

THE INFECTION OF MULES BY *TRYPANOSOMA HIPPICUM* THROUGH MUCOUS MEMBRANES.*

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The importance of ascertaining all the natural modes of infection in any trypanosome disease is evident. In murrina, the trypanosome disease of horses of Panama, it was not possible to incriminate tabanidæ or stomoxys—the only biting flies taken in or near corrals—as carriers of infection, for no evidence of trypanosomes nor of other flagellates could be detected in dissected specimens that had fed on infected mules.

The disease, which has been eradicated in the Canal Zone, is still present in parts of the Republic, in villages and along the trails, and cases have been detected in native ponies near the Zone line. In those localities where biting flies other than the ones examined exist in numbers, it may be that the trypanosome has a true succtorial intermediary host, but the inaccessibility and sparseness of the native population render difficult at the present time an investigation of this part of the question.

One other mode of infection, impossible to investigate directly in the corrals, required looking into; namely, the possibility of infection during coition. This was tested experimentally by noting whether *Trypanosoma hippicum* could pass through the intact mucous membranes of mules.

Koch was of the opinion that sleeping sickness might be transmitted during coitus, for he noted the presence of sleeping sickness in a number of women whose husbands either had died of the disease or were suffering from it at the time.

Hindle¹ has succeeded in infecting rats with *Trypanosoma gambi-*

* Received for publication, January 25, 1912.

¹ Hindle, *Parasitology*, 1911, iv, 24.

ense through the mucous membranes of the mouth and vagina and through the undamaged skin.

Martin and Ringenbach² also succeeded in infecting guinea pigs through the vagina with *Trypanosoma gambiense*.

In my experiments with *Trypanosoma hippicum*, two mules³ and an avirulent strain of trypanosomes were used. The average incubation period of this strain was ten days, *i. e.*, two or three days longer than that of the ordinary strain. The avirulent strain was purposely employed, so as to furnish additional evidence that the infection was acquired by the experiments, which were carried out in a screened stable practically free from flies.

Using an all glass syringe with a smooth nozzle which could not abrade the vaginal mucosa, about ten cubic centimeters of citrated saline solution containing about 0.5 of a cubic centimeter of guinea pig blood richly infected with *Trypanosoma hippicum*, were introduced into the vagina of mule 193. Most of the fluid was ejected upon the ground, but did not come in contact with the skin or any abraded surface on the legs of the animal. On the tenth day, the animal's temperature, which had been uniformly 101° F. morning and evening, rose to 102° F., but trypanosomes could not be detected, though there was some auto-agglutination of erythrocytes. The appearance of this in other experimental mules I had observed just before trypanosomes could be demonstrated in films. On the morning of the eleventh day, the temperature was 102.6° F., and trypanosomes were detected in films for the first time. The following morning the animal's temperature was 103.6° F.

In the other mule, trypanosomes were introduced into the mouth and nasal cavity. Ten cubic centimeters of citrated saline suspension containing 0.5 of a cubic centimeter of guinea pig blood richly

² Martin and Ringenbach, *Bull. Soc. path. exot.*, 1910, iii, 433.

³ In a previous experiment, I attempted to infect three mules with *Trypanosoma hippicum* by means of *Musca domestica* that had fed on infected guinea pig blood. After an incubation period of ten days, one mule became infected. The remaining two were kept under observation for thirty days in a screened corral, daily examinations of the temperature and blood being made. As during this time there was no rise of temperature, no appearance of trypanosomes, and no development of auto-agglutinins in the blood, it was assumed that they had not been infected. They were, therefore, employed in the experiments here recorded.

infected with *Trypanosoma hippicum* were divided into two portions. By means of a syringe, five cubic centimeters were introduced into the mouth, and five cubic centimeters into the right nasal cavity, the muzzle of the mule being elevated so as to avoid waste of the infected fluid. The mucosa was not touched by the nozzle of the syringe. On the ninth day, the animal's temperature rose from 101° to 102.8° F., and trypanosomes were detected in blood films.

These two experiments are of significance because they indicate positively that *Trypanosoma hippicum* can penetrate the mucosa of mules, which in the mouth and vagina is much thicker than that of guinea pigs and rats. From this it is assumed that murrina may be transmitted during copulation.