Report on an inquiry into the nature, causes, and prevention of splenic fever, quarter-evil, and allied diseases: made at the Brown Institution / by W.S. Greenfield.

Contributors

Greenfield, W. S. 1846-1919. Paget, James, Sir, 1814-1899 Royal College of Surgeons of England

Publication/Creation

London: Printed by William Clowes and Sons, 1880.

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INTO THE

NATURE, CAUSES, AND PREVENTION

CF

SPLENIC FEVER, QUARTER-EVIL,

AND ALLIED DISEASES,

MADE AT THE

BROWN INSTITUTION.

BY

W. S. GREENFIELD, M.D., F.R.C.P.,

PROFESSOR-SUPERINTENDENT OF THE BROWN INSTITUTION.

PRINTED BY WILLIAM CLOWES AND SONS, LIMITED, STAMFORD STREET AND CHARING CROSS.

1880.

FROM THE

JOURNAL OF THE ROYAL AGRICULTURAL SOCIETY OF ENGLAND.

VOL. XVI.—S. S. PART 1.

REPORT ON AN INQUIRY

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BROWN INSTITUTION.

The investigations which form the subject of this Report have been carried on during the past year in continuation of those made in the previous year by Professor Burdon Sanderson and Mr. Duguid. I have, however, extended the scope of the inquiry, and have taken up, in addition to splenic fever, the disease of cattle known as quarter-evil, and the diseases of horses known as Cape horse-sickness and Loodiana fever. Both in the experimental and clinical work I have been most ably assisted by Mr. George Banham, M.R.C.V.S., Veterinary Surgeon to the Brown Institution.

It is necessary to state that the investigation is still in progress, many subjects awaiting further opportunities of obtaining material for examination and experiment; and a doubtful opinion must therefore be expressed on many points, which we hope will be cleared up by future observation.

ANTHRAX AND ANTHRACOID DISEASES.

In addition to the disease of cattle most commonly known to English agriculturists and stockowners as splenic apoplexy or

15

anthrax, there are a considerable number of forms of disease which affect either cattle, sheep, or horses, and more rarely other animals, both in this country and in other parts of the world, to which the common term "anthracoid," allied to or resembling anthrax, is applied. Widely diffused as are these diseases, and enormous as is the mortality caused by their attack, we know as yet but little either of their causes or their relation to each other. It was felt that in an inquiry having for its chief object the determination of the causes and prevention of splenic fever, it would be very important to ascertain, as far as possible, the nature of these other forms of anthrax, and the conditions under which they occur, as they might very probably give some clue to the causes of splenic fever.

In England the disease known as Quarter-Ill, Quarter-Evil, Black Quarter, and under many other names, is the one concerning the nature and causes of which the greatest doubt exists.

In France and Germany, and on the Continent generally, the various forms thus separated in England are usually classed together; in France, going under the name of *Charbon*, *Mal de rate*, or *Sang de rate*; in Germany, as *Milzbrand*, *Karbunkel-krankheit* and *Anthrax*. The different forms are sometimes more precisely defined, as I shall show later.

In Russia, the "Siberian Plague" is the name under which the common disease is known, the name having probably arisen from the supposed resemblance of the disease when communicated to man to the true or Levantine Plague. This Siberian plague is very widely spread, reaching throughout Siberia, occurring at scattered spots in Russia, and southwards as far as the Caspian.

In India there are two or three diseases which appear to be closely allied to splenic fever. In the Punjab there is the disease of horses known as Loodiana fever. Elsewhere a similar disease is highly fatal to elephants and camels. The Hara Murree, or Pali Plague, and the Bharsati disease are two other perhaps allied epidemic diseases, concerning the nature of which but little is known.

In South Africa attention has recently been directed to the very fatal disease of horses, known amongst the Boers as throat-sickness, Dikkip, or black tongue. Although presenting many analogies with some of the localised forms of anthrax, it appears to be distinct, inasmuch as true anthrax when affecting horses in the same or other regions is said to present different symptoms. And, as I hope to show, although there are very close analogies in the fungoid organism which is apparently the cause of the disease, there are also certain well-marked distinctions. In

the present Report I propose to limit myself almost exclusively to the subject of the experimental investigations which have been made during the past year at the Brown Institution, referring to other points only so far as may be necessary for the purpose of explanation. In a future Report I hope to deal with the more recent observations on the nature and mode of propagation of anthrax, and the more minute description of the morbid anatomy of the several diseases which have formed the

subject of investigation.

Amongst the many points relating to anthrax which have received attention, the most important is that of the experimental inoculation of bovine animals with virus modified by transmission through the guinea-pig. Some preliminary experiments made by Mr. Duguid at the request of my predecessor Dr. Burdon Sanderson appeared to show, first, that when anthrax is communicated by inoculation to bovine animals through rodents, the animals so infected, although exhibiting severe symptoms, recover; and, secondly, that such animals are less liable to future infection by the same process than others. These experiments I have repeated, varying the method employed; and although in some respects the results are such as to modify the conclusions suggested by the experiments of Dr. Sanderson, they are nevertheless in striking confirmation of the general result, and afford grounds for hope that the continuance of the experiments may serve to establish a prophylactic for the disease. Hitherto I have had no opportunity of submitting an animal thus inoculated to the crucial test of subsequent exposure to contagion or inoculation from an original case of the disease in a bovine animal; but I have done so from a sheep which died of the artificial disease, with a favourable result.

I have also been engaged in studying the microscopic anatomy of the disease in various animals, with a view to discover the mode of diffusion of the poison, and its method of action in the system, and as some of these investigations afford practical suggestions, I propose to refer to them briefly in the present Report.

Inoculation of Bovine Animals with the Poison of Anthrax transmitted through Guinea-pigs.

It is now a generally recognised fact that the contagium or virus of splenic apoplexy (anthrax) is a low vegetable organism, the so-called Bacillus anthracis. This organism is found in

enormous quantities in the blood and certain of the tissues of animals dying of the disease. It has been shown by repeated experiment that it may be artificially cultivated outside the living body, under conditions suitable to its growth and propagation; that it nevertheless retains its virulent property, which may be transmitted from generation to generation, so that a far distant progeny of the bacillus first grown from the blood of the living animal, if introduced into the system of another living animal, gives rise to all the phenomena of the disease.

Moreover, the disease is readily communicable to a large number of known animals, rodents being especially susceptible, guinea-pigs and mice being amongst the most easily inoculated.

In transmitting the disease through rodents to bovine animals, two distinct methods may be employed; the one of direct inoculation, the other of inoculation with the cultivated virus.

The direct method, that employed by Mr. Duguid in his experiments, consists in inoculating the bovine animal with the blood or spleen of a guinea-pig which has died of the disease produced either by inoculating from a cow, or from one of a series of which the first was inoculated from a cow. That is, if guinea-pig A is inoculated from a cow, B from A, C from B, D from C, and then a cow direct from D, it would be an example of direct inoculation. Or it might be inoculated direct from A or B or C. Dried blood, if properly secured, retains in parts its infective power for a considerable time, so that this process of transmission may have been carried over some months.

The "cultivation" or indirect method proceeds on the basis of the fact already indicated, that the poison (the Bacillus anthracis) may be artificially grown in a cultivating fluid in successive generations, and that the last generation may then be inoculated and produce the symptoms and fatal result of the disease.

Seeing that the object of my experiments was to inoculate bovine animals with a virus modified by its transmission through the guinea-pig, it may appear that it would be desirable, in order to have the full effect of such modification, either to inoculate directly from the guinea-pig or from a cultivation not far removed from it. But there would be many practical advantages, if it were found that, having once transmitted the virus through one guinea-pig or a series of them, its modified property were subsequently maintained in the artificial cultivations. I have therefore kept this in view, and have

tested the effect of successive generations of the artificially pre-

pared poison.

It would have been a great advantage if I had been able to test, side by side with these cultivations derived from the guinea-pig, other cultivations derived direct from a bovine animal. But in every case cultivations from the latter have failed, owing to the commencement of decomposition in the material sent to us.

The method of artificial or fractional cultivation is now well-known. The bacillus is capable of growth in many nutrient fluids, amongst others aqueous humour, hay-infusion, serum, urine, and also in many other natural and artificial fluids. The necessary conditions suitable for its cultivation are supply of nourishment and of air, a proper temperature, and the exclusion of sources of putrefaction in the form of germs of other bacteria.

Of the various nutrient fluids, I have employed almost exclusively either aqueous humour or hay-infusion. The former may be readily procured from the eye of a recently killed animal, such as a sheep or ox.

Of the two methods of cultivation most commonly employed, cultivation in cells and in hermetically sealed tubes, I have employed chiefly the latter for my present purpose, as affording a better supply, which can be more easily preserved, and more

readily used for inoculation.

The method of cultivation is very simple. A piece of glasstubing about 4-inch in diameter is drawn out at both ends to a fine capillary tube, and sealed at the ends. The central chamber thus formed, about an inch in length, is half-filled with the cultivating fluid, after opening the tube; the lower end is then re-sealed; a capillary tube, containing a minute quantity of the blood, or only rubbed on the spleen of the animal which has died of anthrax, is passed down into the chamber till it touches the cultivating fluid; it is then rapidly withdrawn and the tube sealed up. The cultivating tube is then placed in an incubator, which is kept at a uniform temperature of about 35° C., and left there for twenty-four to forty-eight hours, at the end of which time an opalescent mass consisting of the growing bacillus has been formed. From this a minute quantity is removed with a capillary tube, and a fresh tube of cultivating fluid is inoculated and placed in the incubator. And a similar procedure may be repeated for an almost indefinite period, if proper precautions be taken. The bacillus thus grown may be preserved in capillary tubes in certain stages of its existence for

a very long time. I may now describe what these stages are, for modern research has shown that this organism passes through several distinct phases of existence, in all, however, preserving its power of germination under suitable conditions,

as well as its infective property.

As seen in the blood and tissues of animals dying of anthrax, the Bacillus anthracis is usually a short protoplasmic rod, measuring from one to four times the diameter of a human redblood-corpuscle in length. These rods, in many cases, swarm in immense numbers in the blood, so that they may appear even to exceed the blood-corpuscles themselves in number, and by their presence they interfere with the normal coalescence of the blood-corpuscles and prevent normal coagulation of the blood. But it is not in every case of anthrax that the Bacilli present this appearance or are found in such numbers. Not uncommonly they may be found in much smaller quantity; and in some cases examined only four or five hours after death, the rods may be very much longer than those just described. This may possibly be due to growth after death of the animal. On minute inspection, the longer rods are usually found to be in process of division into two or more short rods of nearly uniform length.

2

Fig. 1.—Bacillus anthracis.

Rods of Bacillus anthracis as seen in the blood.
 Portions of rods under cultivation.
 Groups of spores.
 and 2 magnified about 500 diameters.
 about 700 diameters.

The rods, which appear homogeneous and uniform, and blunt at the ends, increase in number by a process of elongation and transverse fission in the ordinary process of the disease within the body. If one of the rods which is in course of division be watched, it will be seen that having reached a certain length, a clear space appears in the middle between the two highly refractile segments. This clear space does not as yet indicate a complete separation, for a delicate outline can be seen uniting the two

rods, which later become entirely separate. This outline is the delicate sheath which surrounds the rods, but is not readily

seen during the simple rod stage.

When the organism is cultivated in the manner already described, changes occur which usually follow a certain course. Similar changes may also take place in the carcass of the dead

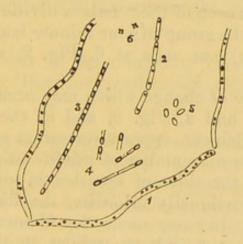
animal, especially if the temperature is favourable.

The short rod elongates rapidly into a long filament, composed of a homogeneous protoplasm enclosed in a very delicate hyaline sheath, and this filament may grow to a great length, curving in all directions and forming loops or spirals. These long filaments are often united in parallel bundles, or cross

obliquely in a very characteristic manner.

As this elongation proceeds, changes take place within the substance of the rod which may assume one of two forms. The more common is as follows: The central protoplasmic mass of the rod becomes somewhat granular in appearance, and the protoplasm appears broken at certain points, clear specks, as seen in the lower straight rod of 2, Fig. 1, sometimes appearing at regular intervals. Then the protoplasm shrinks, leaving clear spaces, with ovoid or oblong highly refractile bodies arranged at regular intervals, as at 3, Fig. 2, or more irregularly or in pairs, as at 4, Fig. 2, and 7, Fig. 3. These oblong or ovoid bodies are the now well-known spores.

Fig. 2.—Stages of Bacillus anthracis under Cultivation.

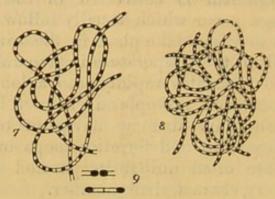


Wavy filament, showing commencing lateral formation of spores, as granular dots.
 Another part of same cultivation, filament dividing, each segment having a terminal dot.
 Filament in which spores have formed at nearly regular intervals.
 Portions of filament with spores after division.
 Isolated spores.
 Sporules formed by division of spores.

One of two things may happen: the spores may remain in situ in the remains of the filament, which persists only as an almost invisible shred of the external hyaline capsule, or they may escape from the filament by a lateral opening. Or, lastly,

the filaments may break up into a number of fragments, each containing one or two spores, as at 4, Fig. 2.

Fig. 3.—Further Stages of Bacillus anthracis under Cultivation.



 Part of convoluted filaments in which spores have formed, and division is commencing in parts, from cultivation of Bacillus anthracis.
 Similar process in another Bacillus not connected with anthrax.
 Portions of filament from 7 more highly magnified.

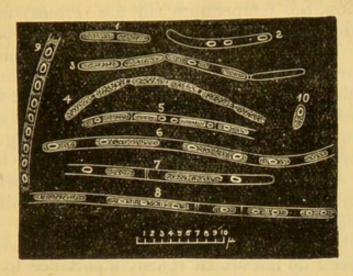
The spores thus formed are capable of long dormancy, for months or years, and may then, given favourable conditions, grow again into rods and reproduce the disease. It is the formation of these spores, and their capacity for resisting changes of temperature and other adverse conditions, which makes the contagion of anthrax so persistent and so difficult to destroy.

In some cases these spores, when kept under conditions which are only favourable to very slow germination, undergo a process of division by transverse fission, each spore first dividing across its length, and then each of these halves divide again transversely, so that each formed a group of four minute bodies, or sporules (as Ewart calls them); as seen at 6, Fig. 2, and still better at 3, Fig. 5.

The other course of change which may occur in the filaments is that seen at 1 and 2, Fig. 2, and at the bent filament in 2, Fig. 1. The filament appears somewhat swollen:—in it (as in 1, Fig. 1) are seen a number of minute bright dots, which as a rule are towards the sides of the sheath, sometimes only here and there, but occasionally crowding the filament with bright oily-looking dots. In some cases the filaments break up into shorter pieces, as at 2, Fig. 2, whilst in this condition. It is probable that this may be regarded as merely a stage in spore formation, which here commences laterally instead of centrally.

The more minute changes which take place in the course of growth and spore formation, will be better seen in Fig. 4, which is taken from cultivations of *Bacillus anthracis*, magnified 1500 diameters.

Fig. 4.—Bacillus anthracis magnified 1500 diameters.



 A short rod (8 μ long) in which the protoplasmic contents have divided previous to fission of the rod. 2. A clear rod containing only spores. 3. A jointed filament composed of three segments, in one of which fission is commencing, in another a spore has formed, whilst another is empty. 4. Part of filament composed of short homogeneous rods united by a narrower sheath. 5, 6, 7. Parts of long filaments showing various arrangements of the protoplasm and spores. 8. From another filament in which spore formation is proceeding more regularly. Commencing fission is seen at some parts of the filament. 9. Part of filament completely filled with regularly arranged spores. 10. From another cultivation showing sporule germinating into a short rod.

(It must be stated that the space between the sheath and the protoplasmic contents did not exist in the original drawing, and that in the other rods it has been necessarily somewhat exaggerated in the woodcut.)

I have thought it well thus briefly to describe the changes usually observed, in order to render clear the frequent references

to these various conditions in describing the experiments.

Details of Experiments on Inoculation of Bovine Animals with Anthrax poison (Bacillus anthracis) transmitted through Rodents.

Case I .- A steer, in good health and condition, was kept under observation for three weeks, in order to ascertain its normal temperature. It was then inoculated with a small quantity of fluid which had been obtained by cultivation of the Bacillus anthracis in aqueous humour, and which swarmed with rods and spores of bacillus. The fluid thus used was the second generation of the cultivation of spleen of a guinea-pig which died of anthrax. This guinea-pig was the third of a series, through which the disease was transmitted from the cow by inoculation. I have already described the method followed in these experiments.

For about twenty-four hours no symptoms were observed, there was only very slight swelling at the seat of inoculation.

On the morning of the second day, *i.e.*, about forty hours after the inoculation, the temperature (in the rectum) was found to be 104° Fahr., the normal temperature having been 100° to 101° Fahr. The animal was drowsy and stupid and fed badly, but no rigour was observed, nor any special symptoms. On the evening of the same day, the temperature was 102.4° Fahr., but on the following day it rose to 106.4° Fahr., and continued to vary between 105° and 107° for three days, during which time the animal was seriously ill, and at times seemed hardly likely to recover. On the eighth day, however, the temperature fell to 104°, and on the ninth to 101°, after which the animal speedily recovered and continued perfectly well.

I was unable to discover any *Bacilli* in a drop of the blood examined, but I do not attach much importance to this fact as any proof that they were not present, for I have often failed to find them in the blood of the general circulation in smaller animals at a period before death, when they must have existed

in the blood in some organs.

The animal, having completely recovered, was again inoculated a second time some weeks from the first inoculation. During the interval after subsidence of the fever produced by the first inoculation, the temperature was at no time higher than 101.4°,

ranging between 100° and 101°.

This second inoculation was made directly with the fresh spleen of a guinea-pig which had died of anthrax, simply rubbed down with saline solution to make it more fluid. One gramme of this fluid was injected with a hypodermic syringe into the right side about the elbow. The fluid swarmed with rods.

In the evening the temperature was 102°. Next day the temperature was 102·4° morning, and 102·6° evening, and the animal showed some indisposition and took food badly. On the third day there was complete recovery of appetite, and the temperature became normal. The subsequent temperatures will be seen by reference to the appended table. (See page 14.)

The third inoculation was made a week after the second. The animal had completely recovered in every respect, and it was desirable to test the power of resistance acquired by the previous inoculations, and to compare the result with a simultaneous inoculation of another animal with the same

material.

In this third inoculation the poison was obtained from a guineapig which had died of anthrax derived from the horse. The spleen having been removed shortly after death, was rubbed down with saline solution, and one-half of the fluid injected into the steer, the other half into the cow (Expt. No. 2). No result whatever followed in the case of the steer, with the exception that on the evening of the following day the temperature was 102°. But the appetite was unaffected; the slight local swelling

produced by the inoculation speedily subsided.

Fourth inoculation.—A period of three weeks having elapsed after the third inoculation, during which the animal was in good health, a fourth inoculation was made. A guinea-pig, inoculated with blood from the guinea-pig used for a previous experiment, which had been dried and kept in a sealed tube for some weeks, died of anthrax. The spleen was considerably enlarged (an unusual occurrence in the guinea-pig), and very soft, and the fluid exuding from the cut surface swarmed with Bacilli. The whole spleen was rubbed down with about a drachm of water, and nearly the whole of the fluid with the particles of the spleen in suspension was injected by means of a hypodermic syringe beneath the skin of the shoulder. result of this inoculation, so far as general symptoms are concerned, was absolutely nil, not the slightest rise of temperature or failure of appetite being produced, and the animal subsequently remaining in perfect health.

Fifth inoculation.—Fourteen days having elapsed since the previous inoculation, during which the temperature of the animal had continued absolutely normal, on no occasion exceeding 101° Fahr., another inoculation was made, the material on this occasion being derived from a sheep, which died of anthrax (see Case IV.). The bloody fluid which flowed from the cut surfaces of the lung was collected, and about one drachm injected beneath the skin of the right shoulder. The fluid injected contained a large number of very long bacillus rods, and caused the usual symptoms of anthrax in a guinea-pig inoculated at the same

time.

On the day following the inoculation, the part was considerably swollen, the animal was rather dull, and did not take food so well as usual, but there were no other symptoms. There was some rise of temperature—103.4° in the morning, 103.6° mid-day, and 104° evening. Next day there was still some slight loss of appetite, but the temperature was normal, and subsequently the animal continued in perfect health. A slight swelling remained at the seat of inoculation, which subsequently changed into a small abscess. This was opened, and discharged a small quantity of ichorous pus.

This animal is still being kept, awaiting the opportunity of making the crucial experiment of direct inoculation from a case

of anthrax in the cow.

CASE I.

200	Burner	and the	EXPERIMEN	T I.	
Day of Observa-	Rec	tal Temperate	ire.	Remarks.	
tion.	Morning.	Noon.	Evening.		
THE PARTY	0	7-10-11-1	- 100	The state of the s	
1	100.6	0	101·	THE RESERVE OF THE PARTY OF THE	
2	101.		101	The second second second	
3	101 ·	In		First inoculation with cultivated	
4	101.4		101.8	fluid.	
5	104		102.4		
6	104.4		106.4	The second secon	
6 7 8	105		106.8	Charles of the Control of the Control	
8	105.4		106.6	STATE OF THE PARTY OF	
. 9	106.		105.6	usin dmi succession nome	
10	104		104		
11	100.		101.		
12	100.4		101•		
Treat p	of partition is	Exp	ERIMENTS II	, and III.	
1	100.		101-	Total and the stance	
2	100.8	o thirty	101.4	DESCRIPTION OF THE PARTY OF THE	
3	100.4	3- 0-	100.8	contract conflicted and the second	
. 4	100.4		102	Second inoculation.	
5	102.4		102.6		
6	100.4		100.	The second is not as the late of the late	
7	101.		101.4	CHIEF DESIGNATION OF THE PARTY	
8 9	100·6 100·2		707.	Singly leader and leavest special	
10	100.4		101.		
11	101.	••	101.2		
12	100.8		100.4		
13	100.4		101.	Third inoculation.	
14	101.2	101.	101.		
15	101 -	101.	101.		
16	101.	101.	101.		
17	101 ·	101.	102.		
			Experiment	r IV.	
7	100.4		101-	A STATE OF THE STA	
1 2	101.		101	The state of the s	
2 3	101.2		101.6	PROPERTY OF THE PARTY OF THE PA	
4	101.		100.6	The state of the s	
5	101.		101.4	Fourth inoculation, with spleen of	
	101.	101.2	101.4	guinea-pig.	
6	101.2		101.4	The state of the s	
7 8	100.4		101		
9	100.		100.4		
	200			A STATE OF PERSONS ASSESSED.	

EXPERIMENT V.

Day of Observa- tion,	Rectal Temperature.			REMARKS.
	Morning.	Noon.	Evening.	
1 2 3 4 5 6 7 8	101· 101· 103·4 101· 101·6 101·2 100·4 100·4	103·4 100·8	101· 101· 104· 101·4 101·6 101·6	Fifth inoculation, direct from sheep

Case II.—The material used for these experiments was derived from a case of anthrax in a horse, the poison having been transmitted successively through two guinea-pigs. The preparation of the material has already been described (part having been used for Inoculation 3 in Case I.).

The other part of the fluid was injected subcutaneously in an old emaciated cow, which, as was discovered subsequently, was in-calf.

On the day of inoculation no symptoms occurred. On the following day the temperature rose to 102.4° morning, $103.^{\circ}$ evening. On the third day the morning temperature was 103.6° , evening 102.4° , but there was no loss of appetite, nor were any other symptoms noticed. On the morning of the fourth day the animal was found dead.

CASE II.

	Day of Observa-	Temperature in Rectum.			
	tion.	Morning.	Noon.	Evening	
	1	101.4	101·4 (Inoculation).	101.6	
DIE GUILLE	2 3	102·4 103·6	102.	103· 102·4	
	4	Found dead.	102	102 4	

Post-mortem (6 to 9 hours after death). Body emaciated. A bloody mucous discharge flowed from nostrils and anus.

Stomach full and healthy. Intestines congested, with bloody extravasation here and there. Spleen somewhat enlarged, weighing 2 lbs. 14 oz., presents hæmorrhagic spots in the capsule. On section, tissue highly vascular, somewhat soft but not diffluent. Liver and kidneys apparently healthy.

Heart: numerous small ecchymoses and petechiæ beneath the endocardium, especially that of the left ventricle, and also in the musculi papillares. Some also more deeply in the muscular tissue, and one or two beneath the pericardium.

Lungs appeared healthy and pale.

On microscopical examination, the blood was found to contain very large numbers of rods of *Bacillus anthracis*, and the tissue both of the spleen and the other organs also swarmed with them. The sanious discharge from the nostrils also contained them in great abundance.

The feetal calf, of about four months, appeared healthy. All its organs were carefully examined for *Bacilli*, but none were

discovered.

It will be observed that this experiment differed from the first case in being inoculated direct from the guinea-pig instead of with cultivated fluid, in the original source of the material, viz. the horse, and in the fact that the animal was old and emaciated and in-calf. The latter points appear to me to be of more importance than the former, as the animal in Expt. I. was inoculated in the same way and with the same material

without any symptom being induced.

Case III.—A calf 6 months old was inoculated with the fourth cultivation of anthrax Bacillus, derived from the guinea-pig. No symptoms whatever were produced. The temperature had been somewhat irregular previous to the inoculation, owing to an attack of diarrhœa. On the ninth day from the first inoculation, the animal appearing perfectly well, it was again inoculated with the first cultivation direct from the spleen, which had been kept sealed up for a fortnight, and swarmed with spores of Bacillus anthracis. To test the virulence of the material, a mouse was inoculated at the same time with a very minute quantity, and was found dead the next day, about 20 hours from the time of inoculation. The spleen of the mouse was found to be crowded with rods of the usual character.

On the day following inoculation the part was somewhat swollen, but there was no perceptible rise of temperature until the second day, when it rose to 106° morning, and 106·4° evening. For the ensuing four days the elevation of temperature persisted, with variations from 105·4° to 106·4°, there was continued swelling of the shoulder, which extended, and, together with this, loss of appetite. On the seventh day, the morning temperature was 103·8°, but as the swelling persisted and showed no sign of subsiding, an incision was made into it, and fomentations applied. The subcutaneous connective tissue was found to be infiltrated with colourless, somewhat gela-

tinous, serous exudation. On microscopic examination no Bacilli or Bacteria were found either in this serous fluid or in the blood.

The animal speedily improved, the swelling subsided, and complete recovery occurred. A sufficient time has not yet elapsed

for further inoculations to be made.

CASE III .- CALF SIX MONTHS OLD.

EXPERIMENT I.

Remarks.		ctum.	Day of		
	Evening.	Morning. Noon. E		Observa- tion.	
n, with 4th genera-	(First inoculation	103.	0	0	1
ated material.	tion of cultiva				
		103.6		104.	2
		105	104.6	104.	2 3
		104.2		103.6	4
		103.4		104	5
	The state of the s	103.8		103	4 5 6 7 8
		103.8		103.	7
		103		102	8
tion, with first gene-	Second inoculati	103*	£ 14.000	103.	9
	A STATE OF THE STA	103.2		103	10
	No. of the State of the last	106.4	105.6	106	11
		105.8	106.4	105.4	12
	The state of the state of	105.8	105.6	105.4	13
	THE REAL PROPERTY.	105.8	106.2	106.4	14
		106.4	100 2	106.2	15
		104.	103 8	103.8	16
subsiding. Eats	(Local swelling				
, , , , , , , , , , , , , , , , , , , ,	well.	102	103	103.4	17
		102	102.	102.4	18
	Quite well.	101.4	101.2	101.	19

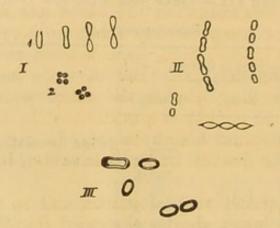
Inoculation of Sheep, with cultivated Bacillus anthracis, in the form of Spores.

Some doubts having naturally arisen as to whether the cultivated material used in the preceding experiments could be regarded as representing the anthrax poison in a virulent form, and an opportunity arising for inoculation of a sheep, I thought it desirable to test the material upon this animal. And as the disease is liable to be communicated to sheep, and in some countries frequently forms a fatal epidemic amongst them, the question whether the poison was modified in its action upon them by transmission through rodents appeared important. The following experiment was therefore performed :-

Case IV.—A young sheep was inoculated with a small quan-

tity of the cultivation of the fourth generation, which had been kept sealed up in the incubator for a week. The characters of the organisms found in the fluid at the time of inoculation are seen in the annexed drawing. There were, in fact, hardly any rods, only spores in various stages of subdivision.

Fig. 5.—Drawings of Elements seen in Fluid with which the Sheep was inoculated.



I. and II. drawn with No. 10 imm. Hartnack, No. 3 oc. 1. Stages of division in spores. 2. Sarcina-like groups of 4 sporules. II. Rows of spores, some in process of division. III. With higher power—spores dividing and divided.

No symptoms whatever were observed, beyond a slight lowering of temperature, as seen in the list of temperatures. (The normal temperature of the sheep averages 103° Fahr.) The animal fed well, and did not appear to suffer in any way, until the evening of the fifth day (96 hours after inoculation), when the temperature was 107°. No special symptoms, however, were observed. Next morning the sheep was found dead.

Examination of the body about twelve hours after death.—Well-marked rigor mortis. No trace of decomposition. No local swelling observed. Skin generally free from exudation or ecchymosis.

Pleuræ healthy. Lungs pretty fully distended. Some small patches of collapse in upper part of right lung. Throughout the whole of both lungs, both on the surface and on section, are very abundant dark red spots, which have the appearance of uniformly disseminated ecchymoses. They are of nearly uniform size, about that of a mustard-seed. There is no appearance of pneumonic consolidation around or between them. Bronchi and trachea contain frothy blood-stained mucus; mucous membrane generally appears somewhat injected.

Pericardium healthy. Heart: ventricles normally contracted. Some small ecchymoses around the base of the left ventricle, in the subserous tissue. Endocardium somewhat deeply bloodstained. Ecchymoses in the muscular wall of the left ventricle, especially in and around the musculi papillares. Valves healthy. Blood in the heart imperfectly coagulated.

Liver small but natural.

Spleen somewhat enlarged and soft; on section, however, not pulpy, though readily broken down.

Kidneys somewhat swollen and soft, but otherwise of natural

appearance.

Stomach and intestines natural.

Brain and cord appear perfectly healthy.

On microscopic examination the blood was found to contain very long rods in comparatively small proportion, i.e., though several were seen in each field, there was no crowding of them together as is often the case. But the rods were of enormous length, many reaching almost the entire width of the field, measuring from .05 to .175 of a millimetre in length, or nearly twenty times the usual length, having, in fact, an appearance often found in the bacilli under cultivation, but rarely in the

In the bronchial and tracheal mucus, and in the bloody fluid exuding from the lungs, similar elongated Bacilli were found.

In the spleen, repeated examination by myself and Mr. Banham, both of the splenic tissue and of the bloody fluid exuding from it, entirely failed to discover any Bacilli. Only a few ordinary micrococci were observed, not a single Bacillus in any of the specimens examined.

Microscopic examination of the lungs after hardening in chromic acid, showed that these scattered spots were actual hæmorrhages. In addition were seen the usual appearances; Bacilli in the capillaries and terminal arteries of the pulmonary

system, and also in the interlobular connective tissue.

Day of Inocula-	Temperatures.		
tion.	Morning.	Evening.	
1	0	104	
2 3	101.6	103	
4	102.6	102·4 103·2	
5	103	107	
6	Found dead.		

I have already described the result of the inoculation of a steer (which had been previously inoculated with cultivated virus from a guinea-pig) with blood from this case. The results produced on the sheep are a striking proof of the virulence of the material after successive cultivations.

To summarise the results of these experiments:-

A yearling steer, inoculated with cultivated Bacillus of the guinea-pig, after forty hours had a rise of temperature with local swelling, followed by high fever, lasting about six days,

this then subsiding and leaving the animal well.

A second inoculation direct from the guinea-pig was practically without effect. A third with similar material, which killed a cow inoculated with it, was likewise without effect. A fourth, with similar but still more active material, was likewise devoid of effect. And lastly, direct inoculation from a sheep was followed by very slight fever, which passed off without any result beyond a small local abscess. This was the only instance in which such an abscess was found.

A cow, old, emaciated, and in-calf, was inoculated direct from the guinea-pig, and rapidly died of anthrax, having had but

little rise of temperature or other symptoms.

A calf, six months old, inoculated with cultivated Bacillus fluid, suffered from intense fever, lasting five days, with considerable local swelling, but the temperature then fell, and the animal recovered. The course of the temperature was closely analogous to that in the first case.

It is scarcely safe at present to attempt to draw any very general conclusions from the experiments which have been performed, but I think enough has been done to show the great importance of the inquiry, and the possibility, if not probability, that by this or some analogous method a means may be discovered of controlling or preventing this widely prevalent and

highly fatal disease.

It must be evident to any one who considers the matter that there are many points which must be determined by experiments of a much more extensive character than any I am able to carry out at the Brown Institution. If, as I hope, it should prove on further experiment that the earlier results are confirmed, and that the inoculation of bovine animals with the Bacillus anthracis cultivated artificially after transmission through guinea-pigs or some other animal serves to render bovine animals totally or partially insusceptible to the disease when transmitted by the usual channels, one great step will have been taken. But there will yet remain the questions: Is the mortality from inoculation by this method a high one, or do even a small percentage of animals die? What are the conditions under which inoculation may be best performed, and does age exercise an important influence in the fatality? And, lastly, for how long a period is protection from attack conferred?

To settle these points, the inoculation of a large number of animals will be necessary, and their subsequent exposure to

sources of contagion at favourable periods.

For the present I propose to continue these experiments in the same direction as those which have already been made, varying the conditions in such a way as to determine, if possible, the readiest method of inoculation, and that affording the best results in conferring protection. If the results hitherto obtained should be confirmed, and if I am able to guarantee definite effects from material cultivated in the manner which I have described, it will be easy to preserve the virus for an almost indefinite period, and to send it to any one who is willing to extend the sphere of observation by making inoculations and watching the results.

OUTBREAKS OF ANTHRAX.

There has been but little information as to outbreaks of the disease (splenic apoplexy); and in no case have I had any opportunity of gaining fresh light upon the etiology of the disease. The facts brought to my notice are embodied in the following reports of Mr. Banham; they concern two outbreaks, from which the material for the experiments was derived.

Outbreak near Romford, Essex.—This occurred in October, amongst a herd of twenty-five cows and two bulls. The cows were turned perfectly healthy into a grass field, the subsoil of which is clay. At the lower end of the field is a stream, which carries the sewage from a village about a mile distant. The cows received no other food, they cannot therefore be said to have been highly fed.

In the first outbreak seven cows died. The two bulls, which had been kept in a shed and never been out at all, also died. The only possible communication between the cows and the bulls were—(1) The cows passed the doors of the place in which the bulls were kept. (2) The men who fed the cows often came into the bulls' stables.

The cows were brought up into the homestead, and then allowed to remain for two months, when the owner thought they might be fairly turned out again. But soon after they had been in the grass-field again, two others died. After that they were brought to the farm again, and remained there without any further loss.

The second outbreak occurred at the end of December, at Alford in Lincolnshire. On Dec. 19, 1879, there were twenty-two cows tied up in a shed, and they appeared to be perfectly well when fed at 3 P.M. It may be mentioned that they were fed on barley, chaff, and straw, tares, hay, mangolds, and linseed-cake. At 3.30 one of the cows seemed stupid, and staggered

about. The attendant was sent for, and found the animal restless, temperature 109° Fahr., pulse could not be taken. The cow died at 5 p.m. The carcass was removed into a yard where the horses were kept at night, and a post-mortem examination was made there. The most characteristic lesions of splenic apoplexy were found. All the parts with which the cow had been in contact were ordered to be thoroughly cleansed and disinfected. Notwithstanding these precautions, on Dec. 23 a horse, which had been worked the previous day, was found dead. On Dec. 25 another was taken ill, and when the attendant arrived he found it unable to rise, and a frothy hæmorrhagic discharge flowing from the nostrils. Blood was also running from the anus. The visible mucous membranes and tongue were highly congested and studded with hæmorrhagic spots. Death occurred fifteen minutes after his arrival.

In connection with this outbreak, I may mention that in the neighbourhood of Alford the disease appears to be endemic,

previous cases having been reported to us.

On a farm in the neighbourhood, the owner lost fifty cattle in February 1878. On June 5, 1879, a yearling steer from the same yard as that in which the previous outbreak had occurred was found dead. A post-mortem examination showed in the abdomen a quantity of black blood; the spleen weighed 8 lbs. and was greatly congested; the gastro-intestinal tract contained bloody fluid throughout, and the mucous membrane showed numerous ecchymoses.

Another case occurred in the same neighbourhood, though at some distance from the above, on June 6, in a heifer grazing in the marsh. No previous case was known to have occurred on this farm for twenty-five years before. The symptoms and post-mortem appearances resembled those in the previous

case.

Distribution of Bacilli.—One of the most striking points in the morbid anatomy of anthrax is the enormous number of Bacilli, and the way in which they crowd the blood and tissues. One might indeed imagine, from the way in which the blood sometimes swarms with them, that they would be found nearly equally in all the organs. This, however, so far as my observations go, is not the case. Nor are they especially abundant in the spleen in many of the cases which I have examined; in some, e.g. the sheep in Case IV., none are found.

In the heart they are often found in very great abundance, crowding the vessels, appearing alongside the vessels in the intermuscular spaces, but not in very great number; and forming here and there masses which completely plug the small vessels and lead to extravasation of blood, which of course also contains Bacilli.

In the lungs they are well seen, owing to arrangement of the capillaries in the alveolar walls; but they more rarely lead to infarction.

They do not usually appear in any great number in the

pleura.

The liver is sometimes very free from them; when found, they are especially seen in the hepatic veins and their intra-

lobular tributaries.

The kidneys usually contain them in very large numbers, chiefly in the glomeruli and their afferent and efferent arterioles, and in the parts of the vessels in immediate continuity with them. Many of the afferent arterioles are almost completely plugged by masses of them; in the glomeruli themselves, especially near the surface of the organ, they lie in parallel bundles, in such a way that the capillaries appear to be striated; and sometimes they form a mass which may occupy a considerable portion of the glomerulus. I have in some of these masses seen the formation of spores: a fact, I believe, of great importance. Rupture of the glomerulus may occur, and the Bacilli, usually accompanied by some blood, escape into the convoluted tubes, where they may grow to a considerable length. Koch states that they do not pass beyond this position, and that he has never seen them in any part of the straight tubes. It must, however, be remembered that he is speaking only of rabbits and mice.

There are many considerations which lead me to believe that either Bacilli or spores must, in the majority of cases, pass into the urine and be voided with it; and that this may serve as one of the most important carriers of contagion. I shall therefore consider it a little more in detail. First, with regard to the glomeruli. It is quite possible that after rupture of the capillary network has occurred, and a quantity of blood has been extravasated, very few of the Bacilli may traverse the tortuous channel and reach the bladder; but what we know of the passage of blood in the urine, under analogous conditions, leads me to believe that some small quantity must pass. Again, I have frequently observed that individual Bacilli can pass through the capillary wall, and even through the walls of large vessels. Some may therefore pass into the urine whilst the network is as yet unruptured.

Again, we know that the urine is an excellent nutrient fluid for *Bacilli*, and that they can go on to the formation of spores. Once escaped, therefore, into the urinary tract, one or two

Bacilli may serve to contaminate the whole of the urine.

Apart from the glomeruli, the relations of the vascular supply of the pyramids may explain a more direct mode of entry of *Bacilli* by rupture of the capillaries, or, of course, they may escape into the bladder; but the importance of the other method is, I think, especially in the possibility of the acquisition of a

more stable form and of spore formation.

Anthrax has been communicated by Feser and others by means of inoculation with the urine of animals suffering from the disease. When we consider the very numerous ways in which the poison thus excreted may, having acquired a stable form, become dried on fodder or hay, or spread on the ground in manure, and subsequently be introduced by various channels into the system, especially of animals which, like cows and sheep, are apt to have their food more or less mingled with excreta, we need seek for no more ready mode of preservation and conveyance of the contagion. This fact, if it leads to more rigorous sanitary precautions (such as the thorough destruction of all litter, &c.) suggested by these observations, may prove of great value in limiting and controlling the disease.

The widespread distribution of the *Bacilli* in the capillaries of all the organs which are concerned in the most important functions of animal life, is suggestive of other modes by which they may cause death than by their direct action on the blood.

Unquestionably, they must interfere with oxygenation, by taking from the red corpuscles their needful amount of oxygen and appropriating it to their own growth; but their mere mechanical effect must also be considered, impeding, as it must, the action of the heart, the circulation and air absorption in the lungs, the excretory functions of the intestines and kidneys, and so on of nearly all the organs. And in corroboration of this view are the often observed facts of preservation of intelligence and some power of action till very shortly before death, of the lowered temperature which often precedes death, and of the absence of the ordinary symptoms of septic poisoning, properly so-called: the defective coagulation depending on the mechanical presence of Bacilli. So far as I have seen, there may be but slight changes in the blood taken from a large vessel, whilst the capillaries of the organ are crowded with bacilli, and the phenomena associated with early decomposition are much less frequent than in many forms of blood-poisoning.

QUARTER-EVIL, BLACK QUARTER, OR BLACK LEG.

The disease commonly known by these names is usually regarded as one of the forms of anthrax. It is widely spread throughout this country; and different names are used to de-

signate it in different counties. Quarter-Ill, Joint-Ill, Quarter-Felon, Speed, Hasty, Puck, or Pook, Schewl, and Inflammatory Fever, all appear to be merely local appellations for the same disease. A similar affection is described as Erysipelas carbunculosum (Armitage), Emphysema infectiosum (Böllinger); and in Germany it would appear to be classed as only a variety of anthrax, as "Milzbrand Karbunkel," or "Milzbrand Emphysem," and "Rauschbrand." In France the corresponding form is called "Œdème charbonneuse," or "Œdème malin." But I cannot positively assert that any of these names indicate a disease identical with Quarter Evil.

Although the disease is well-known to most stock-owners, and in many parts of the country occurs with great regularity at certain periods of the year in some pastures, it is desirable that I should mention the principal common features of the disease under consideration, in order that no confusion may arise. This is the more necessary, as most authors do not separate this disease from anthrax, but consider it is only one manifestation of that disease. I shall point out that there are certain differences from anthrax, in the conditions of its occurrence, its mode of apparent spread, and its pathological appearances, which are sufficiently marked to suggest a provisional separation from anthrax.

For many of the facts with regard to this disease I am indebted to my colleague, Mr. Banham, who has seen a good deal of it in Cambridgeshire.

The animals usually affected are cattle under two years of age.

Young sheep are occasionally victims to it.

It occurs chiefly at certain periods of the year, especially the spring and early summer, though not by any means necessarily limited to that time, cases sometimes occurring in October, and its chief prevalence is amongst young cattle recently put out to graze. In some districts there are certain pastures, even certain fields or paddocks, in which cases occur regularly year after year, whilst in other fields cases never break out. So far as the information received extends, these pastures are commonly lowlying lands, with defective drainage, clay and calcareous soils being especially favourable to its development.

Whatever the cause of the disease, it would appear, if infectious at all, to be so only to a very slight degree. In some places cases occur sporadically year after year, only one or two animals dying, and the others remaining perfectly healthy, removal from the field to another being the only precaution required. Sometimes a number of animals succumb, but it

rarely extends beyond one farm or one lot of stock.

The first symptom observed is that the animal isolates itself

from the herd, lies down, shivers, refuses its food, appears dull and listless, and there may or may not be symptoms of fever. The pulse is irregular, feeble, and rapid, 80 to 120 per minute; if fever is present, the mouth is hot and dry, the conjunctiva reddened, and sometimes spotted with petechiæ. Some small swellings appear, either on the loins, back, neck, head, brisket, or on one or more of the limbs (usually on the shoulder or quarter). These swellings are at first hot and painful; they rapidly increase in size, and then become cold and painless, and form diffused emphysematous swellings, occupying an extensive surface, which, when tapped with the fingers, produce a peculiar crepitating sound, and if cut into, dark frothy bloody fluid exudes. Large surfaces are frequently found in this mortified state without being preceded by smaller carbuncular swellings. Lameness is of course, in these cases, a prominent symptom. The animal usually lies in this condition for from six to eight hours, then becomes stupid and unconscious, and finally dies in a comatose state.

The course and duration of the disease vary; cases have been known to live from three to seven days, and it is stated by Röll that the swellings sometimes break at one or more points, discharging a gangrenous, ichorous, or tenacious bloody fluid; and it is said that the animal may recover, but Mr. Banham says that these cases are rare, if they ever occur, in England. Most commonly the disease runs a quick course, and kills the animal in from twelve to fifteen hours.

The usual post-mortem appearances are: Rigor mortis only slightly developed, the carcass greatly swollen, owing to the subcutaneous emphysema, and also, in some cases, to tympanitis. The blood extravasations are usually confined to one quarter, but they may be found in any part of the body. The blood is usually dark-coloured, fluid, and imperfectly coagulated, and, according to some authors, it has a peculiar odour. Decomposition sets in very rapidly, and the blood soon swarms with bacteria.

The lungs are said to be congested, the bronchi often filled with frothy mucus. The spleen is usually of normal size and consistence, the liver of healthy appearance, the intestines sometimes containing ecchymosed spots, but usually healthy.

But the most striking changes are found in the affected limb or quarter. The integuments are distended by the swelling of the subcutaneous tissue. On cutting through the skin, a large quantity of blackish, almost tarry, frothy fluid, filled with bubbles of gas, is found in this position. The swelling and gangrenous condition are not limited to the subcutaneous tissue, but extend deeply through the intermuscular planes separating the muscles. The muscles themselves are of blackish-red colour, mottled with points of ecchymosis, and there is a similar ædema of the connective tissue between the bundles of muscular fibres, which separates the individual bundles, in a manner analogous to that in which the exudation in the lung in pleuro-pneumonia of cattle separates the pulmonary lobules.

The diseased quarter is said to be very deadly to pigs and dogs, and it occasionally happens that men who dress the carcass, if scratched, die very rapidly with a peculiar form of bloodpoisoning; yet it is stated to be a common practice to cut off and bury the affected quarter, and to cook and eat the rest,

without any known ill-effects.

By some authors * the disease is divided into two forms, the one affecting the quarter, the other chiefly involving the intestines. In the latter form the chief symptoms are the passage of bloody fæces with tenesmus, occasionally, also, the urine being bloody; great tympanitic distension of the abdomen, and rapid prostration. The mucous membrane of the intestines is greatly swollen and intensely congested, or almost gangrenous; they contain soft bloody fæces. This is most marked in the large intestine. The mesenteric glands are greatly swollen, congested, and softened, and contain hæmorrhagic spots. There may be bloody exudation in the peritoneum.

This form of the disease appears to be somewhat allied to the

so-called Mycosis intestinalis.

It may be well here to point out certain features in which the disease usually differs from splenic apoplexy.

1. Its limitation to certain localities and certain periods of the year, and the apparent absence of direct contagion in most cases.

2. The gangrenous emphysematous nature of the swellings, which is not usual even with local forms of anthrax.

- 3. The general course and long duration of the disease in some cases.
- 4. The presence of coma and convulsions for some time before death.

5. The absence of swelling of the spleen.

6. So far as most observations go, the absence of the characteristic anthrax *Bacillus* from the blood.

And, as I shall further show, the disease may be exactly reproduced in rodents by inoculation from various parts of the body, without the discoverable presence of anthrax *Bacillus* in the blood or organs of the inoculated animals.

On the other hand, in no inoculation have I succeeded in

^{*} A full account of these diseases is given by F. Hable in the 'Oesterreichische Vierteljahreschrift für Wissen. Veterinärkunde.' Bd. li. H. I, which has been translated by Mr. Banham.

producing anthrax, even in animals very highly susceptible of infection by its poison in very minute doses, such, for instance,

as mice and guinea-pigs.

There has been great difficulty in procuring any material from this disease for investigation and experiment, only two outbreaks having been reported early enough to allow of a personal inspection. In two or three cases specimens of blood from the affected quarter, or from the jugular vein, and of other fluids, have been forwarded, but in every case the material had not been properly secured from sources of decomposition, and proved inert on inoculation, showing that in this, as in many other diseases, the specific virus is destroyed by decomposition of the fluid containing it.

The most satisfactory case was one in which Mr. Banham was able to make a post-mortem examination and to secure specimens of the various fluids and tissues of the body at a suffi-

ciently early period after death for examination.

In October 1879 an outbreak of Black Quarter occurred in the parish of Madingley, a village about three miles from Cambridge, in a herd of ten or twelve young animals, four of which succumbed to the disease. The usual prophylactic treatment, viz. change of pasture and food, with the administration of saline purgatives followed by vegetable tonics, was adopted, and the other animals remained perfectly healthy.

Mr. Banham states that most of the land in this district has a loamy upper soil, with a clayey subsoil, and in consequence the water remains on the land for a considerable time, making it very "clung" and hard to work, what is commonly called

"heavy" land.

Through the kindness of Mr. Page Wallis, of Cambridge, Mr. Banham was enabled to make a post-mortem examination of the last of the animals which died during the outbreak, twenty hours after death, and I examined the tissues and fluids about five and a half hours later.

Mr. Banham's report of the post-mortem examination is as

follows :--

"The carcass was extremely emphysematous, the subcutaneous tissue more or less distended with gas, and infiltrated with reddish gelatinous fluid, this condition being, however, much more marked on the right shoulder and hind quarter, the left side of the body being less affected. On removal of the skin, the muscles beneath the places most affected with emphysematous swelling were of a dark colour, and infiltrated with frothy, gelatinous, dark-coloured fluid. The muscles most affected were those surrounding the left humerus, both tibiæ, and the right lumbar region and shoulder.

"Abdomen tympanitic, and, when opened, a quantity of redcoloured fluid escaped from the cavity. The stomach and intestines appeared healthy. The spleen was slightly enlarged and softened. Liver normal to the naked eye. Kidneys of

natural size and colour.

"Pleuræ contained an unusual amount of fluid, serous membrane smooth and glistening. Lungs fairly healthy, except at the lower third of the left lung, which appeared airless, or nearly so, firm, of dark-red colour and sank in water (probably collapse). Heart and pericardium healthy. Blood in ventricles of heart had coagulated to a firm clot. Similar coagulation had taken place in the large veins."

Blood from the heart and jugular vein and spleen, serum from the heart and from the affected quarter, were preserved in capillary tubes hermetically sealed, and examined by myself about

five hours later, with the following results:

The serum from the affected parts contained only a very few rods, and some few spores similar to those found in other parts. The serum from the blood of the heart contained very few red corpuscles, but in it were seen scattered spores and pieces of filaments attached to them. These spores were nearly 1.4μ in length, and about 6 or 7μ in diameter, hence they were much larger than ordinary bacteria.* The filaments attached to some also resembled pieces of the ordinary anthrax bacillus rods, they were of very delicate outline.

The spleen was found to contain a very large number of oblong spores, some free, others with pieces of the filaments

attached to them.

From these fluids animals were inoculated with the following results.

Experiment I.—A guinea-pig, inoculated at 12 P.M. Oct. 9, with fluid from the spleen, was found next evening, twenty hours after the inoculation, to be apparently paralysed in the hind limbs, with extremely low temperature. There was swelling and emphysema of the walls of the abdomen, and of the tissues of the inoculated limb. It was killed at 9 P.M., and the tissues and fluids at once examined.

The skin of the thigh and of the adjacent abdominal wall was found to separate very readily; the whole of the subjacent tissues being of blackish, almost gangrenous, appearance. The

^{*} It must here be mentioned that *Bacilli* may be found in blood and tissues after death, which are not anthrax *Bacilli*. The proof of their anthracoid nature is found only in their effect when inoculated in another animal. This point is too complicated for discussion here. For some facts I may refer to a work of Dr. Timothy Lewis, in 'Microscopic Organisms found in the Blood of Man and Animals,' p. 41. Calcutta, 1879.

muscles of the thigh and abdomen were also swollen, of very black colour, and contained numerous ecchymoses. The spleen appeared perfectly healthy, and not apparently enlarged.

On examining the serum from the swollen connective tissue of the thigh, it was found to contain ordinary bacteria and micrococci, the bacteria in very active movement. Serous fluid, which was somewhat blood-stained, obtained from the centre of affected muscles, also contained a few moving bacteria of the common form. The blood from the heart appeared to be perfectly healthy, and contained neither bacteria nor micrococci of the ordinary form. That from the spleen swarmed with bacteria and micrococci, a few of the rods being quiescent, but even these having the form of ordinary decomposition bacteria. The lungs appeared perfectly natural. I could not collect any fluid in the serous cavities.

In this case, then, there were none of the usual characters of anthrax, and neither bacillus rods nor spores were to be discovered in the spleen or tissues; but there are two noteworthy points, viz., that the emphysematous swelling of the inoculated limb and of the abdominal wall, the almost gangrenous state of the tissues and the black condition of the muscles were present, reproducing in this respect the clinical features of the original disease. At the same time there was evidence that decomposition had already commenced during life, in the fact that the splenic tissue swarmed with bacteria. I have never found this before in an animal killed and examined at once. In the other inoculations I was unable to examine the tissues immediately after death.

Experiment II.—A guinea-pig, inoculated in the leg with serum from the affected quarter at the same time as the preceding, was found dead at 8.30 A.M. on October 11. I was unable to examine the body till 5 P.M. The whole of the quarter where the inoculation was made was greatly swollen, the hair and epidermis were detached with great ease, even by a slight touch. In the subcutaneous tissue of this limb and over a great part of the abdominal wall was a quantity of deeply blood-stained exudation. The muscles of the thigh were swollen and black, containing small spots of ecchymosis. The other hind limb was not affected. All the viscera and the serous cavities were apparently perfectly healthy, and the spleen was free from enlargement.

The blood-stained serum from the subcutaneous tissue contained many red blood-corpuscles, a few ordinary moving rod-shaped bacteria and some micrococci, no still rods or spores. The peritoneal serum contained no bacteria at all, nor did the blood from the heart. In the spleen were found only ordinary

moving bacteria. In the pleural serum were a few longer moving rods, some measuring from 8 to 12 μ in length, showing signs of incipient division, but not containing spores.

Experiment III.—Another guinea-pig was inoculated at the same time with blood from the heart, which, it will be remembered, contained spores resembling anthrax spores, and was also found dead thirty-three hours after the inoculation, having had a temperature ten hours after of 106°, and in the evening, twenty-two hours after inoculation, of 100·4°. The appearances, both naked-eye and microscopical, were precisely similar to those in the preceding: no Bacilli nor definite spores were found.

Experiment IV.—A rabbit, inoculated in the leg with fluid from the spleen, presented on the following day a very marked swelling of the whole of the inoculated limb, much resembling that in the guinea-pigs. The temperature was 104° for the next two days, morning and evening; on the third day it was 103° to 103.4°, on the fourth, 102.8°. The swelling gradually declined and the animal recovered. These experiments were not continued further, for at the time they appeared to me to indicate that the material employed must have been too much decomposed to be of value. But on reconsideration I am inclined to think that this may have been an error.

In all four of these experiments the sequel was different, both from that of the inoculation of ordinary septic material, and from that containing anthrax Bacilli. In all there were the peculiar emphysematous swelling and the black condition of the muscles, with ecchymoses scattered through them, which are characteristic of and give the special feature to the disease known as black-quarter. In no other experiments with animal fluids have I seen anything at all similar produced. Moreover, it was produced alike with the blood from the heart, juice expressed from the spleen, and the serous exudate from the

affected quarter.

I would especially notice the fact that the spores found in the spleen gave no signs of growth; that they were, in fact, not reproduced in the slightest degree in the animals inoculated. Now what has commonly been observed is that if material containing a quantity of anthrax spores is inoculated, anthrax Bacilli, being developed more rapidly in the body than other Bacilli, produce the symptoms of splenic apoplexy. Here we have evidently to deal with a poison equally rapid and fatal in its action in some cases, producing decomposition actually during life, but apparently hindering the development of Bacillus anthracis, if that organism is really present.

Some other experiments, previously made, show that the

blood of quarter-evil loses its peculiar properties when decomposition has become at all advanced, indeed that, as has been shown to occur in many cases, such blood may be inoculated in rodents without causing any deleterious effects at the time,

though in some cases later effects are developed.

Some blood taken six hours after death from the jugular vein of a cow which died of black-quarter on June 24 was received by me on June 28. The blood had been placed in a small stoppered bottle, completely filled and well secured; nevertheless when received it was highly offensive. It contained only a few common rod-shaped and other forms of bacteria. About four minims of the blood were injected under the skin of the thigh of a guinea-pig. It was gradually absorbed, the animal showing no symptons whatever. About one drop was similarly injected beneath the skin of the back of a mouse, and no results were produced.

Another outbreak, which occurred near Cambridge, has given further opportunities for investigation of this disease. Two cases being reported on February 24, 1880, Mr. Banham went down to Cambridge to examine them and procure material.

He found that a post-mortem examination had already been made, and only the affected quarters reserved for his inspection. Nothing abnormal was found in the internal organs. In both cases the affected quarter presented in the most striking manner the usual characters, which have already been described, the subcutaneous and intermuscular connective tissue being infiltrated with blood-stained serum, and the muscles of blackish-

purple colour mottled with spots of hæmorrhage.

Blood and serous exudation from these parts were secured in capillary tubes about twenty-four hours after death, the weather being cold and almost frosty in the interval. On examining these fluids about thirty-two hours after death I found no sign of decomposition. In the blood and the blood-stained serum which flowed from the quarter, the only striking fact was the entire absence of coagulation, the blood-corpuscles remaining entirely isolated. I was unable, with the highest power which I had at hand (12th oil-immersion of Zeiss), on prolonged examination to detect any bacteria of any form whatever, even micrococci being absent in the freshly-opened tubes containing only blood. No traces of Bacilli or their remains were discovered. It is the more remarkable that bacteria should have been absent, as it is not uncommon for the blood and tissues of the affected parts to be found in a state of decomposition directly after death, sometimes even during life as we have already seen.

Inoculation experiments were made with the blood and serum

from the affected quarter, as in the preceding case.

Experiment I.—A rabbit was inoculated in the thigh with a drop of the serous fluid which had been collected. No symptoms or local results followed.

Experiment II.—A rabbit was inoculated with 2 minims of bloody fluid (chiefly blood) from the affected quarter. No

symptoms, either local or general, ensued.

Experiment III .- A guinea-pig was similarly inoculated with

the blood. No effect was produced.

At first sight these experiments seem to contradict the results obtained with the previous case. But I must point out a striking difference in the method employed, or rather in the material used. In the former case most of the results were obtained by inoculation with either the spleen or the blood from the general circulation, inoculation with which gave rise to the local phenomena of the disease. Moreover, all the fluids used contained bacteria of some form. In the present case only the local serum and blood were employed, the other fluids not being obtainable, and no bacteria could be discovered in the fluids inoculated. It would, therefore, appear probable that the specific virus does not reside in the local lesion, or is present in a far less active form there than elsewhere. And this again would appear to suggest that the affection of the quarter or limb is secondary to and not the cause of the general disease.

I do not desire to speculate further on this subject, which can only be solved by further experiment. Two conclusions alone appear to me to be warranted. First, a scientific one, that the disease must be different from anthrax, which is so readily communicable to lower animals, especially rodents, that if this were a form of anthrax, some of our experiments must have produced that disease, instead of reproducing either the identical quarter-evil or no effect whatever. Second, a practical conclusion, that the danger of infection is as great or greater from the unaffected parts of the body as from the obviously affected and gangrenous quarter. The practice of burying or burning the obviously diseased part only, and taking no steps to destroy or disinfect the rest of the carcass, is therefore clearly

fraught with danger to other animals.

I reserve for a future occasion the description of the microscopical examination of sections of the hardened organs of animals dying of the original disease, and as a result of inoculation from it. I may, however, state that the principal results of this examination were to establish the identity of the inoculalated with the original disease, and to confirm the conclusions drawn from the naked-eye appearances, but beyond this they have not afforded any clue to the pathology of the disease.

In connection with the experiments on this disease it is right

that I should mention the following observation, made in their course. That flagellated protozoa may exist in the blood of healthy animals is now well known from the observations of Rättig, Wedl, Lewis, and others. But I am not aware of any similar recorded observation, and it may be found to have a

pathological bearing at some future date.

An apparently healthy white mouse was inoculated in the tail with some blood from the diseased quarter, and as it presented no symptom beyond some swelling of the tail, it was supposed to have escaped all mischief. However, the swelling did not subside, but became gangrenous, and towards the middle of the second week it began to emaciate and the belly swelled; finally, it died on the fourteenth day after the inoculation.

Examination five hours after Death.—On opening the abdomen there was found to be considerable opalescent exudation in the peritoneum; the abdominal wall was also swollen and infiltrated. The tail, where inoculated, was considerably swollen. The other organs were fairly healthy. Before, however, I examined the organs, I took, as usual, some of the exudation in capillary tubes, and also some serum from the pleura and pericardium,

and examined these microscopically.

I naturally anticipated that I should have found the ordinary characters of semi-purulent serous effusion in the peritoneal fluid, but my astonishment was great when I found the fluid to be swarming with minute organisms swimming actively about in every direction, moving apparently by means of two long

cilia or flagella.

These organisms I may now describe more fully. They were ovoid, rounded, or more commonly, when in motion, pyriform or balloon-shaped; very translucent, of delicate hyaline structure, sharply defined. At the posterior extremity were two long fine lashes, moving with a wavy motion, much like that of cilia, and apparently serving to propel the organism forwards. In each could be seen a central clear space running lengthwise, wider at its central part, and apparently contracting and expanding. At the anterior extremity a more refractile part could be seen, apparently forming a sort of oral aperture. Their protoplasm was highly contractile, and they changed shape, elongating and contracting very readily. In size they were, when at rest and round, about from 2 to 4.5 µ in diameter; when elongated, some reached 6 μ in length, and 1.5 to 2 μ in width. I was able to observe their movements for more than half an hour, but it was not until they were quiescent that the other cilia could be seen, and they were not well seen until stained. A more thorough study of their character was possible after staining with methylaniline violet, and mounting in glycerine

jelly. It was then found that they had each at least six lashes, sometimes apparently eight, two of these being at the caudal extremity, and the remaining four attached to the anterior extremity, and, so far as I could see, around the oral aperture. These cilia or flagella were extremely delicate, so as hardly to be visible without staining. In length they were from 5 μ to 8 μ , or nearly twice as long as the length of the body. After staining, the body of some presented numerous fine granules, perhaps due to coagulation of the protoplasm; in each one the anterior extremity stained more deeply, and in some formed a distinct ring. In all the central space came out very clearly.

There can, I think, be little doubt that the anterior cilia were not visible during life, owing to their very active movement; that the posterior, which seemed to propel the body,

were really serving rather as rudders to steer it.

I supposed at first that these might be parasites which had escaped from the intestine, bladder, or elsewhere, and I therefore examined very carefully the contents of these viscera throughout their whole extent, and also the blood, &c., but failed to find any trace of similar organisms in any of these; whilst all the specimens from the peritoneal exudation swarmed with them, and contained no other organised constituents of importance.

What is their nature and significance I must leave for future

observation to discover.

OTHER ANTHRACOID DISEASES.

Amongst the diseases of the class of blood-poisonings which appear to be allied to anthrax are two which especially affect horses, and are known respectively as Cape Horse-sickness and Loodiana fever. At present it is uncertain whether these are identical, and also whether they are or are not produced by the

same poison as anthrax.

The former disease, "Cape Horse-sickness," is one to which attention has been specially directed of late by the Zulu war. It appears to be endemic (or enzōotic) in certain regions, notably of Natal and Zululand, occurring chiefly in low-lying lands and in valleys, though cases sometimes occur at high altitudes. The disease usually prevails at particular seasons of the year, especially in moist hot weather, or when rain occurs after heat, disappearing almost entirely both in hot dry weather and also in the cold season. Almost universal popular belief and recent experience attribute the attacks to

eating the wet grass under such conditions, and it has been observed that cases are especially prevalent when horses are

allowed to graze on, or are tethered out, in wet grass.

The onset is usually sudden, with dulness, loss of appetite, rise of temperature, injection of the conjunctivæ, accelerated breathing, cough, and other symptoms, resembling those of inflammation of the lungs. There is evidence of considerable exudation in the bronchi, and in some cases a large quantity of mucus is discharged from the mouth and nostrils. The temperature is usually considerably elevated, reaching 105° to 108° Fahr. In one form of the disease there is great swelling of the tongue and of the cellular tissue of the throat, causing death by suffocation.

The disease is usually fatal, sometimes in a few hours, more rarely in two or three days, and cases of recovery are on record.

The post-mortem examination shows usually rapid decomposition. There is often a quantity of straw-coloured fluid in the pleura; the lungs are engorged, and along the margin and in patches beneath the pleura is a large quantity of yellow lymph. On section of the lungs, blood and serum flow freely from the cut surfaces, and frothy yellow mucus from the bronchi.

Ecchymoses are invariably found beneath the endocardium of the left ventricle, the heart is soft and flabby, and there may be

serous effusion in the pericardium.

The following account of a case of this disease, for which I am indebted to Mr. R. Moore, M.R.C.V.S., gives a good illustration of the symptoms and post-mortem appearances which are usually observed. Material was sent to me for examination:—

"The horse had been doing very little work, and was in good condition. He was noticed to be dull in his work the day before he was taken ill. He was first noticed to be ill at 3 o'clock P.M., and was seen by me at 6 o'clock the same evening, when he was presenting the following symptoms: Temperature at the mouth very high; pulse imperceptible; visible mucous membranes highly congested and of a livid hue, especially round the gums; breathing very laborious; extremities and surface of body cold; crepitation in all parts of both lungs on auscultation. At 9 P.M. the characteristic discharge of froth issued freely from the nostrils, and he expired about 10 P.M.

"Post-mortem Examination .- Body in good condition; flesh

dark in colour.

"Chest .- Large accumulations of lymph in patches in the

areolar tissue on the surface of the lungs and pericardium. Both lungs large in appearance, but not of a very dark colour. When cut, they presented great capillary congestion, which pervaded every inch of the lungs. The bronchial tubes, both large and small, were very much congested, and from them issued the froth. No part of the lungs heavier than water. Pleura slightly affected.

"Heart.—Deposit of lymph in pericardium. Endocardium

of right ventricle congested and livid, almost black.

"Liver and Spleen of normal size, but darker in colour than

"Kidneys .- The left kidney very much congested, the right one healthy.

"Intestines.—There were patches of inflammation over all

the bowels and mesentery.

"The Blood very dark and fluid."

The condition of the lungs here described is, as I shall hope to show, of considerable interest in relation to some experiments which I made with the disease upon animals. The lung affection is evidently a marked and constant feature of the disease, and the principal conditions are subpleural exudation and a sort of bronchial catarrh. These I have been able to reproduce by inoculation of the disease in guinea-pigs and mice, with characters apparently almost identical with those seen in the original disease. Moreover, these lesions were in both situations attended with the presence of bacilli in the affected parts in the initial stages of the morbid process, disappearing as it reached its maximum.

Of the original disease I have only had the opportunity of examining one specimen, a piece of the affected lung. This was not in a condition for examination for bacteria. The principal change was evidently an inflammation, slight in degree, which followed the lines of the bronchi, but in many places was evidently outside them. Some of the bronchi, especially those of medium size, showed signs of catarrh, but there were a large number of which the mucous membrane appeared

absolutely healthy.

But in a large number of sections there were very marked changes around the vessels supplying the walls of the bronchi. Some of these were completely plugged by masses of leucocytes, and around them for some little distance were the ordinary appearances of inflammation. There were here and there patches of exudation, extending to close beneath the bronchial mucous membrane.

The epithelial lining was in many of these perfectly healthy

in appearance, but here and there the sub-mucous tissue considerably swollen and infiltrated. But the absence of bronchial catarrh was in most parts very striking; only rarely was there any exudation in the air-cells. The pleura was thickened in scattered patches, apparently by exudation into its substance, which was partly inflammatory, partly hæmorrhagic but no superficial effusion was observed.

Let me anticipate what I shall have to say with regard to the results in rodents, for the sake of drawing a parallel between the condition here described and that found in them. In two mice, the one inoculated from the other, the first having been inoculated from a guinea-pig, I found these changes. In the first, marked pulmonary congestion with slight subpleural exudation. In this lung I found commencing inflammation of the subpleural tissue with slight hæmorrhagic exudation on the surface. Both in patches of the lung tissue, and in these deeper pleural layers and in the exudate I found here and there abundant bacilli, agreeing both with those cultivated from the peritoneal

In the second animal there was no pleural exudation. I found, however, that in some of the larger bronchi, those near the root of the lung, there was incipient catarrh; that this was not general, but limited to some parts of the circumference of the tube examined, and that side by side with affected bronchi were others absolutely healthy.

exudation and those seen in the other animals. These were only found locally at the spots of commencing inflammation, and

Then I found that these affected parts corresponded with vessels which were the seat of commencing inflammation, and that around them in the tissues between them and the bronchi were long bacilli, corresponding exactly with those seen in the peritoneum, and in the lung and pleura of mouse No. 1. These bacilli one could trace passing through the walls of the vessels, and some were also seen in the blood within the vessels. As the inflammation began, they seemed to disappear, indicating the probability that inflammation is attended by their destruction.

Similar bacilli were also found in the blood-vessels of the kidney, but I have as yet failed to find them in other

I may now describe these experiments. Some blood, which had been preserved in carefully filled capillary tubes, and some mucus from the nostrils were the materials used. The blood when examined was found to contain some spore-like bodies, a few remains of short rods, and also a few micrococci. A

guinea-pig which was inoculated with the blood died in seventeen hours. When examined, four hours after death, there was very slight inflammation and serous exudation at the seat of inoculation, the heart was filled with coagula, the organs generally appeared natural. The blood contained numerous rods and also free spores; that taken from the heart containing the greatest abundance, more than the splenic blood.

Another guinea-pig, inoculated at the same time as the first, did not die till twelve hours later; it presented similar appearances, but there was more exudation at the points of

inoculation.

Some blood-stained serum from the heart of guinea-pig No. 1 was kept in capillary tubes at normal temperatures for four weeks, and it was then used to inoculate two mice, with very minute quantities. One of these died in twelve hours, and was examined after nine hours. There was no local inflammation in the tail where it had been inoculated, and no inflammation of the peritoneum. The spleen was large and rather soft, the other viscera healthy, except slight injection of the pleura. Blood taken from the right ventricle was found to contain very numerous long rods, many of them measuring 80μ in length, some containing spores.

Another mouse was inoculated in the tail with a minute quantity of serum collected in the peritoneum of the previous one, and, like it, died in twelve hours. When examined, three and a-half hours after death, some inflammation was found extending along the surface of the abdomen, and some slightly turbid exudation in the peritoneum; the latter containing a large number of long rods like those in the previous case.

On comparing together specimens of blood from these animals and some from cases of undoubted anthrax, a very close similarity is discovered. The chief differences are that the bacilli in most cases of true anthrax are much more abundant; and that the bacilli in Cape fever were more slender. But I find a considerable difference in this respect in bacilli in anthrax; in specimens from some animals they are decidedly smaller than in others. In other respects, their varying length, junction of two segments at an acute angle, or formation of long rods made up of nearly equal segments, they are very similar in appearance.

In the viscera I did not find any important changes beyond

those already mentioned in speaking of the lungs.

It is indeed a most striking feature of all these forms of blood-poisoning that the changes in the organs are so slight in degree or may be quite undiscoverable. There is one other point of distinction from anthrax which I must note, viz., that the growth of the bacilli in cultivating fluids is much less voluminous than that usual with anthrax

bacillus, although in its main features it is very similar.

I was able to cultivate the organisms found in the blood, spleen, and peritoneal serum of the guinea-pig for several generations; and also those from the peritoneum and pleura of the mice subsequently inoculated. The various stages were as follows: they were repeated again and again in successive cultivations, so that the various stages could be watched side by side in crops of different ages.

The rods first formed were long and slender, measuring sometimes (as in the mouse examined three hours after death) as much as 15μ to 90μ in length, and about 6μ in thickness.

They were usually quite motionless, but some of them moved

very slowly along with a slightly wavy motion.

These rods, when cultivated, grew rapidly to an enormous length, some extending almost the entire width of the field (about $200 \,\mu$). In these longer ones there were sometimes, at an early period, indications of subdivision into shorter rods, from 10 to 30 μ in length; but they often produced the twisted rope-like forms before any subdivision. Once or twice a moving stage was observed in rods about 15 to 25 μ in length; but this was uncommon.

In these elongated rods the formation of spores proceeds in the usual manner, though, on account of their smaller size, the steps could not be so readily watched as in the ordinary anthrax bacilli.

These spores were pretty regularly arranged in a definite order in the rods, sometimes in couples, *i.e.* the alternate spaces narrower.

In the rods which had undergone partial division before the formation of spores, these were frequently more closely set together; and the filaments broke down more rapidly into a mass of spores, some of which had escaped from the filament,

others had been formed by its division.

Although these observations are not sufficiently extensive to warrant any definite and final conclusions, they seem to me to indicate that the disease, although allied to anthrax, may yet be distinguished from it by the predominant affection of the lungs, by the relatively small number of bacilli which are found in the blood and organs, as well as by the size of the bacilli themselves.

In true anthrax the growth of bacilli is enormous, their distribution general throughout the body, and in many of the organs the blood-vessels are often found to be plugged by masses of them.

In the Cape horse-fever they are produced in relatively small numbers, and display a preference for certain sites, circulating in the blood and wandering out of the vessels at certain points, at which they appear to set up inflammatory changes, and lead to characteristic results which form the anatomy of the disease.

In anthrax the blood is the site of their growth, and the sole anatomical characteristic in most cases is the swelling of the spleen, which is due to the peculiar relations to the blood. At the same time it must be allowed that cases of anthrax do occur in which the morbid process does not specially affect the spleen, an example of this is seen in the sheep inoculated with anthrax.

I am not yet in a position to say whether these two diseases are identical, viz., anthrax and Cape fever, or whether the bacilli are the same in both; my own belief is that they are

not identical, but are two members of one group.

With regard to Loodiana fever, which appears to be very closely allied to, if not identical with, the Cape horse-sickness. I can say but little. There appears to be usually more swelling of the throat, with infiltration of the intermuscular planes, than in the common form of Cape fever.