# Recovery of Rabies Virus From Colonial Bats In Texas

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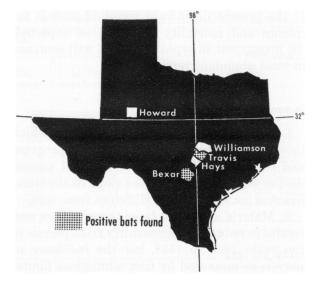
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THE ISOLATION of the rabies virus from a leaf-nosed bat taken in Blumenau, Brazil, was first reported by Haupt and Rehaag (1) in 1921.

Hurst and Pawan (2-4) demonstrated the natural infection of the vampire bat (Desmodus rotundus) with rabies virus and observed that these bats were capable of transmitting the agent for several months as symptomless carriers. Pawan (5) showed that fruit-eating bats of the genus Artibeus have also survived for months as asymptomatic carriers, even though their saliva became infectious. Johnson (6) recorded that he and Ten Broeck recovered the rabies virus from the vampire bat (D. rotundus) in Mexico. Málaga-Alba (7) later found rabid vampire bats in Sonora, Chihuahua, and Tamaulipas, three states in Mexico which border the United States.

The existence of rabies in bats in the United States was suspected in December 1951 when Dr. J. E. Hogan of Big Spring, Tex. (Howard County), notified the Texas State Health Department about a woman who had become ill approximately 3 weeks after being bitten by a

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bat she had picked up by the roadside. The bat was not tested for rabies or specifically identified. Sulkin and Greve (8) reported that the patient had symptoms suggestive of bulbar poliomyelitis after becoming ill, and she died shortly afterward in a Dallas hospital. On autopsy, Negri bodies were discovered in brain material.

The first findings of rabies in bats of the United States came from the Florida State Board of Health Regional Laboratory at Tampa (9) in June 1953 when Negri bodies were found in the brain of a Florida yellow bat (Dasypterus floridanus) which had attacked a child. Later, the rabies virus was recovered from brains of 5 additional Florida yellow bats and from 1 Seminole bat (Lasiurus seminolus). These 2 species are noncolonial, insectivorous, and indigenous to the southeastern United States.

Several weeks after the Florida report, the rabies virus was recovered from the brain of a bat (*Lasiurus cinereus*) which without provocation had attacked a woman near Carlisle, Pa. (10).

The present report concerns the recovery of the rabies virus from colonial, insectivorous bats collected in the central and south central parts of Texas. Two hundred bats were taken in Howard, Bexar, Hays, Travis, and Williamson Counties, Tex., in November and December 1953. They included 151 Mexican free-tailed bats (Tadarida mexicana), 42 cave myotis (Myotis velifer), and 7 pipistrels (Pipistrellus subflavus). Taxonomic assistance in identifying the bats was provided by Dr. W. Frank Blair, mammalogist, University of Texas.

# Methods of Detecting the Virus

All bats collected were individually numbered. In the laboratory, their brains were removed aseptically, and impression smears were prepared and examined for the presence of Negri bodies.

The brains were prepared for inoculation into mice immediately or preserved in a 50-percent glycerin-saline solution for processing as time permitted. The brains were macerated in sterile mortars, suspended in a broth containing penicillin (1,000 units per cc.) and streptomycin (250 units per cc.), and eluted in a cold vault for at least 2 hours. The glycerinated brains were washed in a sterile saline solution prior to being treated as the fresh brains.

The supernatant fluid from each brain suspension pool was then inoculated into white mice by the intracerebral and intraperitoneal routes. Each pool was prepared from the brains of 1 to 5 bats of the same species, with a single exception.

### **Findings**

A rabieslike agent pathogenic for mice was obtained by inoculating 4 mice with brain tissue removed from 1 T. mexicana and from 1 M. velifer (inadvertently mixed). Both of these bats were taken at Camp Bullis in Bexar County, Tex., in November 1953. The T. mexicana was taken from an officer's quarters at the camp, and the M. velifer from a bat retreat known as head-quarters cave. Seven days after inoculation, 1 mouse showed symptoms of rabies, and 3 mice were dead. The brains of all 4 mice were found to contain Negri bodies.

The brain taken from the ill mouse was found to be bacteriologically sterile, and its passage resulted in the death of 1 of 6 mice and abnormal behavior in the remaining 5 on the fifth day. Negri bodies were found in the brains of these mice. Aerobic and anaerobic cultures for

bacteria were negative. The agent was infectious for mice when injected into the peritoneal cavity, but the incubation period was somewhat prolonged.

The identity of the agent was further established by means of the serum-virus neutralization test. The antiserum had been prepared in rabbits against a strain of fixed rabies virus.

Confirmation of the source of the rabies virus was obtained by a second isolation from the original bat brain pool. It was unfortunate that the brains of the two different species of bats were mixed since it then became questionable which species, if not both, was infected. Examination of the smears of the original bat brains revealed Negri bodies in the brain of the T. mexicana but not in the brain of the M. velifer.

A similar pathogenic agent was obtained from the brain of a T. mexicana taken inside a trailer shed on the premises of the Texas State Health Department in Austin (Travis County) on December 4, 1953. The bat was found in a torpid condition on a wall near the ground, whereas other bats in the area were active. On the seventh postinoculation day, all mice inoculated with the bacteriologically sterile brain suspension from this bat were paralyzed. On the eighth day, 1 mouse was dead, and the remainder were sacrificed. Negri bodies were demonstrated in brain smears from these mice as well as in a second series of mice inoculated with a brain suspension from the dead mouse.

A second recovery of the virus from the original bat brain suspension verified the origin of the infectious agent. Confirmatory evidence of the identity of the virus was afforded by its neutralization with fixed virus antirabies serum.

#### **Discussion**

The behavior of the canine rabies virus in insectivorous bats has been studied in a limited way by Reagan and Brueckner (11) who successfully inoculated two species (*Eptesicus fuscus*, *Myotis lucifugus*). Several passages were made in the big brown bat, *E. fuscus*.

The significance of the finding of rabies virus in the common Mexican free-tailed bat (*T. mexicana*) is unknown. Since the two infected

bats were taken in unnatural locations, the possibility exists that the animals ill with rabies leave their colonies and seek solitude. If true, this would lessen the chances for spread of the infection. These are, of course, preliminary observations.

The present information on rabies in bats in the United States is too limited to formulate specific control measures. Certainly the destruction or abatement of the *T. mexicana* is not justified, but it does appear that the public should be warned relative to the dangers inherent in the handling of sick bats.

These insectivorous bats are present in large numbers in caves and dwellings in the southwest and are known to migrate into Mexico. Sanborn (12) has reported the recovery of a banded specimen in Mexico which had flown 800 miles from the Carlsbad Caverns in New Mexico.

Further studies on the percentage and species of infected bats in Texas and their importance in the epizootiology of rabies are now being conducted by the Texas State Department of Health.

## Summary

Two strains of rabies were recovered from the brains of colonial bats in Texas. One strain was obtained from pooled brain tissue comprised of 1 Tadarida mexicana, which contained Negri bodies, and 1 Myotis velifer, which contained no Negri bodies. The second strain of rabies virus was isolated from the brain tissue of a single T. mexicana. These were among a total of 200 colonial, insectivorous bats collected and tested for rabies by the Texas State Department of Health in November and December 1953. The following species were

included: 151 T. mexicana, 42 M. velifer, and 7 Pipistrellus subflavus.

This is the first reported recovery of rabies virus from naturally infected colonial bats in the United States.

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# Key to Bats of Western North America

A key to the bats of western North America (north of Mexico) will aid in identifying 27 species of bats which are now known to be from North America north of the Mexican border and west of the 110th meridian. The key was prepared by Philip H. Krutzsch of the Museum of Natural History, University of Kansas, and is included as No. 133 in the series of *Natural History Miscellanea* published by the Chicago Academy of Sciences, 2001 N. Clark Street, Chicago, Ill.