MELIOIDOSIS IN A HORSE.

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The symptoms and the morbid anatomy of melioidosis in man are almost indistinguishable from those of glanders; the disease was first described by Whitmore, in 1913, under the title of "A glanders-like disease, occurring in Rangoon." It is, however, clearly differentiated from glanders by its etiology: *Bacillus whitmori*, the causative organism of melioidosis, possesses distinctive characters, such as its active motility and the curious corrugated appearance of its growth on glycerine agar, which distinguish it at once from *B. mallei*. Agglutination, absorption and fixation tests show that *B. whitmori* is closely related to certain strains of the glanders bacillus from India and Java, but that its kinship with the strains of *B. mallei* in the National Collection of type cultures is very remote.

We have seen the disease occur naturally not only in man but also in rats, cats, dogs, guinea-pigs and rabbits, and it almost invariably ends in death. It has not been recognised, hitherto, in horses. There are very few of these animals in the Federated Malay States and none of the people, whom we have seen suffering from melioidosis, had anything to do with them. Nevertheless, constrained by the close resemblance of melioidosis and glanders, we attempted to infect horses, both Arabs and country-bred ponies, by the subcutaneous, intravenous and intranasal inoculation of virulent, freshly-isolated cultures of *B. whitmori*. A small abscess appeared at the site of inoculation, when the virus was administered subcutaneously, but it soon healed; in no instance was there any evidence of generalization, and we concluded (1925) that "horses are apparently immune to infection with melioidosis." In this we were wrong; though they may be highly resistant they are certainly not altogether immune.

On February 26, 1925, Mr Macgregor, the government veterinary surgeon, brought some thick, yellow, blood-streaked pus to the laboratory. This pus had been coughed up by a valuable race-horse, named Elkins, which had been imported from Australia six months before. Mr Macgregor's attention had been drawn to the horse because it was ailing and had lost condition.

The specimen of pus was such a menagerie of organisms that we thought it must have been collected from the stable floor by the syce, and this was exactly what had happened. Media, inoculated with the pus, became hopelessly overgrown with moulds; but a guinea-pig, into which a little was injected subcutaneously, died three days later with a localised necrosis at the site of

the puncture, an enlarged, granular spleen and subcutaneous congestion. *B. whitmori* was cultivated from the heart, spleen and lungs.

The horse became no worse, and for several days no more pus could be obtained; but, a week later, the veterinary inspector found some pus dripping from the horse's left nostril and from this we isolated *B. whitmori* once more.

Agglutination tests were carried out with the blood of eight horses from the racing-stables, including Elkins. The type strain (Ragaviah) of *B. whitmori* was agglutinated by Elkins's blood diluted 1 in 8000, but the blood of none of the other horses agglutinated it when diluted beyond 1 in 200. Elkins's agglutinins were therefore fifty times as strong as those in the blood of any of the healthy horses.

Mallein tests were applied with some old samples of mallein from the Lister Institute and, subsequently, with some from Java; but the results were negative. In the meantime it was very difficult to know what to do with the horse; the purulent discharge was intermittent and appeared only after long intervals during which the animal was probably not a source of infection, because *B. whitmori* could not be cultivated from its nose. It became no worse, but improved so much on a course of arsenic that, when it was examined by three veterinary surgeons on March 15, they could find nothing wrong with it, but agreed to isolate it for two months so that it might be kept under observation. At the end of this period the horse seemed to be in perfect health and it was sent to a race-meeting. It did not run well, and when it was brought back to Kuala Lumpur it looked ill and, occasionally, there was a slight purulent discharge from the nose. On the ninth and again on the twentieth and the twenty-third of June, specimens of this discharge were sent to the laboratory and on each occasion *B. whitmori* was isolated. Subsequently the horse improved, with rest, but it never got into really good condition. In September, 1926, it was sold by its owner to a medical practitioner in Kuala Lumpur, who had it shot as soon as he learned its history. To our surprise we were not able to find any active lesions in the body. There was an old fibrous scar at the lower edge of the right lung in front and there were some enlarged and necrotic glands at the hilum; but the nose, the Schneiderian membrane and the accessory sinuses showed no trace of disease. Cultures were made from the glands, blood and various organs, but *B. whitmori* was not isolated. Guinea-pigs were inoculated with the blood and with material from the lung, spleen and glands, but they remained healthy. The horse's blood still agglutinated the homologous culture "Elkins" and the type strain "Ragaviah" in high dilutions.

The strain of *B. whitmori*, which was isolated from this horse, had the usual attributes of the species; it stained in bipolar fashion with Leishman's stain, it was motile, it produced a characteristic corrugated growth on glycerine agar and a pellicle on the surface of liquid media. It digested gelatine at 37° C. within two days; it produced acid in lactose, saccharose, glucose and mannite, and, after four days, a very little acid in dulcite.

A saline emulsion of the organism was agglutinated at 1 in 15,000, which

was the full titre, by an immune serum prepared with the type strain (Ragaviah) of *B. whitmori*. A rabbit, immunised by four intravenous injections of the culture from the horse, gave a serum which agglutinated this horse-strain in a dilution of 1 in 8000 and also agglutinated the type culture at the same titre. The serological identity of the two strains was confirmed by cross-absorption tests.

Two guinea-pigs, *A* and *B*, were inoculated with the second sample of pus. Guinea-pig *A* was given an injection under the skin. It died on the fourteenth day with large suppurating buboes in the groins, an enlarged granular spleen and one tubercle in the lung. *B. whitmori* was recovered from the buboes and from the spleen. Guinea-pig *B* was inoculated by the introduction of a platinum loopful of pus into the right nostril. It died on the twenty-fifth day with the nasal orifices blocked by dried discharge and with two abscesses in its neck, each one an inch in diameter. *B. whitmori* was cultivated from the thick pus in these buboes.

The effect of the culture on rats was tested, because of the importance of these animals as the probable disseminators of the disease. One rat (*M. griseiventer*) was inoculated subcutaneously with the fifteenth part of an agar slope. It died in four days with miliary tubercles in the lungs and spleen from both of which *B. whitmori* was recovered. A second rat was given a feed of bread soaked in an emulsion of the organism. It died ten days later, with one nodule in the spleen and two or three in the liver; *B. whitmori* was cultivated from both these organs.

SUMMARY.

A race-horse, which had been imported into the Malay States from Australia, got out of condition and occasionally coughed up thick pus. Five specimens of pus were brought to the laboratory between February and July, 1925, and on each occasion *B. whitmori* was isolated from it. The blood of the horse agglutinated the standard strain of *B. whitmori* in high dilutions. The morphological characters, and the biological reactions of the organism isolated from the horse were the same as those of the type strain (Ragaviah) of *B. whitmori*; and cross-agglutination and cross-absorption tests showed that they were identical. The strain was highly pathogenic for guinea-pigs and rats. The horse was killed in September, 1926, eighteen months after *B. whitmori* was first isolated from the nasal pus. No active lesions were found at autopsy and *B. whitmori* was not isolated from the organs.

REFERENCES.

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(*MS. received for publication* 18. x. 1926—Ed.)