

Some Experimental and Epidemiological Observations on a Particular Strain of Pleuro-Pneumonia.

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In a previous article* I described the specific pathological anatomical lesions which are produced in the different tissues as a result of the injection of lung-sickness virus. The strain used happened to be of extreme virulence, and provided the necessary material for the following experiments. These were all carried out at the Institute, and they showed so many interesting details that a full report is advisable. The history of the origin of this lung-sickness strain is as follows:—About 400 head of cattle were inoculated prophylactically against lung-sickness, subsequent to an outbreak of this disease in the herd. The Government Veterinary Surgeon, who inoculated the cattle, reported that the pleural exudate of an animal killed for the purpose was used. No details of the *post-mortem* examination of this animal were given, but it has to be particularly emphasized that nothing unusual was observed, as for instance swelling of the joints, and in our experiments this proved to be of considerable importance. The inoculation was carried out as usual into the tail. Shortly afterwards the Government Veterinary Surgeon was called to inspect the cattle owing to the fact that several animals had died and a large number showed progressive necrosis of the tail. In his report of the investigation, the following observations were recorded: A number of the cattle had purulent abscesses at the seat of injection, and in some of them necrosis extended half-way up the tail. The tails of about fifty head of cattle were amputated and for about the same number, similar measures were considered necessary. About ten or twenty were sick, and were expected to succumb in the following few days. The visible lesions were distinct swellings of the joints; in about 100 animals which showed no reaction in the tail, decided swellings of one or several joints were observed. In some, one joint, and in others, two or three joints were visibly affected. The movements of the animals were very stiff, and a few were unable to rise. Generally, the knees, stifles, fetlocks, hip-joints were affected, and in a few cases also the hocks, and in one case the neck. Nearly all the inoculated animals showed symptoms of stiffness. Those which reacted in a normal way to the inoculation and showed no swellings of the joints had an uncertain gait, and a rattling noise could be detected when they were walking. In some cases the temperature was taken at the time, and found to be 105° F. Generally speaking, young animals were most affected, but cows and oxen also showed lesions in the joints and were unable to rise. Joint evil had never been observed on this farm previously. During

* Notes on the "Pathological Anatomy of Pleuro-pneumonia *contagiosa bovum*" (Commem. Publication, Govt. Vet. Bac. Lab., 1909).

the following days about forty-five animals died. One sick calf, "D", was sent to the laboratory, but it died during transit. It was used for the following experiments.

The *post-mortem* examination* showed polysynovitis, tendovaginitis, sero-fibrinosa. A small quantity of the liquid in the joints was drawn up by means of a syringe, diluted in eighty times its volume of *Martin's* bouillon, and filtered through a *Chamberland* candle, F. The filtrate was then mixed in the test tube with 2 per cent. cattle serum, and kept in the incubator. After four days a distinct, typical growth of the pleuro-pneumonia organism was observed. Two small fibrine flakes of the same joint were rubbed in a mortar with a small quantity of *Martin's* bouillon, and 5 c.c. of the liquid was injected into two oxen.

(1) Ox 672 inoculated on the 15th December, 1908, in the dewlap. From the 10th day it showed a distinct swelling at the seat of injection, and hot, slightly swollen fetlock joints in both fore-legs. Twelve days after inoculation the animal was killed in extremis. The fetlock joint and the hocks were considerably swollen. *Post-mortem* examination showed the lesions of a myositis, synovitis sero-fibrinosa.† A culture was obtained with the exudate of one of the joints and gave a pure growth of lung-sickness.

(2) Ox 670 was also injected in the dewlap on the same day. Eight days later it showed a high temperature, swelling at the seat of injection, and slightly swollen joints. On the 12th day a polyarthritus had formed in nearly all joints. The animal died on the 3rd January, 1909. *Post-mortem* examination‡ showed a myositis, synovitis, and tendo-vaginitis sero-fibrinosa. The cultures obtained with the exudate of one of the joints gave a distinct growth of pleuro-pneumonia. In both cases, therefore, the same symptoms and pathological anatomical lesions were produced by means of the inoculation of the joint exudate in the calf "D". The culture proved the nature of pleuro-pneumonia. The swelling of the joints was especially pronounced on the fetlock, but nearly all the joints of the different bones showed lesions. The exudate in the joints increased, and there was a watery serous imbibition of the peri-articular connective tissue; in a few joints were fresh fibrinous filaments and a slight affection of the cartilage. We omitted to make cultures from each joint separately, and were therefore unable to ascertain if all joints were affected in the same way. The result of the cultures proved that the lesions in the joints were nothing else than metastasis of the lung-sickness virus circulating in the blood, and it was a peculiarity of this strain that it affected the joints to such a degree. Individual disposition must be excluded since 400 animals of different ages were attacked. On looking through the literature we find that *Nocard*, *Marek*, and *Huytra* state that only calves are susceptible for joint lesions as a sequel of a pleuro-pneumonia infection. This specific aggressivity of our lung-sickness strain on the articulations could be produced in the third generation of cultivated virus, but we failed to do the same with a culture made with material from one of the joints of the animals which had been injected with cultures.

Two cattle, 688 and 685, were injected on the 18th January, 1909, with 0.5 c.c. at the tip of the tail with culture from calf "D", generation 1. The only symptom observed during a long observation time was a slight swelling of the tail. Control animals inoculated with culture of a different strain showed the same slight local symptoms. The strain of calf "D" had probably lost its specificity through being grown on artificial medium.

* *Loc. cit.*, Notes on the "Pathological Anatomy of Pleuro-pneumonia contagiosa bovum," page 149.

† *Loc. cit.*, pages 149 and 150.

‡ *Loc. cit.*, pages 150 and 151.

These experiments give an explanation of the fatal results of inoculation, inasmuch as it was shown that they were sequels due to the pleuro-pneumonia micro-organism, which we had isolated in pure culture. That such a specific and virulent strain of lung sickness can do more damage was further illustrated by the following observations at the Institute:—

In the middle of February, 1909, an animal, No. 583, which was standing in the same stable where the cattle used for the above inoculation experiments stood, showed symptoms of pleuro-pneumonia, but a definite diagnosis was not made, because this disease was not surmised. It was thought that a perforation by a foreign body might be the cause of the symptoms. The temperature was rather high, the animal was off feeding and rumination. It was killed on the 23rd February, 1909. *Post-mortem* examination proved it to be a typical case of pleuro-pneumonia.

We failed at first to understand this spontaneous case of lung-sickness because in the herd of cattle to which this animal belonged, no outbreaks of lung-sickness had been noted for a number of years. The animal in question (583) joined this herd at the beginning of May, 1908, and ever since June in that year had been standing in the same stable—that is to say, for a year and a half it had been practically isolated. It was used for experiments in connection with the feeding of suspected poisonous plants. In November, 1908, cattle 671, 675, and 674 were put in the same stable and near this animal, 583. Ox 671 was used for the production of lung-sickness virus, and a pure culture of this virus was tested on cattle 675 and 674.

In the middle of December, cattle 670 and 672, which had been used in the experiment on the genesis of the joint-swelling were also placed in the same stable. The question now arises whether these animals were responsible for this spontaneous outbreak of lung-sickness. Although in the literature no definite case of a similar nature could be found, it was generally admitted that inoculated animals do not transmit the infection. *Nocard* and *Leclainche** state, based on observations of *Rossignol*, that it is impossible to infect a clean herd by means of the inoculation. They write as follows:—“ . . . L'inoculation caudale, meme quand elle est compliquée d'accidents consécutifs amenant la chute de la queue, ne constitue pas un danger au point de vue de la contagion pour des animaux sains qu'on a mis à cohabiter avec des inoculés pendant une période de quatre mois consécutifs. Les soins hygiéniques, donnés indistinctement, pendant quatre mois, aux inoculés, aux témoins et a des veaux nés pendant le cours de l'expérience, par la même personne, sont également impuissants à faire contracter la péri-pneumonie a des animaux sains, placés comme témoins au milieu de sujets inoculés. . . .”

The reports of *Hutcheon*† from the Cape Colony are entirely different. He states that the majority of farmers believe that through the inoculation, lung-sickness may be introduced into an absolutely healthy herd. He writes as follows:—“ . . . Is it possible for them to communicate lung-sickness to healthy cattle direct by inoculation into the tail? The large majority of European experts express the confident opinion that you cannot, and when the disease does appear in an apparently clean herd after inoculation, they assert that the infected animals must have contracted the infection previous to their inoculation, or before the inoculation has had sufficient time to confer immunity on them. Of course, it is impossible to prove the contrary, more especially as inoculation against lung-sickness is usually resorted to only when

* *Nocard* and *Leclainche*, “Les Maladies microbiennes des animaux”, page 487.

† *Hutcheon*, “Lung-sickness of Cattle”, *Agricultural Journal*, Cape Colony, December, 1905.

the disease has already appeared in the herd itself, or amongst some herds in the near neighbourhood. But from the numerous instances in which I have observed the disease to follow inoculation when no other reasonable explanation could be offered except that of direct communication by the inoculation. I am thoroughly convinced that inoculation is capable of communicating lung-sickness direct to a healthy susceptible animal. I am quite ready to admit that such cases are comparatively rare, but they do occur and are the means of introducing the disease into a previously healthy herd. I entertain no doubt whatever, and it does not appear so improbable that such cases should arise under special circumstances. . . .”

If we compare these notes with our observations, we can support the views of *Hutcheon*. Whether the infection was carried from cattle 671, 675, and 674 which were inoculated in November, or whether the other experiments in December were responsible for the spontaneous outbreak in 583, could not be immediately decided. If we take the incubation time of ten to forty days as usual, the animals in the last experiments, 670 and 672, strain “D”, must be the cause. All these animals showed lesions in the skin at the tail or dewlap, through which a considerable amount of serum was exudated. This material was of an unusual virulence, and its distribution was probably affected in the same way as in the infection from sick to healthy animal standing in its neighbourhood. Undoubtedly the last-mentioned two animals are responsible for the infection of 583.

Cultures were made with the pleural exudate from the animals, and a distinct and strong growth was obtained. This culture was used for the prophylactic inoculation of a number of cattle on the station, with the intention of testing its special virulence. About twenty cattle were inoculated with 0.5 c.c. of culture at the tip of the tail. All showed an enormous swelling of the tail and of the lymphatic glands on the os ischium. This proved that the reactions were unusually severe. Two animals, ox 662 and cow 585, showed swellings along the tail to the pelvis; all the organs in the pelvis were enormously swollen, and the animals had to be killed. Cow 585 showed symptoms of chronic intoxication, typical for pleuro-pneumonia; details of the *post-mortem* examination appeared in the article referred to previously.

The inoculated animals were of different ages, but none showed lesions of the joints, neither did the *post-mortem* examination reveal any indications of a joint affection. This result therefore does not give the direct proof that the lung sickness strain of 583 originated from calf “D”, but we stated before, that passing a strain through a culture or, as in this particular case, through an animal, its specificity to act on the joints becomes lost. On the other hand, the severe reactions and the fatal results of two animals prove that a most virulent strain caused the death of 583. Therefore the spontaneous lung-sickness case and all its sequels probably originated from the calf “D”, which was inoculated with the synovial exudate. The use of this strain for inoculation in practice was avoided, because as previously stated, a strain does not lose its virulence in the first generation by passing through cultures, and knowing this we feared that this virulent strain might give similar fatal results when used for prophylactic inoculation. The results of these observations was that subsequently all experiments with lung-sickness material were carried out in isolation stables.

The following conclusions may be drawn :—

(1) Lung-sickness virus collected from a sick animal and used for prophylactic inoculations in cattle, produced in animals of different ages a polyarthrititis sero-fibrinosa. This phenomenon was not due to individual disposition, as is occasionally observed in calves, but it was specific for the strain

of virus used by us. The subcutaneous inoculations of synovial liquid of animals affected by this particular strain produced, besides the typical local reaction, a poly-arthritis in all animals experimented on. The synovial liquid represented a pure virus, and the micro-organism of pleuro-pneumonia could be cultivated from it. The specific action of the strain became lost in sub-cultures or by passage through an animal, and no secondary joint affections could be produced subsequently by inoculation of cultures.

(2) Through the experimental or prophylactic inoculations of lung-sickness material, pleuro-pneumonia could be introduced into a healthy herd. The first generation of the culture maintained its virulence as a lung-sickness strain. From a practical point of view, the isolation of all inoculated animals is advisable. A strain used for the prophylactic inoculation of cattle should—whenever such is possible—be first of all tested on a few animals.

