

of it. Extreme care has been taken, however, to make the number of the dots bear fairly accurately a general proportion to the density of the degeneration, and the same proportion in one drawing as in another.

---

“The Inter-relationship of Variola and Vaccinia.” By S. MONCKTON COPEMAN, M.A., M.D. Cantab., F.R.C.P. Communicated by LORD LISTER, F.R.S: Received November 13,—Read November 27, 1902.

The term “*variolæ vaccinae*” employed by Jenner, as a synonym for cow-pox, has been generally accepted as affording evidence that in so naming this disease, “small-pox of the cow,” he was desirous of placing on record his belief that cow-pox, or vaccinia, was intimately related to human small-pox, if indeed it were not directly derived from it.

This theory, however, appears to have found but scanty favour in Jenner’s day, and even at the present time the value of the practice of vaccination is, by some, impugned on the plea that inoculation of one disease—cow-pox—could not be expected to exert any really protective influence against the ravages of small-pox—a disease considered by them of totally different origin.

In the hope of obtaining definite information on the subject, many observers, during the long period which has elapsed since the introduction of vaccination, have set themselves the task of attempting to solve, by experimental methods, the problem of the true relationship of vaccinia to variola.

These attempts have been, for the most part, directed to the possibility of giving rise to cow-pox by the introduction, in one or another manner, of the virus of small-pox into the system of the bovine animal. In the great majority of such attempts, which have been much more numerous than is generally supposed, the results have been entirely negative, although so numerous have been the experimenters, who from time to time have attacked the problem, that the total number of instances in which an apparently successful result has been obtained, is now considerable.

So far as I am aware, the first recorded experiments are those of Gassner of Gunsberg, who, in 1801, succeeded, after no less than ten fruitless attempts, in directly variolating a cow with small-pox virus. The lymph thus obtained was employed for the vaccination of four children, from whom other seventeen were subsequently vaccinated. None of these exhibited any signs of small-pox.

It is impossible, here, to do more than mention the names of other investigators who have engaged in research of this nature at various times from the commencement of the last century up to the present time. I therefore merely append in the foot-note\* a list of such names, placed, as far as possible, in chronological order. It is a noteworthy fact that every observer mentioned, with the exception of Chauveau and his colleagues of the Lyons Commission, and Martin, claim to have obtained, on one or more occasions, positive results as regards the production of typical vaccinia, generally after one or more removes from the animal originally variolated.

But it must, I think, be admitted that many of the earlier experiments, more particularly, are practically worthless owing to the conditions under which they were carried out. Some of the main objections are based on the frequently concomitant use of vaccine and of variolous lymph on the same animal, and the want of care as to the cleanliness and freedom from vaccine contamination of lancets and "points" used in the experiments.

As regards my own work, carried out on similar lines to those adopted by previous observers, and of which a full account was published in the 'Journal of Pathology and Bacteriology,' in 1894, it may here be mentioned that I obtained an undoubtedly successful result in one series only, out of four attempts. In four subsequent variolation experiments, carried out several years later (1901) in connection with work, a detailed account of which is set out in the

\* Chronological list of observers who have carried out variolation experiments on bovines :—

1801.	Viborg .....	Copenhagen
1828.	McMichael .....	Egypt.
1830.	Sonderland .....	Barmen.
„	Numann .....	Utrecht.
1832.	Macphail .....	Baltimore.
1836.	Thiele .....	Russia.
„	Martin .....	Attleborough, Mass.
„	MacPherson .....	India.
1839.	Reiter .....	Munich.
„	Ceely .....	England.
1840.	Badeock .....	„
1863-65.	Chauveau .....	Lyons.
1868.	Shortt .....	India.
1871.	Chauveau .....	Lyons.
1881.	Voigt .....	Hamburg.
1886-90.	Fischer .....	Carlsruhe.
1889.	King .....	Madras.
1890-91.	Eternod and Haccius .....	Geneva.
1892.	Simpson .....	India.
„	Hime .....	England.
„	Klein .....	„
„	Copeman .....	„

present paper, my attempts at direct transference of human small-pox material to the calf met with no success.

All my earlier experiments were conducted at the Brown Institution, in order to avoid any possibility of contamination with vaccinia. As a further precaution new scalpels were used, which were invariably first carefully sterilised in the flame of a spirit lamp, and, after use, the table was, on each occasion, thoroughly washed with carbolic acid (1 in 20), while during the intervals of use it was kept exposed to the air under an open shed. Similar precautionary measures have been observed throughout the course of my later work.

The difficulty experienced by myself and the numerous other investigators, to whom reference has already been made, in attempts to transmit human small-pox directly to bovines, whether cows or calves, is not infrequently cited as a reason for regarding with distrust the theory expounded by Jenner, that cow-pox, whether carried through the horse as intermediary host or not, was originally derived from small-pox in the human being.

But a great deal, at any rate, of the small-pox which was prevalent at the time that Jenner lived and wrote was of that comparatively mild variety which, under the name of inoculated small-pox, was intentionally produced in healthy subjects, with the object of thereby conferring protection against subsequent attack by the disease in virulent form.

So mild indeed at times were the results of inoculations in the hands of such operators as Adams and the brothers Sutton that, as we learn from contemporary records, in many instances but little obvious effect was observed, with the exception of the local vesicle arising at the site of insertion of the small-pox virus, and the patients suffered but little inconvenience. Thus, more particularly in certain of Adams' cases, as may be gathered from his own account of the circumstances, the visible effect produced so closely resembled the results then beginning to be known as following on the Jennerian process of vaccination, that numbers of his patients were with difficulty persuaded that he had not, contrary to their desire, intentionally vaccinated rather than variolated them. The gradual evolution of a strain of lymph of such tenuity, according to Adams himself, was obtained by attention to the mode of life and general treatment of persons undergoing the process, together with careful selection of the sources (preferably the primary vesicle) from which the virus was obtained.

The majority of persons thus inoculated are not likely to have been incapacitated, as the result of the operation, to a much greater extent than are those who undergo efficient vaccination at the present day, and doubtless, therefore, they would be, for the most part, capable of following their ordinary avocations during the progress of the induced

disorder. On the other hand, this would hardly have been possible in the case of persons contracting small-pox in the ordinary way, among whom the disease was apt to exhibit such virulence as to account for the death of perhaps 50 per cent. of those attacked.

Not only were the effects following on inoculation comparatively mild, but the disease in this form was intentionally brought into many country districts which otherwise might not have become invaded by small-pox. In the light of these facts, it has for some time past been borne in upon my mind more and more convincingly that it was probably from the *inoculated* form of small-pox, rather than from the ordinary variety of the malady, that much, at any rate, of the cow-pox, in the pre-vaccination era, was derived. It is not difficult to understand how that the cracks so often found on the udders of cows might become infected by a milker with fingers contaminated by contact with the inoculation sore upon his arm.

I determined therefore, if possible, to put the matter to the test, and, learning that in Nubia, in Burmah, and in certain parts of India the inoculation of small-pox is still practised, I made numerous endeavours to obtain the necessary material, but unfortunately without success.

In default, therefore, of inoculated small-pox in the human subject, I made trial of the monkey, which, as I have shown in a previous communication to the Royal Society, is readily susceptible to the disease, the various phases of which in this animal closely resemble those observed in man, but in a much milder form; the occurrence of a generalised eruption being exceptional.

The different series of experiments, protocols of which I append, have been carried out at intervals, determined mainly by the possibility of procuring the necessary small-pox material. The work was commenced in April, 1898, with a supply of small-pox lymph received from the Medical Officer of Health for Middlesborough, in which town an epidemic of the disease was then in progress. For subsequent supplies I am indebted to the Medical Officer of Health and the Medical Superintendent of the Small-pox Hospital at Glasgow, to the Medical Superintendent of the West Ham Small-pox Hospital at Dagenham, near London, and to the Medical Superintendent of the Hospital Ships of the Metropolitan Asylums Board.

The methods employed in the investigation have been briefly as follows:—

*Collection of Material for Inoculation.*

In the first instance this was obtained in a manner similar to that formerly employed in obtaining human vaccine lymph. Discrete vesicles, mature, but still containing clear lymph, on one or another portion of the body of a patient suffering from small-pox, were punctured

with a sterilised lancet, and their fluid contents received into fine capillary tubes, which were subsequently sealed in the flame of a spirit lamp to admit of transport. This operation, however, is a most laborious one, and was subsequently abandoned, at my suggestion, in favour of collection, in the post-mortem room, of vesicle pulp at a suitable stage of the eruption, by means of a small Volkmann's spoon, after the fashion now invariably used in the Government lymph laboratories in obtaining, from the calf, material for the production of glycerinated lymph.

After removal from the body the small-pox pulp is first carefully weighed, and then ground up in a small glass mortar, with the gradual addition of usually four times its weight of a sterilised 50 per cent. solution of pure glycerine in normal saline solution. After thorough emulsification, what is not required for immediate use is stored in tubes, resembling small test-tubes, which are then corked, sealed with liquefied paraffin to which carbolic acid has been added, and set aside in a chamber kept at a temperature a few degrees above freezing point. Both storage-tubes and corks are sterilised before use.

Bacteriological examination by the method of plate-culture often shows a comparatively small number of extraneous micro-organisms in a specimen of small-pox emulsion prepared in the manner described, but whenever possible it has been stored at a temperature of about 15° C. for some weeks prior to using it for inoculation.

#### *Species and Age of Monkeys Inoculated.*

For my original experiments on the transference of human small-pox to the monkey, a brief account of which was presented to the Royal Society in 1893, I employed the rhesus monkey, for the reason that Professor Sherrington and myself had, at the time, a stock of these animals, which had been obtained for other experimental work. Having at that time obtained successful results in every one of my inoculations, I employed the same species of monkey in the greater number of the experiments comprised in the present research. As, however, during the progress of the work I learnt that Dr. Eilerts de Haan, who, in Batavia, had been working on similar lines to myself, had made use most successfully of the macaque monkey, I also obtained a few specimens of this species, in order to compare the results of variolation in these animals with those that I had previously observed in the rhesus monkey. But after two or three inoculations of the macaque with small-pox material, I came to the conclusion that the results following on the operation were not ordinarily as typical as in those experiments in which rhesus monkeys had been employed. At the same time the macaque is in this country more expensive and more difficult to obtain than the rhesus, so that I reverted to the use of the latter species in subsequent work.

It would appear also that, as in the human subject, young animals are more susceptible to small-pox than are adults, since it was in those instances in which monkeys probably not more than a year old were variolated that the most successful results were obtained. In one instance, however, in which the monkey was believed to be not more than a few months old, the extremely fine downy hair, after shaving, grew again so rapidly as to render somewhat difficult the photographing of the effect produced by the operation.

#### *Mode of Operation and Collection.*

In the earlier experiments inoculation of the monkey with human small-pox emulsion was carried out by rubbing it well into scarified patches or linear incisions of the skin of the upper arm or of the inside of the thigh, after previous shaving and cleansing of the skin. Subsequently, however, in accordance with the suggestion of Dr. de Haan, a shaved area on the back of the animal was utilised for inoculation. In this situation the results of the operation were found to be equally good, and there is less liability of damage to the vesicles from the monkey scratching itself.

The eruption having arrived at maturity, after the lapse of a period extending from five to eight days from inoculation, the altered epithelium was removed either with a small Volkmann's spoon or by scraping with a scalpel, after cleansing the inoculation area, between the blades of pressure forceps. The resulting epithelial pulp was then rubbed up in a small glass mortar, with the gradual addition of about six times its weight of normal saline solution, containing, when it was desired to preserve and purify the emulsion, 50 per cent. of glycerine.

Experience has shown that in monkeys a year or more old, which have been inoculated, the vesicular stage of the eruption is at its height, as was formerly observed in the human subject, by the eighth day; but in younger animals the process tends to be hastened, and in some of the later and most successful cases, the eruption was completely vesicular as early as the sixth day (120 hours). The particular *breed* of monkey does not appear to exert any influence in this respect.

#### *Transference to the Calf and Human Subject.*

The methods employed for transference of the localised disease in the monkey, after one or more passages through that animal, to the skin of the calf need not be set out in detail, being similar to those ordinarily used in the process of calf vaccination. But it may here be stated that at no stage of the investigation have these experimental calves been brought into contact with, or even placed in the same room as, the calves used in the current work of the Government Vaccine Establish-

ment. They were fed and otherwise attended to by a man specially detailed for the purpose. All instruments employed for the vaccination of monkeys, calves, or children were previously sterilised by boiling or passing through the flame of a spirit lamp. Attention may perhaps be called to the fact that the skin of the scrotum in the calf affords a specially favourable site for inoculation experiments, especially if, when the incisions are made, the skin is made tense by pressing down the testicles. The first transference from the monkey to the calf does not usually afford a perfect result. Indeed a second, third, or even later passage from calf to calf was usually required before the most typical vesiculation was obtained.

In certain cases children were vaccinated with lymph obtained from the experimental calves, and in all instances the resulting vaccination ran a perfectly normal course. With lymph of similar origin I also successfully vaccinated myself. But none of the strains of vaccine lymph, derived originally from human small-pox in the manner described, have been brought into general use.

## PROTOCOLS OF EXPERIMENTS.

### FIRST SERIES.

*February 21, 1898.*—Glycerinated samples of small-pox lymph received this day from Medical Officer of Health of Middlesbrough. Patients living and aged respectively 20, 27, and 34 years; all had been vaccinated in infancy.

*April 1.*—At Brown Institution, inoculated small rhesus monkey with small-pox emulsion of 21.ii.98 in five linear incisions on left arm, and in fourteen on abdomen, after previous shaving and cleansing of the skin, by means of soap and water, followed by warm boric acid lotion. Monkey isolated in separate room and attendant vaccinated as a precautionary measure.

*April 5.*—All insertions on both arm and abdomen evidently "taking."

*April 8.*—Distinct vesiculation at site of all incisions on arm and most of those on abdomen. Monkey etherised and substance of vesicles removed with sharp spoon into small previously weighed and sterilised test-tube. Scrapings weighed (0.6 gramme) and ground up with six times the weight of 50 per cent. watery solution of glycerine. Resulting emulsion taken up into twelve capillary glass tubes.

*April 13.*—Monkey looks well. All incisions healed up. No sign of generalised eruption.

### *Calf Experiments.*

*April 9.*—At the Animal Vaccine Establishment, Mr. Stott inoculated Calf No. 1 (No. 4363) on two scarified patches, in twelve incisions on scrotum, and forty-four in perineum and on abdomen, with contents of two capillary tubes of glycerinated pulp prepared from vesicles of monkey. Incisions made with scalpel previously sterilised.

*April 12 (72 hours).*—Practically nothing to be seen.

*April 14 (120 hours).*—All insertions on scrotum appear to have "taken," and, in addition, four (not quite so well) on abdomen. Insertions on perineum seem to have failed. Large bullous-looking vesicle on upper scarified patch.

From this and from the vesicular lines on scrotum and abdomen collected pulp by scraping, after clamping with compression forceps.

The same day (April 14), Calf No. 2 (No. 4369 in A.V.E. records) was inoculated on perineum, scrotum, and abdomen with material obtained from Calf No. 1.

*April 19.*—All inoculated lines, with exception of two on abdomen, "taken" well, eruption being markedly vesicular. Vesicles clamped and scraped; pulp being immediately employed for inoculation of Calf No. 3 (No. 4673 in A.V.E. records) in a number of long incisions on the perineum, scrotum, and abdomen.

*April 24.*—All lines of incision "taken" well; eruption perfectly typical of vaccinia.

From this calf, six children vaccinated at A.V.E. same day.

*May 1.*—Children returned for inspection, in ordinary course. All vaccinations completely successful. Photographed arms of two of these children, which presented most perfect eruption.

About a month later I hunted up the parents of all six children, when I learnt from the mothers' statements that in every case the vaccination had pursued a perfectly normal course.

#### SECOND SERIES.

*March 3, 1900.*—At the West Ham Borough Hospital, Dagenham, I removed small-pox vesicles from body of a man, æt. 56, who had died 24 hours previously from semi-confluent form of disease. Material removed in test-tube and placed in ice-chest.

*March 5.*—After removal of some shreds of epithelium for histological purposes, the remainder (0.25 gramme) ground up with twice its weight of 50 per cent. solution of glycerine, and the greater portion stored in amber-coloured capillary tubes. The test-tube was afterwards swabbed out, and cover-glass specimens made for microscopical examination.

With some of the glycerinated emulsion inoculated, at the Brown Institution, rhesus monkey (young female) on shaved area of back, about 3 inches by 2 inches, previously well washed with warm boric acid solution. Incisions twelve in number made "en échelon."

*March 8.*—Inoculation has evidently "taken," as tips of each incision are distinctly raised, and whole prospect is that of a typical calf vaccination of about same age (72 hours).

*March 12.*—Eruption beautifully perfect; edges of vesicular portion a little irregular, and centre of each line of incision occupied by commencing "crust." No general eruption visible. Two photographs taken.

Removed lower half of vesicular area with sharp spoon (upper portion left in order to watch further development), and glycerinated resulting pulp. Material used for inoculating Monkey No. 2 and also a calf (No. 606).

*March 12.*—Monkey No. 2 inoculated in fourteen incisions with glycerinated pulp from Monkey No. 1. Technique as before.

*March 19.*—Eruption not so perfect as in Monkey No. 1. Vesicles not so defined and plump. Monkey very wild, and has made sites of incisions bleed by dashing from side to side of cage, which may be in some degree the cause. Photograph taken. Vesicular pulp removed and ground up with four times its weight of dilute glycerine. With some of this emulsion *Monkey No. 3* (young male rhesus) inoculated same day in ten linear incisions on shaved area of back.

*March 26.*—All insertions "taken" well. Photographed. Material collected and glycerinated, pulp being diluted about fifteen times, by mistake. Some used



for inoculation in twelve insertions of *Monkey No. 4* (young male rhesus) same day.

*April 2.*—All places have “taken” well, although material used for inoculation had been so diluted. Photographed. Vesicles scraped and pulp glycerinated.

#### *Calf Experiments.*

*March 12.*—Inoculated Calf No. 1 in half dozen long linear incisions by method usually employed in current vaccinations at Government Establishment, with glycerinated pulp from *Monkey No. 1*.

*March 16.*—Lines of incision slightly raised and red.

*March 17.*—Dr. Fremlin found few small vesicles had developed. These he clamped, inserting material obtained thereby on Calf No. 2.

*March 21.*—There were evident signs of “taking” at all points of insertion, the lines of incision being elevated and with a tendency to vesiculation. But appearances not considered sufficiently typical to permit of material removed being utilised for vaccination of children.

At this point, owing to unforeseen circumstances, this particular series of experiments was discontinued.

#### THIRD SERIES.

*February 25, 1901.*—At the Jenner Institute inoculated medium-sized macaque monkey with small-pox emulsion, made by working up scrapings from P.M. cases of the disease (received from Dr. Thomson, of the Belvedere Hospital, Glasgow), in a small amount of pure glycerine. Technique as in previous experiments. Incisions made on the monkey's back with blunt scalpel, which had lost its temper by constant passing through the flame, so that all incisions did not apparently penetrate to the true skin.

*March 4.*—“Taken” well, though not throughout all insertions as in first monkey of the last series, but as failure had only occurred where there was no mark of incision, it was probably for reason mentioned above, as the eruption which had appeared was good. Not markedly vesicular; lines of incision which were covered with slight crust being surrounded by a pinkish papular eruption. Photograph taken.

Scraped with aid of compression forceps, and rubbed up material in small amount of NaCl 0·7 per cent. solution.

*Monkey No. 2* inoculated this day, with emulsion of scrapings from *Monkey No. 1*. Technique as before.

*March 16.*—All insertions “taken”; slightly more vesicular than in *No. 1*. Photographed. Compression forceps applied and scrapings removed and rubbed up in small mortar with NaCl solution.

*Monkey No. 3* inoculated immediately, on the back, with emulsion of material obtained from *No. 2*. Animal very young; hair downy and not easily shaved.

*March 18.*—All insertions have “taken” and have wide whitish vesicular margin, but appearance rather spoiled as hair on back has grown so rapidly. Lymph oozed up when compression forceps applied. Photographed. Scrapings rubbed up with NaCl solution, of which small quantity was used immediately for inoculation of *Monkey No. 4*. Remainder glycerinated, tubed and stored in ice-chest for future trial on calf.

*March 23.*—*Monkey No. 4*, an old animal, had not apparently taken as well as *No. 3*, so series was discontinued.

*April 20.*—*Monkey No. 3* vaccinated in six incisions on outside of thigh with current vaccine lymph of known potency.

*April 27.*—No result, although monkeys not previously protected take vaccinia as successfully as in the human subject.\*

### *Calf Experiments.*

*March 22.*—Calf No. 1 (1332), at the Government Animal Vaccine Establishment, inoculated with small quantity of glycerinated emulsion of vesicular pulp from Monkey No. 3.

*March 27.*—Tiny papules and vesicles which had made their appearance along lines of incisions removed with Volkmann's spoon and glycerinated.

*March 29.*—Material collected on March 27 inserted into three long incisions on Calf No. 2 (1342).

*April 3.*—Fifth day. Lines of all three incisions occupied by good vesicles, Photograph taken by Dr. Green. Vesicle pulp (0·37 gramme) removed and glycerinated.

*April 24.*—Calf No. 3 (1390), inoculated (with portion of material collected on April 3) in thirty-six incisions on abdomen and scrotum.

*April 29.*—All insertions had "taken" well, vesicles surrounded with slight pink areola. Vesicles scraped and pulp glycerinated. Emulsion stored in ice-chest.

*October 3.*—Four c.c. of this emulsion used for vaccination of calf at the Jenner Institute, by numerous linear incisions extending nearly whole length of abdomen, in manner usually employed at Government stations.

*October 8.*—Appearance indistinguishable from normal vaccination. 63 grammes of vesicle pulp collected and glycerinated.

The glycerinated emulsion prepared from material removed from the calf on October 8 was subsequently employed for the vaccination of other calves, a strain of lymph being thus obtained which continued to give excellent results both on children and calves. But the strain was never brought into general use, and all the glycerinated emulsion remaining was eventually destroyed.

### FOURTH SERIES.

*April 29, 1901.*—At the Jenner Institute young rhesus monkey shaved on back, as in previous experiments, and inoculated in a dozen linear incisions with glycerinated emulsion of S.P. vesicle pulp received from Dr. Thomson, of the Belvedere Hospital, Glasgow, on March 26, 1901.

*April 2 (120 hours).*—Had "taken" so well that I decided to collect; lines of incision distinctly vesicular. After taking photo, washed inoculated area, and removed pulp with aid of clamp forceps. Thin lines of altered epithelium came off as in a good calf vaccination. No "crusting."

Monkey No. 2 inoculated same day with material obtained as above and subsequently triturated in small glass mortar with small quantity of equal parts of glycerine and normal saline solution.

*May 9 (120 hours).*—Every insertion "taken" successfully. More markedly vesicular in places than Monkey No. 1. Photograph taken, followed by usual process of collection and glycerination of vesicle pulp.

Monkey No. 3 inoculated on back in eight diagonal incisions. Emulsion remaining over taken up into capillary tubes, of which two given to Dr. Fremlin for trial on calf at A.V.E. on May 15.

*May 13 (120 hours).*—Eruption of perfectly vesicular character along course of all incisions made, the centre in each instance being occupied by thin linear crust.

---

\* Copeman, 'Journal of Pathology and Bacteriology,' May, 1894.

Drs. Blaxall and Fremlin, on seeing the animal, both described eruption as being equal to that which in case of calf vaccination would be entered in official records as v.g. (very good). Monkey photographed and vesicle pulp collected and glycerinated. Portion of emulsion used same day for inoculation of Monkey No. 4. Another portion handed over to Dr. Fremlin for trial on calf at A.V.E.

May 18.—Monkey No. 4 “taken” well. Vesicle pulp collected, emulsified, tubed and stored in ice-chest.

Series not continued beyond this stage, as laboratory man had failed in attempts to obtain further supply of young rhesus monkeys.

#### *Calf Experiments.*

May 8.—Emulsion of vesicle pulp from *Monkey No. 1*, used at A.V.E. for inoculation of Calf No. 1418.

May 13.—Dr. Blaxall noted “No vesiculation, slight thickening in one line.”

May 15.—Material from Monkeys No. 2 and No. 3, inoculated on calves at A.V.E. by Dr. Fremlin.

May 20.—Both calves had “taken” to a certain extent, the result being most marked in calf inoculated from Monkey No. 3. Material collected, glycerinated, and stored. Portion subsequently handed to Medical Director of Jenner Institute for further trial.

May 26.—Inoculated calf at A.V.E. with emulsion of second removal from Monkey No. 3.

May 31.—Perfect vesicular eruption along course of all incisions. General effect indistinguishable from that obtained with the current vaccine lymph of the Government Establishment.

In view of successful results following on vaccination of children with former lymph stocks raised in similar fashion, it appeared unnecessary to employ this particular lymph for infantile vaccinations. But on vaccinating my own arm with it direct from the calf, I succeeded in raising by the eighth day a fairly typical vesicle, an effect in excess of that obtained by me on my own person at previous attempts at vaccination.

No further transference of this lymph was attempted.

My first series of experiments had not long been concluded, when I came across a reference to an account of similar work which had been carried out by Dr. Eilerts de Haan. The reference occurred in a paper by Dr. Bruno Galli-Valerio,\* and on hunting up Dr. de Haan’s original paper, which is entitled, “Vaccine et Rétrovaccine à Batavia,”† I found that it contained an account of a lengthy series of experiments on the variolation of monkeys and on the transference of the resulting affection to calves.

Dr. de Haan’s work proved of special interest to me for the reason that, quite independently, we had been able to corroborate one another’s work, except as regards the transference of the strain of variola vaccine to the human subject—a final test which Dr. Eilerts de Haan did not, as he says, feel justified in attempting, in view of the unfortunate experience of Chauveau, in connection with his abortive

\* ‘Centralblatt für Bakteriologie,’ March 28, 1893, p. 380 *et seq.*

† ‘Annales de l’Institut Pasteur,’ 1896, p. 169.

attempts at variolation of the cow. Dr. de Haan's own words may be quoted:—"Je reconnais qu'il manque à ma démonstration d'avoir rapporté la variole mitigée du singe sur l'homme ; c'est une expérience que je ne me suis pas cru en droit de faire. L'expérience de Chauveau enseigne à être prudent, et je ne me croirais autorisé à faire cette tentative que si le vaccin ordinaire dont je me sers me manquait au moment d'une épidémie. Mais j'espère qu'on répétera mes expériences à ce sujet."

For the purpose of his experiments Dr. de Haan made use of the macaque monkey (*Macacus cynomolgus*), which is common in the Dutch East Indies, and therefore was readily obtainable. His inoculations were, in each instance, made on a portion of the animal's back, which was first shaved and then cleansed with soap and water, followed by a solution of boric and salicylic acids. In his first series of variolations of the monkey, the small-pox lymph employed was obtained from a Javanese child, no statement however being made by him as to the age of the child, whether or not it had ever been vaccinated, or at what stage of the disease the lymph was taken. Transferred to the monkey, this lymph gave rise, in a week's time, to well-marked vesicles at the site of inoculation, while, in addition, a few papules were observed on the lips and the extremities. Seven subsequent primary variolations were, however, successfully carried out on monkeys, in only one of which was any evidence of generalization observed.

From the contents of the vesicles of the monkey first-mentioned, a second was inoculated, which in seven days developed vesicles at the inoculated points only. From this second monkey, in due course, a third animal was inoculated ; from this a fourth, and so on, through a series of seven monkeys.

From the sixth monkey of this series, a calf was inoculated, which five days later presented an appearance indistinguishable from a typical vaccination. From the seventh monkey also, of the series, a calf was inoculated, with the result again, that after an interval of five days, perfect vaccine vesicles appeared at the site of each insertion of the lymph. From this calf another was vaccinated with complete success.

In a second series of experiments, lymph at the fourth remove, in the monkey, from human small-pox, gave rise to perfect vesicles when inserted on the skin of a calf, and the strain of vaccine lymph thus obtained was carried on successfully, through eight removes from calf to calf. Monkeys and calves all failed to react to subsequent inoculation with the strain of vaccine lymph in current use.

The results of my own experiments may be briefly summarised as follows :—

In each of the separate series of experiments the human small-pox lymph or pulp was first inoculated directly on calves, and in every instance, so far as could be observed, with altogether negative results. But with monkeys success was as invariably obtained, and when, after one or more passages through this animal, the contents of the local inoculation vesicles were employed for insertion on the calf, an effect was now produced which, after two or three removes in that animal, was indistinguishable from typical vaccinia.

Moreover, from the contents of vesicles raised in this manner on the calf, a number of children have, in turn, been vaccinated, some of whom were afterwards kept under observation for as long a period as a couple of months.

Every such vaccination "took" normally, and in no case was any bad result subsequently observed by myself, or reported by the parents of the children; no "generalisation" of the eruption occurring in any instance.

In conclusion, I desire to call attention to the somewhat remarkable fact that a mild and strictly localised form of small-pox, such as is induced in the monkey by the inoculation of material from cases of the generalised disease in man, should, when transferred to the calf, "take" readily with the production of a vesicular eruption of non-infectious character in that animal, whereas it is well known that successful transference of small-pox direct from man to calf can only be accomplished with the utmost difficulty.

The experimental results obtained in the course of the research, an account of which has been set out in this paper, all tend, then, to confirm the view that the vaccinia of Jenner's time was derived, in all probability, from a comparatively mild form of human small-pox.

In addition, I think it will be admitted that the work has afforded conclusive evidence of the essential identity of the virus of small-pox and cow-pox or vaccinia.

---