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Received October 8, 1951. P.S.E.B.M., 1951, v78.

### Effect of Rabies Street Virus in the Cotton Rat and the Swiss Albino Mouse. (19187)

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The rabies virus strain V 308 used in the present study was isolated in Swiss albino mice (21-day-old Webster Carworth strain) from a dog brain sent to this laboratory for a routine examination for rabies. The history of this strain is given in a previous paper which dealt with electron microscope studies of Negri bodies(1). The strain was pathogenic for 2 species of bats(2). Furious rabies was produced in the Syrian hamster by various routes of inoculation with this strain (3).

**Methods.** Each of four 3-week-old cotton rats (*Sigmodon hispidus*) was inoculated intracerebrally with 0.03 cc of a 10% virus-bearing brain suspension of the 2nd mouse passage which titrated  $10^{-4}$  in mice. Five days after injection the cotton rats showed tremors and paralysis, but no evidence of furious rabies was noted. The brains of 2 paralyzed cotton rats were removed aseptically. Touch preparations from the Ammon's horns of these brains were stained with Seller's stain(4) and upon examination under an optical microscope were found to contain Negri bodies. A 10% suspension in physiological saline was made from the 2 brains. This material was used to initiate the present study in cotton rats and Swiss albino mice. By intracerebral inoculation it titrated  $10^{-4.5}$

in cotton rats and  $10^{-4.2}$  in mice. Diagnosis of rabies was made on the following factors: presence of symptoms of rabies in the rat or mouse and demonstration of Negri bodies in the Ammon's horn of the brain of the animal showing symptoms. The presence of Negri bodies was determined by staining touch preparations of the Ammon's horn with Sellers' stain and examining the slides with the optical microscope. Twenty-seven healthy cotton rats (21 days old) were divided into 9 groups of 3 animals each. The 3 rats of each group were administered the virus by one of the following methods: intracerebral, intraperitoneal, intradermal, intramuscular, intralingual, and intracardial injection and by oral, rectal, and intranasal instillation. In all routes of exposure the inoculum consisted of 0.1 cc of the above described suspension, except in the intracerebral group where the inoculum was 0.03 cc. Twenty-seven healthy Swiss mice of the same age were divided into 9 groups of 3 each and were treated in a like manner. After inoculation all groups were observed daily for symptoms of rabies. After symptoms of rabies appeared and the animal became paralyzed or moribund, it was sacrificed and the brain removed and examined for the presence of Negri bodies. Some animals died during the night and the brains of these

TABLE I. Response of Cotton Rats and Swiss Mice to Rabies Virus. All showed symptoms.\*

| Min-max incubation period† (in days) |            |
|--------------------------------------|------------|
| Cotton rats                          | Swiss mice |
| 7-8                                  | 9-10       |
| 7-8                                  | 7-8        |
| 9-11                                 | 7-8        |
| 7-8                                  | 7-8        |
| 12-14                                | 10-11      |
| 8-11                                 | 7-8        |
| 5-6                                  | 6-7        |
| 5-10                                 | 8-11       |
| —                                    | 14         |

\* Except cotton rats by intranasal inoculation.

† Incubation period—time between exposure to virus and onset of symptoms.

dead rats or mice were also examined for Negri bodies.

**Results and discussion.** Table I shows the response of cotton rats and Swiss mice injected with rabies virus by different routes of exposure. All rats and mice exposed, except the cotton rat group exposed by intranasal instillation, died from the disease or were sacrificed when symptoms appeared.

The intranasal group of rats was held 30 days without showing symptoms. They were then sacrificed and touch preparations of their brains examined. No Negri bodies were seen.

No cotton rats or Swiss mice showed symptoms of furious rabies as found in Syrian hamsters infected with this strain. The incubation period in rats and mice was between 5 and 14 days by these various routes of exposure. The cotton rats were as susceptible as the Swiss mice to this V 308 strain of rabies

virus. This strain of mice was the one recommended by Webster as being especially susceptible to neurotropic viruses(5).

**Summary.** A strain of rabies isolated from a dog brain and passaged intracerebrally 2 times in Swiss albino mice and one time in cotton rats has been successfully transmitted to the same rodents by the following routes of exposure: intracerebral, intraperitoneal, intradermal, intramuscular, intracardial, intralingual, oral and rectal. The cotton rats and Swiss mice were equally responsive to this strain by the above routes of exposure. Mice were susceptible to intranasal instillation but cotton rats exposed by this method remained symptom free during a 30-day observation period and there was no evidence of Negri bodies when their brains were examined at the end of that time. This strain of virus, which had produced furious rabies in Syrian hamsters in a previous experiment, produced the dumb form of rabies in the cotton rat and Swiss albino mice.

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Received October 18, 1951. P.S.E.B.M., 1951, v78.

### An Adsorption Technique for Partial Purification of Japanese Encephalitis Virus in Chick-Embryo Tissue Suspensions. (19188)

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Crude suspensions of chick-embryo tissues infected with the virus of Japanese encephalitis contain a miscellany of tissue materials differing widely in particle size, composition, and physical properties. As a preliminary step toward the isolation of the virus it would

be desirable to utilize some simple method to remove most of the extraneous material, leaving a partially purified and considerably more uniform suspension of unimpaired infectivity.

A certain amount of the extraneous material can be removed by differential centrifugation.