. 19.

used as another control was seed-lymph used for the production of lymph at these laboratories and was very active, as shown by frequent inoculations on calves.

I.

The first series of animal experiments was made on guinea-pigs, in view of the fact that some observers had expressed the opinion that the Australian disease was a form of modified small-pox; and modification, if sufficiently extended, might give the material a vaccine character. Drs J. Burton-Cleland and E. W. Ferguson had found that the material which they employed gave typical vaccine vesicles when inoculated direct on bovines. As stated in a previous paper on vaccinia[1], if a non-immune buck guinea-pig be vaccinated on the scrotum, vaccine vesicles develop with great facility about 72 hours later. The present experiments were made in a place remote from calf vaccine work, and the technique was arranged to prevent any infection being conveyed outside. 70 buck guinea-pigs were inoculated on the scrotum with material from the Australian disease. No trace either of vesiculation or of any reaction of any kind was subsequently noted, the small incisions healing normally. 70 control pigs, inoculated at the same time and in the same manner with calf vaccine, developed in each case typical vaccine vesicles 72 hours later.

This failure to react to the crusts is no proof that the cases from which the crusts were taken were not small-pox, but it is of evidential value in tending to show that whatever modification the disease may have undergone from ordinary small-pox in its transition to "mild" small-pox (Armstrong, Burton-Cleland, Ferguson), such transition had not carried it to vaccinia. Subsequent experiments showed that the failure of the material to react was not due to loss of its own specific activity.

II.

The second series of inoculations was made on calves. The experiments on Calves 1–4 were made in the stables where routine calf vaccination is carried out, at the Lister Institute. Those on Calves 5 and 6 were made in a place remote from all such work, and the technique was arranged to obviate the possibility of any infection being conveyed away from the animals.

[1] Green, *Journal of Hygiene*, 1914.

CALF 1 (Heifer). (*a*) Was inoculated on a shaved area on the abdomen with the Australian disease. 120 hours later there were found along the lines of inoculation a series of yellow vesicles about the size of lentils; the appearance was unlike that of ordinary vaccine vesicles, being much smaller and more discrete. On the other hand their appearance was unlike that resulting from an inoculation of small-pox material direct from man to the calf, being, from my experience, too developed, even for a first inoculation positive result.

(*b*) At the same time a second area was inoculated with calf vaccine, and 120 hours later typical vaccine vesicles had developed on the site, these bearing practically no resemblance to the small yellow vesicles of the Australian disease.

CALF 2 (Bull). (*a*) Was inoculated on the shaved scrotum with the Australian disease. 120 hours later small yellow vesicles had developed as in the case of Calf 1.

(*b*) At the same time an area on the abdomen was inoculated with calf vaccine, and 120 hours later typical vaccine vesicles of first-class quality, and bearing no resemblance to the vesicles of the Australian disease, had developed here. The results on this calf resembled those on Calf 1.

CALF 3 (Heifer). (*a*) Was inoculated on a shaved area on the abdomen with vesicular contents removed from the Australian disease vesicles of Calf 1. The vesicular material had been removed at 120 hours after inoculation, and had been stored for two weeks at 4° C. 120 hours later the result on this site was *nil*.

(*b*) A second area was inoculated with similarly stored vesicular material from the Australian disease vesicles of Calf 2. 120 hours later vesicles had developed at the site, whose appearance was somewhat suggestive, but not convincing, of poor class vaccine vesicles. Passage through three subsequent calves failed to improve the quality of this vesicular material, *i.e.* to make it resemble vaccinia more closely clinically.

(*c*) A third area was inoculated with vaccine lymph and 120 hours later normal vaccine vesicles of good class had developed.

CALF 4. (*a*) A shaved area on the abdomen was inoculated with the stored material of the Australian disease vesicles of Calf 1. 120 hours later the result was *nil*.

(*b*) A second area on the abdomen was inoculated with stored material of the Australian disease vesicles from Calf 2. 120 hours

later vesiculation had resulted, the vesicles resembling somewhat poor class vaccine vesicles. Passage through two further calves failed to improve the quality of the vesicles.

(*c*) A third area was inoculated with vaccine lymph and 120 hours later normal vaccine vesicles of good class had developed.

CALF 5. Was inoculated with vesicles of the Australian disease removed from Monkey No. 2 (*vide* series of monkey experiments). Vesicles resembling moderately fair quality vaccine vesicles had developed 120 hours later.

CALF 6. Was inoculated with the Australian disease vesicles removed from Monkey No. 4. 120 hours later vesicles resembling vaccine vesicles of moderately fair quality had developed.

CALF 7. (*a*) A shaved area on the abdomen was inoculated with the Australian disease. 120 hours later there was a distinct reaction, with small papules and vesicles along the lines of inoculation, the result being similar to that of Calf 1, but not quite so marked. Here again the result was not typical of vaccinia, and it was too marked for variola inoculation direct from man.

(*b*) A fortnight later a second shaved area on the abdomen was inoculated with:

(1) Variolous material direct from a case of confluent small-pox in a man. 120 hours later the results were *nil*, no reaction of any kind being noticeable;

(2) Vaccine lymph. 120 hours later typical vaccine vesicles, but not of first-class quality, had developed.

CALF 8. (*a*) A shaved area on the abdomen was inoculated with:

(1) Vaccine. 120 hours later typical vaccine vesicles had developed.

(2) Variolous material from man, passed once through a monkey, on which it had given marked typical vesiculation. 120 hours later small vesicles had developed, not typical of vaccinia (but not so well developed as those of Australian disease); they approximated more to the Australian disease vesicles than to the vaccinal vesicles however.

(*b*) A fortnight later another area on the abdomen was shaved and inoculated with Australian crust emulsion. 120 hours later there was slight vesiculation at the site about the same appearance as on Calf 5, resulting from inoculation with Australian disease.

The results of the above experiments are more easily seen in Table I.

There are thus, with regard to these calf experiments, two main questions to consider:

1. *The likeness-relation of the Australian disease to Vaccinia and to Variola.*

The most prominent fact in all the calf experiments was that the vesiculation of the Australian disease, such as it was, was not generally typical of the vaccinia vesicles on the same calf, nor, I may be allowed to remark, was it typical of any vaccine vesicles that I remember on any calf. After passage through a monkey, however, the material gave rise to vesicles undistinguishable from those of moderately fair vaccine vesicles. The general impression arrived at was that the Australian disease, both by the appearance of the vesicles, and by their time-development, possessed some vaccino-variolal relationship. Allowing that there were possibilities of such a relationship, the Australian disease would have to be classed as distinct on the one hand from vaccinia in its lesser ability to produce typical vaccinal vesiculation, though it approximated more closely after passage through a monkey; and on the other hand from variola, for without doubt it possessed a greater facility for vesiculating on a calf than did the variola in the above experiments.

In the foregoing experiments the vesicle-likeness was consistent, but it differed widely from the vesicle-likeness reported in the experiments by Drs Burton-Cleland and Ferguson. In the experiments by these observers there was a tendency for vesicles to develop very freely at any time, even in the case of repeated re-vaccinations at short intervals.

2. *The immunity-relation of the Australian disease to Vaccinia and to Variola.*

There are only two sets of data from which to draw deductions, Exps. 7 and 8 in Table I. This scarcity is due to two causes: (*a*) the comparative difficulty of obtaining calves at the present time, and maintaining them for a period necessary for the development of possible immunity and obtaining re-vaccination results; (*b*) the inferiority of the results obtained from inoculating calves with the Australian disease, and incidentally with variola—even when this had been passed through a monkey.

Taking these two sets of data for what they are worth, however, it appeared that successful vaccination with variola and vaccinia afforded no protection against subsequent inoculation with Australian disease, and the Australian disease gave no protection against vaccinia. The

failure of variola to "take" on Calf 5 is of no account, as variolous material inoculated on the calf direct from man (*i.e.* not passed through a monkey) rarely gives definite vesiculation.

TABLE I.

Calf Experiments.

Calf No.	Vaccination	Result	Vaccination	Result
1.	*A*	+		
		+		
	L	+		
		+		
2.	*A*	+		
		+		
	L	+		
		+		
		+		
		+		
3.	*A* (from Calf 1)	−		
	L (from Calf 2)	+		
	L	+		
		+		
		+		
4.	*A* (from Calf 1)	−		
	A (from Calf 2)	+		
	L	+		
		+		
		+		
5.	*A*	+		
	(from Monkey No. 2)	+		
		+		
		+		
6.	*A*	+		
	(from Monkey No. 4)	+		
		+		
7.	*A*	+	*V*	−
			L	+
				+
				+
				+
8.	*L*	+		
		+		
		+		
		+		
	V	+	*A*	+
	(Passed once through monkey),			

A = Australian disease, *L* = Calf lymph, *V* = Variola.

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These two results, showing no protection, are in accord with the majority of the results of the calf experiments of Cleland and Ferguson. The accompanying Table II has been compiled for the sake of easier reference from the paper of Cleland and Ferguson[1], which should of course be consulted for full information.

It will be seen that in three instances vaccination gave a positive result following repeated successful reactions with the Australian

TABLE II.

Cleland and Ferguson's Calf Experiments.

Calf No.	2/11	4 days later	21/10	4th day?	7/11	4th day?	24/11	4th day?	11/12	4th day?	30/12	4th day?
1.	*A*	+ ?	*L*	+	*A*	+	*A*	−	*A*	+	*L*	+
				+								
				+								
	16/10		7/11		24/11		11/12		30/12			
2.	*A*	+	*A*	+	*A*	−	*A*	+	*L*	+		
				+				+		+		
	7/11		24/11		11/12		30/12					
3.	*A*	+	*A*	+	*A*	+	*L*	+				
		+										
		+										
	11/12		30/12									
4.	*A*	+	*L*	−								
		+										
		+										
	24/11		17/12		9/1							
5.	*L*	+	*L*	−	*A*	+						
		+										
		+										
	15/1											
6.	*A*	+										
		+										
		+										
	15/11		22/1	9 days later								

Monkey Experiments.

1.	*A*	+	*L*	− ?
	22/1	9 days later		
2.	*L*	?		

A = Australian disease, *L* = calf lymph.

[1] *Proceedings of the Royal Society of Medicine, Section of Epidemiology and State Medicine*, Vol. VIII. No. 2, p. 1.

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disease, and only one case a negative. In one case, Calf 5, the authors obtained a positive result of Australian inoculation following successful vaccination, *i.e.* complete protection was afforded only in 25 per cent. of the cases. In view of this fact it is difficult to understand Cleland and Ferguson's claim—"This inoculated disease and vaccinia are mutually more or less completely protective against each other provided sufficient area is inoculated." It would seem, indeed, that the number of Cleland and Ferguson's experiments is far too small to permit of the attempt to select one of the five (the sixth was not, of course, a cross immunity experiment at all) as establishing a law. The only deduction that it would seem safe to draw is the obvious one before mentioned, that 75 per cent. of the cases of inoculated Australian disease failed to protect against subsequent vaccination.

As before stated, the results of the experiments on calves in the present series of experiments were not such as to allow one to place much reliance on them in any way, and for this reason the series of experiments was not extended, extension preferably being made with monkey experiments, which gave very much better inoculation results.

III.

The third series of experiments was made on rhoesus and bonnet monkeys. The work was done in a place remote from all calf vaccine work, in order that no question of any accidental inoculation could arise, and under conditions of strict isolation to prevent the possibility of any infection being conveyed away from the monkeys. Each animal during the experimental period was kept in a separate cage by itself. Each animal was inoculated on an area of shaved skin over the scapulae, in order to reduce to a minimum the prospect of the results being spoilt by scratching. Each shaved area was about 6 square cm., and two linear incisions were made close together, allowing sufficient room for subsequent inoculations.

Monkey 1. *Inoculation.* 1. xii. 14. Vaccinia.

Result. Typical vaccination vesicles of rather poor quality.

Inoculation. 12. i. 15. Australian disease.

Result. Definite vesiculation undistinguishable from results of calf vaccine of poor quality.

Conclusion. Here a successful vaccination with vaccinia afforded no protection against the subsequent inoculation of Australian disease.

MONKEY 2. *Inoculation.* 1. xii. 14. Australian disease.

Result. Well developed vesicles, resembling typical vaccine vesicles of good quality. Subsequent inoculation on Calf 5 gave vesiculation resembling that of calf vaccine.

Inoculation. 12. i. 15. Vaccinia.

Result. 19. i. 15. Full well developed vesicles of "good" quality.

Inoculation. 9. ii. 15. On one area vaccinia, and on another Australian disease.

Result. 16. ii. 15. *Nil* in each case.

Conclusion. This experiment suggested that while Australian disease failed to protect against vaccinia, the original inoculations of vaccine and Australian disease afforded protection against subsequent inoculation of the virus of these diseases—lymph against lymph and Australian disease against Australian disease.

MONKEY 3. *Inoculation.* 1. xii. 14. Australian disease.

Result. 8. xii. 14. Definite vesiculation, but tendency for discrete vesiculation to form along the lines of incision and for vesicles to be irregular.

Inoculation. 12. i. 15. Vaccinia.

Result. 19. i. 15. Typical vesiculation with tendency to irregularity.

Conclusion. There was manifestly no protection afforded here by the Australian disease against vaccinia.

MONKEY 4. *Inoculation.* 8. xii. 14. Australian disease.

Result. 15. xii. 14. Well developed vesicles undistinguishable from vaccine vesicles of good quality. This vesicular material inoculated on Calf 6 gave vesicles resembling vaccine vesicles of fair quality.

Inoculation. 12. i. 15. Vaccinia.

Result. 19. i. 15. Typical vesiculation of good quality.

Inoculation. 9. ii. 15. Australian disease on one area, vaccinia on another.

Result. 16. ii. 15. *Nil* in each case.

Conclusion. The experiment would appear to indicate that Australian disease failed to protect against subsequent vaccinia, but that these two diseases protected against a further subsequent double inoculation on separate areas of the same diseases.

MONKEY 5. *Inoculation.* 9. ii. 15. Australian disease.

Result. 16. ii. 15. Rather poor vesiculation.

Inoculation. 2. iii. 15. Variola.

Result. 9. iii. 15. *Nil.*

Conclusion. Australian disease possibly protected against variola.

MONKEY 6. *Inoculation.* 9. ii. 15. Australian disease.

Result. 16. ii. 15. Good vesiculation.

Inoculation. 2. iii. 15. Vaccinia on one area, variola on the other.

Result. 9. iii. 15. Vaccinia doubtful, variola good vesiculation.

Conclusion. Australian disease did not protect against variola, and probably not against vaccinia.

MONKEY 7. *Inoculation.* 9. ii. 15. Vaccinia.

Result. 16. ii. 15. Good vesiculation.

Inoculation. 2. iii. 15. Vaccinia on one area, variola on another.

Result. 9. iii. 15. Vaccinia *nil*, variola *nil.*

Inoculation. 23. iii. 15. Australian disease.

Result. 30. iii. 15. Slight papulation.

Conclusion. Vaccinia protected against vaccinia and variola, but not against Australian disease.

MONKEY 8. Died before any cross immunisation could be attempted.

MONKEY 9. Died before any cross immunisation could be attempted.

MONKEY 10. *Inoculation.* 9. ii. 15. Variola.

Result. 16. ii. 15. Fairly good vesiculation.

Inoculation. 2. iii. 15. Australian disease on one area, vaccinia on another.

Result. 9. iii. 15. Australian disease good vesiculation, vaccinia *nil.*

Conclusion. Variola protected against vaccinia, but not against Australian disease.

MONKEY 11. *Inoculation.* 16. ii. 15. Variola.

Result. 23. ii. 15. Fair vesiculation.

Inoculation. 2. iii. 15. Australian disease on one area, vaccinia on another.

Result. 9. iii. 15. Australian disease definite but slight vesiculation, vaccinia *nil.*

Conclusion. Variola protected against vaccinia, but not against Australian disease, or only to a very moderate degree.

MONKEY 12. *Inoculation.* 23. ii. 15. Mixed aerobic and anaerobic growth, cocci in broth, cultivated from Australian disease crusts.

Result. 2. iii. 15. *Nil.*

Inoculation. 2. iii. 15. Australian disease.

Result. 9. iii. 15. Definite vesiculation.

Inoculation. 23. iii. 15. Vaccinia on one area, variola on another.

Result. 30. iii. 15. Vaccinia fair vesiculation, variola *nil.*

Conclusion. (1) Cocci cultivated from crusts not pathogenic. (2) Australian disease did not protect against vaccinia, but may have protected against variola.

MONKEY 13. *Inoculation.* 2. iii. 15. Vaccinia on one area, variola on another.

Result. 9. iii. 15. Vaccinia fair vesiculation, variola strongly marked vesiculation.

Inoculation. 23. iii. 15. Australian disease.

Result. 30. iii. 15. Fair vesiculation.

Conclusion. Vaccinia and variola have not protected against Australian disease.

MONKEY 14. *Inoculation.* 2. iii. 15. Australian disease on one area, variola on another.

Result. 9. iii. 15. Australian disease fair vesiculation, variola poor vesiculation.

Inoculation. 23. iii. 15. Vaccinia.

Result. 30. iii. 15. Good vesiculation.

Conclusion. Australian disease does not protect against vaccinia, but neither did variola protect against vaccinia; it must be noted that the variola gave poor result.

MONKEY 15. *Inoculation.* 10. iii. 15. Variola.

Result. 17. iii. 15. Good vesiculation.

Inoculation. 23. iii. 15. Australian disease on one area, vaccinia on another.

Result. 30. iii. 15. Australian disease fair vesiculation, vaccinia *nil.*

Conclusion. Variola protected against vaccinia, but not against Australian disease.

MONKEY 16. *Inoculation.* 10. iii. 15. Variola.

Result. 17. iii. 15. Very marked vesiculation.

Inoculation. 23. iii. 15. Australian disease on one area, vaccinia on another.

Result. 30. iii. 15. Australian disease *nil*, vaccinia poor vesiculation.

Conclusion. Variola protected against Australian disease, and not against vaccinia, or only very slightly.

MONKEY 17. *Inoculation.* 10. iii. 15. Variola.

Result. 17. iii. 15. Rather poor vesiculation.

Inoculation. 23. iii. 15. Australian disease on one area, vaccinia on another.

Result. 30. iii. 15. Australian disease *nil*, vaccinia poor vesiculation.

Conclusion. Variola protected against Australian disease, but not against vaccinia, or only partially.

MONKEY 18. *Inoculation.* 10. iii. 15. Australian disease.

Result. 17. iii. 15. Vesiculation.

Inoculation. 23. iii. 15. Vaccine on one area, variola on another.

Result. 30. iii. 15. Vaccine first-class vesicles, variola fairly good vesicles.

Conclusion. Australian disease gave no protection against vaccine or variola.

MONKEY 19. *Inoculation.* 10. iii. 15. Australian disease.

Result. 17. iii. 15. Vesiculation fair.

Inoculation. 23. iii. 15. Vaccinia on one area, variola on another.

Result. 30. iii. 15. Poor vesiculation in each case.

Conclusion. Australian disease afforded no protection against vaccinia or variola, or only partial protection.

MONKEY 20. *Inoculation.* 10. iii. 15. Australian disease.

Result. 17. iii. 15. Vesiculation.

Inoculation. 23. iii. 15. Vaccinia on one area, variola on another.

Result. 30. iii. 15. Vaccinia good vesicles, variola very good.

Conclusion. Australian disease had afforded no protection against vaccinia or variola.

MONKEY 21. *Inoculation.* 10. iii. 15. Vaccinia.

Result. 17. iii. 15. Fair vesiculation.

Inoculation. 23. iii. 15. Australian disease on one area, variola on another.

Result. 30. iii. 15. Australian vesiculation good, variola *nil.*

Conclusion. Vaccinia afforded protection against variola, but not against Australian disease.

MONKEY 22. *Inoculation.* 10. iii. 15. Vaccinia.

Result. 17. iii. 15. Fair vesiculation.

Inoculation. 23. iii. 15. Australian disease on one area, variola on another.

Result. 30. iii. 15. Australian disease *nil*, variola *nil.*

Conclusion. Vaccinia protected against variola, and may have protected against Australian disease; at any rate the Australian disease failed to develop after previous successful vaccination.

The foregoing results have been condensed and tabulated in Table III.

TABLE III.

Monkey No.	Inoculation and Date	Result	Inoculation and Date	Result	Inoculation and Date	Result
1.	1/12/14 *L*	8/12/14 +	12/1/15 *A*	19/1/15 + Died		
2.	1/12/14 *A*	8/12/14 + + +	12/1/15 *L*	19/1/15 + + +	9/2/15 *L* ⋮ *A*	16/2/15 – ⋮ – Died
3.	1/12/14 *A*	8/12/14 + +	12/1/15 *L*	19/1/15 + + Died		
4.	8/12/14 *A*	15/12/14 + + +	12/1/15 *L*	19/1/15 + + +	9/2/15 *L* ⋮ *A*	16/2/15 – ⋮ –
5.	9/2/15 *A*	16/2/15 +	2/3/15 *V*	9/3/15 –		
6.	9/2/15 *A*	16/2/15 + +	2/3/15 *V* ⋮ *L*	9/3/15 + ⋮ ?		
7.	9/2/15 *L*	16/2/15 + + +	2/3/15 *V* ⋮ *L*	9/3/15 – ⋮ –	23/3/15 *A*	30/3/15 +
8.	9/2/15 *L*	16/2/15 + Died				
9.	9/2/15 *V*	16/2/15 + Died				
10.	9/2/15 *V*	16/2/15 + +	2/3/15 *A* ⋮ *L*	9/3/15 + ⋮ –		
11.	16/2/15 *V*	23/2/15 +	2/3/15 *A* ⋮ *L*	9/3/15 + ⋮ –		
12.	23/2/15 Cocci from *A*	2/3/15 –	2/3/15 *A*	9/3/15 +	23/3/15 *L* ⋮ *V*	30/3/15 + ⋮ –
13.	2/3/15 *L* ⋮ *V*	9/3/15 + ⋮ +	23/3/15 *A*	30/3/15 +		
14.	2/3/15 *A* ⋮ *V*	9/3/15 + ⋮ +	23/3/15 *L*	30/3/15 + + +		
15.	10/3/15 *V*	17/3/15 + +	23/3/15 *A* ⋮ *L*	30/3/15 + ⋮ –		

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TABLE III—(*continued*).

Monkey No.	Inoculation and Date	Result	Inoculation and Date	Result
	10/3/15	17/3/15	23/3/15	30/3/15
16.	*V*	+	*A* : *L*	− : +
		+		
		+		
	10/3/15	17/3/15	23/3/15	30/3/15
17	*V*	+	*A* : *L*	− : +
	10/3/15	17/3/15	23/3/15	30/3/15
18.	*A*	+	*L* : *V*	+ : +
				+ : +
				+ :
	10/3/15	17/3/15	23/3/15	30/3/15
19.	*A*	+	*L* : *V*	+ : +
	10/3/15	17/3/15	23/3/15	30/3/15
20.	*A*	+	*L* : *V*	: +
				+ : +
				+ : +
				: +
	10/3/15	17/3/15	23/3/15	30/3/15
21.	*L*	+	*A* : *V*	+ : −
	10/3/15	17/3/15	23/3/15	30/3/15
22.	*L*	+	*A* : *V*	− : −

V = variola, *L* = calf lymph, *A* = Australian disease.

As in the case of the calf experiments it will be convenient to consider this series of monkey experiments in two main aspects.

1. *The likeness-relation of the Australian disease to Vaccinia and to Variola.*

In this series (as has been previously remarked in the general statement) there is little or none of the ambiguity attaching to the question that appeared in the case of the calf experiments. Without exception the Australian disease vesicles resembled typical vaccine vesicles so closely that it was impossible to distinguish one condition from the other. In time-relation too the two appeared identical, there was indeed no apparent clinical distinction.

2. *The immunity-relation of the Australian disease to Vaccinia and to Variola.*

For the clearer consideration of this problem the experiments have been divided into sub-series in the following four tables (Tables IV, V, VI and VII).

In the first sub-series (*Table IV*). Those experiments have been collected in which monkeys were in the first place inoculated with

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TABLE IV.

(*Sub-series* 1.)

Monkey No.	Inoculation and Date	Result	Inoculation and Date	Result	Inoculation and Date	Result
	1/12/14	8/12/14	12/1/15	19/1/15	9/2/15	16/2/15
2.	*A*	+ + +	*L*	+ + +	*L* ⋮ *A*	– ⋮ – Died
	1/12/14	8/12/14	12/1/15	9/2/15		
3.	*A*	+ +	*L*	+ Died		
	8/12/14	15/12/14	12/1/15	19/1/15	9/2/15	16/2/15
4.	*A*	+ + +	*L*	+ +	*L* ⋮ *A*	– ⋮ –
	9/2/15	16/2/15	2/3/15	9/3/15		
5.	*A*	+	*V*	–		
	9/2/15	16/2/15	2/3/15	9/3/15		
6.	*A*	+ +	*L* ⋮ *V*	+ ⋮ +		
	10/3/15	17/3/15	23/3/15	30/3/15		
18.	*A*	+	*L* ⋮ *V*	+ + + + ⋮ + + ⋮ +		
	10/3/15	17/3/15	23/3/15	30/3/15		
19.	*A*	+	*L* ⋮ *V*	+ ⋮ +		
	10/3/15	17/3/15	23/3/15	30/3/15		
20.	*A*	+	*L* ⋮ *V*	+ ⋮ + + ⋮ + + ⋮ +		

V = variola, *L* = calf lymph, *A* = Australian disease.

TABLE V.

(*Sub-series* 2.)

Monkey No.	Inoculation and Date	Result	Inoculation and Date	Result	Inoculation and Date	Result
	1/12/14	8/12/14	12/1/15	19/1/15		
1.	*L*	+	*A*	+ Died		
	9/2/15	16/2/15	2/3/15	9/3/15	23/3/15	30/3/15
7.	*L*	+ + + +	*L* ⋮ *V*	– ⋮ –	*A*	+
	10/3/15	17/3/15	23/3/15	30/3/15		
21.	*L*	+ +	*A* ⋮ *V*	+ ⋮ –		
	10/3/15	17/3/15	23/3/15	30/3/15		
22.	*L*	+ +	*A* ⋮ *V*	– ⋮ –		

A = Australian disease, *V* = variola, *L* = calf lymph.

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TABLE VI.

(*Sub-series* 3.)

Monkey No.	Inoculation and Date	Result	Inoculation and Date	Result
10.	9/2/15 *V*	16/2/15 + +	2/3/15 *A* ⋮ *L*	9/3/15 + ⋮ −
11.	16/2/15 *V*	23/2/15 + +	2/3/15 *A* ⋮ *L*	9/3/15 + ⋮ −
15.	10/3/15 *V*	17/3/15 + +	23/3/15 *A* ⋮ *L*	30/3/15 + ⋮ −
16	10/3/15 *V*	17/3/15 + + + +	23/3/15 *A* ⋮ *L*	30/3/15 − ⋮ +
17.	10/3/15 *V*	17/3/15 +	23/3/15 *A* ⋮ *L*	30/3/15 − ⋮ +

A = Australian disease, *V* = variola, *L* = calf lymph.

TABLE VII.

(*Sub-series* 4.)

Monkey No.	Inoculation and Date	Result	Inoculation and Date	Result
12.	2/3/15 *A* ⋮ *L*	9/3/15 + ⋮ + +	23/3/15 *V*	30/3/15 −
13.	2/3/15 *L* ⋮ *V*	9/3/15 + ⋮ + + ⋮ + +	23/3/15 *A*	30/3/15 + +
14.	2/3/15 *A* ⋮ *V*	9/3/15 + ⋮ + ⋮ +	23/3/15 *L*	30/3/15 + + +

A = Australian disease, *V* = variola, *L* = calf lymph.

Australian disease and subsequently with vaccinia and variola—one after the other, or simultaneously.

In the second sub-series (*Table V*). Those experiments have been collected in which monkeys were in the first place inoculated with vaccinia, and subsequently with Australian disease and variola—one after the other, or simultaneously.

In the third sub-series (*Table VI*). Those experiments have been collected in which monkeys were in the first place inoculated with variola, and subsequently with Australian disease and vaccinia—in every case simultaneously.

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In the fourth sub-series (*Table VII*). Those experiments, only three *m* number, have been collected which do not conform with those of the previous groups, and consist of initial double inoculations, with subsequent cross immunisation inoculations, as shown in the Table.

In sub-series 1. The eight initial inoculations of Australian disease, as shown in Table IV, were all successful in yielding vesicles, and some of the vesicles were of noticeably good quality, and well developed. Of these there were three cases of subsequent inoculation with vaccinia alone which all yielded vesicles, one case of subsequent inoculation with variola, which gave no reaction, and four cases of subsequent inoculation with vaccinia and variola simultaneously, which all gave definite typical vesiculation. In the whole eight cases therefore of this sub-series there was only one case (Monkey 5) to suggest that Australian disease afforded protection against vaccinia or variola. On the other hand the remaining seven cases indicate that no protection whatever had been afforded by Australian disease against subsequent vaccinia or variola. It only seems possible therefore to state that the percentage of immunity afforded by Australian disease in this subsection was *nil* or very small.

That these animals were capable of developing immunity is shown in the cases of Monkeys 2 and 4, where re-inoculations of Australian disease and vaccinia respectively gave negative results.

In sub-series 2 (*Table V*). Consisting of four experiments, all initial inoculations with vaccinia gave typical vesiculation. All four animals were subsequently inoculated with Australian disease, and in three instances this was followed by vesiculation typical of vaccinia vesicles; the remaining one failed to react—a result of 75 per cent. against, and 25 per cent. for protection.

In sub-series 3 (*Table VI*). The five monkeys were all primarily inoculated with variola and gave vesiculation undistinguishable from the vesicles of vaccinia and Australian disease. Each case was subsequently inoculated with Australian disease and vaccinia simultaneously; in three cases the Australian inoculation was followed by typical vesiculation and in two cases by no reaction, indicative of conferred immunity in 40 per cent. and absence of conferred immunity in 60 per cent. If we take the cases of sub-series 2 and 3 together, vaccinia or variola may have conferred immunity in 33·3 per cent. cases, and did not confer it in 66·6 per cent. It must be noted however, that in sub-series 3 the possible protection afforded against Australian disease occurred in the same percentage as the protection afforded by variola against vaccinia.

In sub-series 4 (*Table VII*). In Monkey No. 12, Australian disease or vaccinia protected against subsequent inoculations with variola; in Monkey No. 13, vaccinia and variola failed to protect against Australian disease; and in Monkey No. 14 Australian disease and variola failed to protect against vaccinia.

Taking the four sub-series as a whole, evidence as to the ability of Australian disease to protect against vaccinia or variola was small or absent, but vaccinia or variola possibly protected against Australian disease in 30 per cent. of the cases; vaccinia protected against variola in 100 per cent. of the cases, and variola protected against vaccinia in 50 per cent. of the cases.

In comparing the foregoing percentage of vaccinia and variola protection one against the other, it is important to remember that the variola was inoculated experimentally, *i.e.* the infective material gained both a more intimate contact with the tissues than would be the case in usual variola infection, and was applied in much larger doses. The same point must be noted in connection with the Australian disease, and the fact that protection was afforded against variola and not against Australian disease, has the more significance, inasmuch as it occurred under this more stringent application of infection.

It may be noted in the foregoing text that when vaccinia or variola has failed to develop after previous successful inoculation with Australian disease the fact has been stated as such; but when variola has failed to develop after previous successful vaccination it is definitely assumed that this is in consequence of the vaccination. This variation of expression is owing to the circumstance that it is an accepted fact that vaccinia protected against variola, but it is not yet an accepted fact that Australian disease protects against vaccinia or variola.

Briefly stated, the conclusions to be drawn from the foregoing experiments as a whole are that, clinically, the Australian disease:

1. Bears no likeness-relation to vaccinia in guinea-pigs, possibly some slight likeness-relation to vaccinia in calves, also to variola in calves when the variola has been passed through a monkey; this relationship would seem to be an intermediate one, between vaccinia on one hand and variola on the other. In monkeys Australian disease is practically undistinguishable from vaccinia or variola.

2. From the monkey experiments, which afford the only evidence of value in this respect, the Australian disease bears a slighter immunity relationship to vaccinia and to variola than either vaccinia or variola bear to each other, and this in spite of the fact that the clinical

relationship of the Australian disease shows certain definite signs of being intermediate between vaccinia on the one hand and variola on the other.

SUMMARY.

(1) Inoculation of pathologically active crusts on guinea-pigs caused no reaction, and in this respect a marked difference was demonstrated clinically between the disease of which the crusts were a product, and vaccinia.

(2) On calves the disease tended to show some vaccino-variolal relationship, from the appearance of its vesicles, but it appeared to be distinct on the one hand from vaccinia, and from variola on the other.

(3) Calf experiments, two only in number, suggested that vaccinia and variola failed to protect against Australian disease, and that Australian disease failed to protect against vaccinia or variola.

(4) On monkeys the vesicles of Australian disease were undistinguishable in appearance from those of vaccinia or variola, but the evidence for immunity relationship between Australian disease and vaccinia and variola was of the slightest; Australian disease affording no protection against vaccinia or variola, vaccinia possibly protecting against Australian disease in 25 per cent. of the cases, and variola possibly protecting against Australian disease in 40 per cent. of the cases. Vaccinia protected against variola in 100 per cent. of the cases, and variola against vaccinia in 50 per cent. of the cases.