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SURVEY OF RABIES AMONG FREE-FLYING BATS FROM THE BIG BEND REGION OF TEXAS

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In 1951, a 43 year old Texas woman contracted rabies from a bat bite. This was the first reported human case of rabies involving a bat (Sulkin and Greve, 1954; Brass 1994; Clark, 1994). Since then, bats have been implicated as a major reservoir for the rabies virus, especially in the Americas (Constantine, 1988). Understanding the prevalence and distribution of enzootic rabies in bats is fundamental in understanding the importance of bats in the epidemiology of rabies (Brass, 1994).

Because of their association with rabies, thousands of bats are examined annually by public health agencies for the presence of the virus. Brass (1994) reported that, on average, about 4% to 10% of bats submitted to diagnostic laboratories test positive for the rabies virus. Constantine (1988) estimated that in the United States, about 10 % consistently test positive annually. In Texas, 11% of 1,758 bats tested by the department of health between 1984 and 1987 proved to be infected (Schmidly, 1991), whereas 14 % of 2,931 bats tested between 1990 and 1994 were rabid (Anonymous, 1990; 1991; 1992; 1993; 1994).

Although these figures suggest that a considerable proportion of the bat population is afflicted with

rabies, they are misleading. Constantine (1988), Schmidly (1991), and Brass (1994) all recognized a bias in the sampling techniques of bats submitted to public-health facilities. These often were sick or dead bats found near human dwellings (Schmidly, 1991; Brass, 1994). Such bats are considered more likely to be infected with rabies than those selected from a random sample of the population (Brass, 1994). Also, if surveyed bats are collected from typical roost sites, rabid bats are taken more easily than healthy bats (Constantine, 1988). Therefore, a more reliable and useful survey of the incidence of rabies among bats requires the sampling of bats capable of flight (Constantine, 1988).

The Big Bend area is of particular interest because of its exceptional abundance and diversity of bats (Easterla, 1973; Schmidly, 1991). Several accounts on the natural history of bats in the Big Bend area have been presented (Baker, 1956; Anderson, 1972; Easterla, 1973; Schmidly, 1977), however, data on the incidence of rabies are lacking. The objective of this study was to examine the incidence of rabies among free-flying bats from the Big Bend region of Texas, and to compare the results with figures reported statewide by the Texas Department of Health.

MATERIALS AND METHODS

All bats assayed in this study were acquired from within Big Bend Ranch State Park (BBRSP), situated in southwestern Texas just north of the Rio Grande in Brewster and Presidio counties. The park consists of over 275,000 acres of Chihuahuan Desert habitat, with more than 100 springs and several permanent streams. The numerous water-associated habitats of this area provide an excellent situation for the sampling of bats in their natural environment.

From May to August, 1994, bats were sampled using mist nets (Kunz and Kurta, 1988). At dusk, nets were deployed across springs, streams, or other small bodies of water. A total of 85 nets was set at 45 localities throughout the park. Bats captured were removed from the nets, identified, and sacrificed. They then were prepared as standard museum skins and skulls (DeBlase and Martin, 1981). These voucher materials are deposited in the collection of Recent mammals in the Natural Science Research Laboratory of the Museum of Texas Tech University.

During preparation, brain tissue for testing was removed from each cranium using a technique modified from Greenhall (1965). A 21 gauge hypodermic needle affixed to a 5 cc syringe with its plunger withdrawn was inserted into the foramen magnum of each bat skull. The plunger was then depressed, and the brain, or portion thereof, subsequently was expelled from the cranium through the foramen magnum via positive pressure. In Greenhall's (1965) technique, the brain was aspirated from the cranium. This new procedure provided adequate quantities of brain tissue for the rabies assay, and yet avoided damage to the skull, allowing it to be used for other studies.

Following expulsion, brain tissues temporarily were stored in liquid nitrogen until they could be deposited in an ultracold freezer for long-term storage. Samples of brain from the bats were tested for rabies by immunofluorescence as described by Dean and Ableseth (1973). All samples were scored as positive or negative for rabies. A 95 percent confidence interval for the resulting binomial parameter was calculated following Dowdy and Wearden (1991).

RESULTS AND DISCUSSION

Brain specimens from 171 bats were tested for the presence of the rabies virus. Species of bats tested (with number of each species tested in parentheses), all of which are known hosts of rabies in the United States (Constantine, 1988; Brass, 1994), were as follows: *Mormoops megalophylla* (42), *Myotis californicus* (8), *M. ciliolabrum* (1), *M. thysanodes* (1), *M. velifer* (5), *M. yumanensis* (1), *Eptesicus fuscus* (12), *Pipistrellus hesperus* (60), *P. subflavus* (1), *Plecotus townsendii* (4), *Antrozous pallidus* (27), and *Tadarida brasiliensis* (9). All 171 specimens tested negative for rabies.

Sample sizes for each individual species were not large enough for independent analysis, therefore the general bat population was treated as a whole. The proportion of positive bats in the entire sample of 171 bats was zero. Thus, assuming random sampling, a 95 percent confidence interval on the proportion of rabid bats at BBRSP is [0, 0.0174]. Alternatively stated, assuming random sampling and given 171 non-rabid bats out of a sample size of 171, the percentage of rabid bats in the population is $\leq 1.74\%$ ($p \leq 0.05$). Therefore, there is only a 5% chance that the true prevalence of rabies in the population was $>1.74\%$ (Fig. 1).

This frequency of rabies in bats at BBRSP (0% to 1.74%) is consistent with that reported in similar studies. Constantine (1988) estimated that $<0.1\%$ to 0.5% of bats capable of flight are infected with rabies, and Baer and Smith (1991), in a summary of four similar studies, reported frequencies of clinically-normal rabid bats at 0% to $<1\%$.

Undoubtedly, a large amount of the discrepancy between rabies prevalence in random or free-flying bats and that obtained from laboratory-submitted bats is due to the aforementioned bias in samples submitted to health departments. However, samples obtained from free-flying bats conceivably could be biased in the opposite direction. Bats ill with rabies at an advanced stage may lose their capacity to fly or behave abnormally, thus excluding themselves from possible capture (Constantine, 1988; Baer and Smith, 1991; Brass, 1994). Presumably, the actual prevalence of rabies in bats lies somewhere between figures derived from "random" sam-

Figure 1. Probability of finding all bats from a sample size of 171 to be negative for rabies, plotted against the true proportion of rabid bats in the population. There is a <5% chance of observing no positives if the true proportion of rabid bats is >1.74%.

pling of free-flying bats and those based on laboratory-submitted specimens, and it is essential that data from both sources be considered in order to understand the incidence of rabies in bats. Still, according to Brass (1994:142), "Sampling biases notwithstanding, 'random' collection of healthy-appearing individuals must serve as our first and best approximation of disease prevalence in natural populations."

The incidence of rabies at BBRSP during the summer of 1994 was well below the statewide levels reported by the Texas Department of Health over several years. However, the results of this study apply only to a small portion of Texas during a relatively short period of time. Cycles in sylvatic rabies are known to vary geographically and temporally, usually predominating in a single reservoir species (Brass, 1994). Hill and Smith (1984) suggested that the absence or low level of rabies in a bat population may indicate that bats are at stable numbers and not overstressed due to overcrowd-

ing and excessive use of food resources. If this is the situation at BBRSP, it is conceivable that if environmental conditions change, outbreaks of rabies within bat colonies could occur, thus changing the frequency of rabies in bats throughout the area. Because of the epizootic nature of rabies, and the abundance of bats and increase of visitor usage at BBRSP, as well as other public lands in the area, we suggest continued monitoring of the bat population in the region.

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It was through the efforts of Horn Professor J Knox Jones, as director of Academic Publications, that Texas Tech University initiated several publications series including the Occasional Papers of the Museum. This and future editions in the series are a memorial to his dedication to excellence in academic publications. Professor Jones enjoyed editing scientific publications and served the scientific community as an editor for the Journal of Mammalogy, Evolution, The Texas Journal of Science, Occasional Papers of the Museum, and Special Publications of the Museum. It is with special fondness that we remember Dr. J Knox Jones.

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