

Development of rabies in native carnivores kept in bat-proof and arthropod-proof cages in a bat cave indicates bat-to-carnivore transmission by a nonbite route.

Rabies Transmission by Nonbite Route

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TWO PERSONS who entered Frio Cave, a large limestone cavern near Uvalde, Tex., where rabid bats had been identified, subsequently died of laboratory-confirmed rabies (1,2). One death occurred in 1956, the other in 1958. Both men were reliable observers and early in their illness, while still conscious and alert, both denied knowledge of being bitten by bats or other mammals. These observations suggested the possibility of some nonbite mode of transmission of rabies, at least under the atmospheric conditions in Frio Cave or caves similarly inhabited by millions of bats.

In 1960 and 1961, three experiments were undertaken to test this hypothesis. Several varieties of wild carnivorous animals, including foxes, coyotes, opossums, raccoons, ringtails (cacomistles), and skunks, as well as domestic dogs and cats, were exposed in cages for periods of 7 to 30 days. Special efforts were made, particularly in the third experiment, to exclude all possibility of exposure to the bites of wild animals or of arthropods.

In the first experiment, beginning July 8, 1960, 13 carnivores (2 domestic dogs, 2 domestic cats, 2 raccoons, 2 ringtails, 2 coyotes, 2 gray foxes, and 1 striped skunk), each confined separately in metal 1- by 2-inch mesh cages, were placed in a bat-occupied room within Frio Cave. Previously the animals had been caged in isolation from 9 to 21 months, presumably ruling out incubation of the disease prior to exposure in the cave. Serums of all the animals collected before the experiment were negative for rabies

antibody by the standard serum neutralization test.

The cages were arranged on racks 3 feet above the cave floor. The legs of the racks had smooth metal guards to prevent ascent of dermestid beetle larvae, which littered the cave floor. Within the cages the animals were exposed to contact with the cave atmosphere, bat excreta, various bat ectoparasites, and other arthropods, such as adult dermestids, and there is little doubt that such contact occurred. In addition, it is possible that feral animals, such as carnivores and bats, either could have bitten caged animals through the cage mesh, or small species could have entered the cages through the mesh.

On July 15, 1960, after 7 days, the carnivores were removed from the cave and placed under observation at our station. Of the 13 animals 1 fox, 2 coyotes, and 1 ringtail died of rabies. Incubation periods ranged from 31 to 113 days. Rabies virus was isolated and confirmed by serum-virus neutralization tests.

This experiment was followed by a second week-long test, beginning September 7, 1960. Four each of the following species were placed in the cave: coyotes, gray foxes, striped skunks, ringtails, raccoons, domestic dogs, and domestic cats. The animals were divided into two groups of equal composition. The cages of one group were protected behind ¼-inch metal mesh to prevent contact with bats and other vertebrates. The cages of the other group were protected behind mosquito netting to provide assurance against contact with large arthropods.

Although these animals were held in the same cave room as those in the July experiment, the bats vacated the room as the animals were

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moved in and did not return, although they could be seen in other rooms of the cave. Bat ectoparasites and other arthropods, however, remained in the room throughout the 7-day period. None of the carnivores in this experiment developed rabies.

In 1961 another collection of animals was exposed in the same cave room from June 28 to July 29, during the time suckling and lactating female bats were present. The animals were arranged in separate groups designated 1-4, each group differing from the others in the degree of protection provided against the cave fauna.

Group 1 animals (two coyotes and two gray foxes) were housed separately in escape-proof 1-inch mesh cages and provided no other protection. Thus, they had no assurance against contact with bats, other vertebrates, arthropods, bat excreta, or the cave atmosphere.

Group 2 animals were also housed separately in cages kept within three larger boxlike 1/4-inch metal mesh enclosures and separated from the mesh on all sides by at least 6 inches. These animals were protected against bats and other vertebrates, but they were exposed to arthropods, bat excreta, and the cave atmosphere. Animals comprising this group were two each of coyotes, gray foxes, silver foxes, striped skunks, spotted skunks, raccoons, ringtails, domestic dogs, domestic cats, and opossums.

Cages of group 3 animals were similarly enclosed using 1/18-inch plastic mesh in place of the 1/4-inch metal mesh. The animals were provided feed which had been sealed and stored within the enclosure, and they were watered by means of a closed-hose system supplied from a tank outside the cave. Animal care was achieved by means of sleeves, built into the enclosure, and into which the animal caretaker could insert his arms to work. These animals were thus protected against bats, other vertebrates, and certain large arthropods, but they were exposed to smaller arthropods, bat excreta, and the cave atmosphere. The group included two each of coyotes, ringtails, and gray foxes.

Group 4 animal cages were kept within two elongated boxlike enclosures, A and B, carefully sealed to exclude air except at the opposite ends, which were open except for 1/26-inch dacron mesh, reinforced with 1/18-inch plastic

mesh. These enclosures were designed to expose animals only to air, which penetrated the enclosure by drifting horizontally from one mesh-covered end to the other. The mesh would prevent entry of large arthropods. In an attempt to insure against exposure to tiny crawling arthropods, such as bat mites, a moat was constructed at each end, just inside the mesh. The moat consisted of a continuous band of glycerine-soaked spun glass padding, 1-inch thick and 12 inches wide, stapled in a continuous channel extending across the floor, up one wall, across the top of the enclosure, and down the other wall. Feed was stored within the enclosures before they were sealed and taken into the cave. Water was available through a closed-hose system, as for group 3 animals, and animal care was accomplished through built-in sleeves. The animals were visible through transparent plastic wall panels and ceiling. Animals included in this group were four each of coyotes, gray foxes, and ringtails. They were housed in individual cages and two of each species were equally distributed in the two enclosures, A and B.

A few mishaps occurred. A number of early deaths in ringtail and opossum groups were apparently the result of the extremely oppressive cave room atmosphere, which was hot, humid, and heavily charged with ammonia. On one occasion the legs supporting one of the three group 2 enclosures were found to have

Rabies status of coyotes and foxes in Frio Cave in four types of cage enclosures

Cage group and number and type of animal	Number of rabies deaths	Days of exposure
<i>Group 1</i>		
2 coyotes-----	2	27
2 gray foxes-----	2	27
<i>Group 2</i>		
2 coyotes-----	2	27
2 gray foxes-----	2	27
2 silver foxes-----	2	27
<i>Group 3</i>		
2 coyotes-----	2	27
2 gray foxes-----	2	27
<i>Group 4A</i>		
2 coyotes-----	2	24
2 gray foxes-----	2	24
<i>Group 4B</i>		
2 coyotes-----	2	30
2 gray foxes-----	2	30

collapsed, and the ends of the enclosure were opened wide enough for bats to squeeze inside, were they so inclined. After the enclosure of group 4B was removed from the cave, sprayed with insecticide, and opened, dermestid beetles were found breeding in the animal feed and feces, but no other arthropods could be detected. One or more dermestids may have been sealed in the enclosure before it was taken into the cave, but we must assume that contamination may have occurred in the cave. A careful search of the enclosure of group 4A indicated its interior was devoid of arthropods.

Animal groups 1, 2, and 3 were in the cave 27 days. Group 4A was kept in the cave 24 days; group 4B was removed after 30 days.

During the 6-month period of observation following their removal from the cave, 22 animals developed rabies. These were all 10 coyotes and all 12 foxes in cage groups 1 through 4 (see table). In each instance the virus was isolated and identified by the serum neutralization test.

Incubation periods, counted from the first day of exposure in the cave, ranged from 28 to 109 days. Prior to placement in the cave, all but two animals had been caged in isolation from 6 to 20 months. Exceptions were a coyote in group 1 and a fox in group 4 which were trapped only 4 months previously. The silver foxes had been born in captivity 3 years previously.

These findings support consideration of an airborne medium, such as an aerosol, as the mechanism of rabies transmission in this instance. Further studies are in progress.

REFERENCES

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