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Rabies in Hooded and Striped Skunks in Arizona

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ABSTRACT: Arizona is home to four species of skunks, and rabies is enzootic in the region in which their ranges overlap. Examination of state health data from 1985 to 2004 revealed an irregular 4–10 yr periodicity in the number of cases annually, which may be related to past precipitation patterns. The number of rabid skunks peaked during springtime. Locations of rabies epizootics changed over time, but there was no evidence of a large-scale geographic spread. Skunks live-trapped during 1996–2002 had a low prevalence of rabies-virus neutralizing antibodies. This study was the first to document rabies in hooded skunks (*Mephitis macroura*).

Key words: Arizona, hooded skunk, Mephitis macroura, Mephitis mephitis, rabies serology, striped skunk, radiotracking.

Rabies is an important zoonotic disease worldwide, killing some 55,000 people annually (WHO, 2006). In the United States, mandatory pet vaccination programs and, in some areas, an aggressive oral vaccination program for wildlife have greatly reduced human exposures and mortality. However, wildlife still represent the source for the majority of rabies exposures in the US (Krebs et al., 2005). Rabies in Arizona has been present since at least the turn of the last century (Parker, 1975) and has been associated with several bat rabies virus variants, a unique fox rabies virus variant, and the south-central skunk rabies virus variant (Krebs et al., 2005).

Striped skunks (*Mephitis mephitis*) are one of the most common mammals reported with rabies each year (Krebs et al., 2005). In Arizona, this species exists sympatrically with three additional skunk species (Hoffmeister, 1986): hooded skunks (*Mephitis macroura*), western spotted skunks (*Spilogale gracilis*), and hognosed skunks (*Conepatus leuconotus* [= *C. mesoleucus*; Dragoo et al., 2003]). All four species are present in the southeastern portion of Arizona (Hoffmeister, 1986), which coincides with the area where the south-central skunk rabies virus variant is found (Krebs et al., 2005). Because state health laboratories' records often do not provide species information on "skunk" rabies cases, data on the occurrence of rabies in individual skunk species often are unavailable (Dragoo et al., 2004).

The objectives of this study were to document the spatiotemporal occurrence of rabies in skunks submitted for testing from Arizona during 1985–2004 and to report the incidence of rabies and the prevalence of rabies antibodies in the four species of skunks that were captured and tested during a study of mesocarnivore population ecology from 1996 to 2002.

Rabies case data for skunks tested from 1985 to 2004 were provided by the Arizona Department of Health Services (AZDHS). These submissions were from animals associated with known or suspected human exposures, as well as nuisance skunks captured as part of routine rabies surveillance at Fort Huachuca Military Reservation and the nearby city of Sierra Vista. All animals were tested by AZDHS, and samples from positive animals were submitted to the Texas state laboratory, or Centers for Disease Control and Prevention (CDC), Atlanta, GA, USA, for rabies virus typing either by direct fluorescent antibody (DFA) testing or by rapid fluorescent focus inhibition test (RFFIT). Data from 1985 to 2000 included information on individual skunks as well as date submitted and location where collected. Data for 2001-2004 consisted of annual summaries and included the number of rabid skunks by county. Reliable

species identifications were not available with these data.

All four species of skunks were captured during studies of white-nosed coatis (Nasua narica) during 1996-2000 (Hass and Roback, 2000; Hass and Valenzuela, 2002) and hooded and striped skunks during 2000-2002. Study areas included Fort Huachuca Military Reservation, 120 km southeast of Tucson, Arizona (31.55°N-110.35°E, 1515 m elev.); Audubon Appleton-Whitell Research Ranch (ARR), 3 km west of Fort Huachuca; Las Cienegas National Conservation Area, 8 km north of ARR; and San Pedro Riparian National Conservation Area, located along the eastern boundaries of Fort Huachuca and Sierra Vista. Skunks were captured in Tomahawk live traps (models 204 and 207, Tomahawk Live Trap Co., Tomahawk, Wisconsin, USA) and were anesthetized with ketamine hydrochloride (Fort Dodge Animal Health, Fort Dodge, Iowa, USA) and xylazine hydrochloride (Lloyd Laboratories, Shenandoah, Iowa, USA). A 5:1 mixture was initially used, but anesthetized skunks often exhibited bradycardia; skunks appeared to better tolerate a 10:1 mixture. Sedated animals were removed from traps and weighed, and standard morphometrics were taken. When possible, 0.5-1.0 ml of blood was collected via jugular venipuncture using a 25-ga needle and 3-cc syringe; collected sera were tested for rabies virus-neutralizing antibodies. An aluminum eartag (size 3, National Band & Tag Co., Newport, Kentucky, USA) was attached to one or both ears. Some adults during 2001–2002 were fitted with VHS radio collars (Holohil System Ltd, Ontario, Canada; Model 2AM; 35 g). Radiocollared animals were tracked to their dens at least weekly. Trapping and handling protocols were designed to minimize stress to captured animals (American Society of Mammalogists, 1998).

As part of a trap-vaccinate-relocate trial, nine skunks captured in the housing area at Fort Huachuca were vaccinated (IM with 1ml of Imrab3; Merial Limited, Athens, Georgia, USA) and translocated to a new location 8.5 km away. All translocated skunks were fitted with radiocollars. Translocated skunks were monitored daily for the first 2 weeks after release, then approximately every other day until they died, the signal was lost, or 30 June 2002, when the study ended.

To examine the relationship between precipitation and rabies in skunks, monthly precipitation data were gathered from four sites in southeastern Arizona for the period 1980–2004: Sierra Vista (31.55°N, -110.28° E, 1515 m elev.); Nogales $(31.35^{\circ}N, -110.93^{\circ}E, 1175 \text{ m elev.});$ Duncan (32.72°N, -109.10°E, 1104 m elev.); and Tucson (32.25°N, -110.92°E, 725 m elev.). These sites had the most rabid skunks reported during 1985-2000. Data were acquired from the Western Regional Data Center (www.wrcc.dri.edu/ climsum.html; accessed 1 August 2005). Monthly precipitation totals were averaged over the four sites and subject to ordinary least-squares regression with natural logarithm-transformed skunk cases as the dependent variable. All statistical tests were conducted using Stata 8.0 (StataCorp, 2003).

Between January 1985 and December 2004, 506 rabid skunks were identified at Arizona state health laboratories. The highest numbers were recorded during 1985–1987 and 2001 (Fig. 1). The average number of rabid skunks varied monthly, peaking during January and April (Fig. 1). With few exceptions, rabid skunk cases were limited to southeastern Arizona where the ranges for all four skunk species overlap (Fig. 2). Distribution of cases within this area varied between years, and all rabid skunks were infected with the south-central skunk rabies virus variant. Outside of this area, few rabid skunks have been reported. One rabid skunk was reported in Sedona during 1985, and 19 cases were reported in Flagstaff between January and May 2001. The 2001 cases from Flagstaff were all associated with the

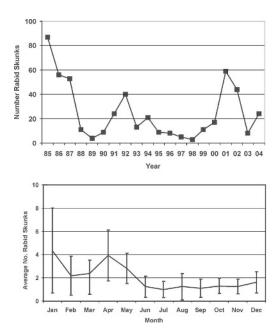


FIGURE 1. Annual totals (top) and monthly averages (bottom) of skunks testing positive for rabies by the Arizona Department of Health Services, 1985–2004. Submissions include potential exposures, and animals captured during routine surveillance in Cochise County. Ninety-five percent confidence intervals calculated by sampling with replacement 1,000 times.

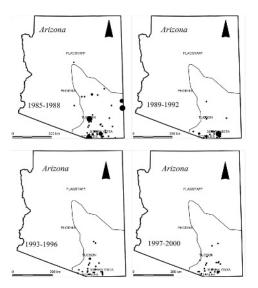


FIGURE 2. Spatial distribution of rabies in Arizona during 1985–2000. Size of dot is proportional to number of rabid skunks. Line represents the border of the overlap zone for the four species of skunks.

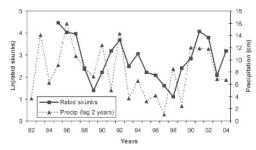


FIGURE 3. The natural logarithm (Ln) of the number of rabid skunks, plotted with precipitation during lactation (May and June) 2 yr previously.

big brown bat variant of rabies virus (Smith et al., 2001), and in this outbreak skunk-to-skunk transmission was suspected (AZDHS, 2005). During 2003 a rabid skunk from south of Flagstaff was found infected with a gray fox rabies virus strain, and in 2004 five skunks from Flagstaff were recovered with the same bat rabies virus strain that was reported during 2001 (AZDHS, 2005).

Precipitation data were divided into discrete periods reflecting skunk biology in southeastern Arizona: gestation (April-May), lactation (June–July), weaning (August-September), and winter (October-March). A significant relationship was detected between the natural log of the number of rabid skunks and two measures of precipitation: precipitation during lactation during the previous 2 yr and during the winter 2 yr prior to the reported case $(F_{3.16}=13.50, P<0.01,$ $R^2 = 0.55$; lnskunks = 0.10 [lactation-1] +0.12 [lactation-2]+0.05 [winter-2]; see Fig. 3). Examination of residuals revealed no evidence of heteroscedascity or autocorrelation.

During field studies from 1996 to 2002, 124 skunks were captured 207 times, including three hog-nosed skunks, 65 hooded skunks, 18 spotted skunks, and 38 striped skunks. Fifty-one plasma samples from skunks were tested by the CDC for rabies virus-neutralizing antibodies, and six brain samples from animals found dead were tested for rabies virus and for molecular typing. Antibodies to rabies virus were detected in samples from one hooded skunk and one striped skunk. Both animals were part of a trap-vaccinaterelocate trial and had been vaccinated 5 mo prior to capture.

Brain samples from one hog-nosed skunk and one hooded skunk tested positive for rabies virus, and both were infected with the south-central skunk rabies virus variant. The hog-nosed skunk was found dead along a dirt road. The hooded skunk, a young female, had been captured and radio-collared in early August 2001. She was observed at 14:30 hr on 20 February 2002 moving rapidly through her home range. She stopped periodically to scratch at the dirt and leaf litter under the oak canopy. Although approached to within 5 m, she did not behave aggressively but did appear to be more agitated than usual. Her carcass was recovered on 23 February, 5 km from where she was last observed, and well outside her known denning range. The remains were located under a tree, and most of the carcass had been consumed; remains were consistent with predation or scavenging by an avian predator (O'Gara, 1978). The skull was sent to the Museum of Southwestern Biology at the University of New Mexico and prepared as a voucher specimen (MSB144500). Rabies in hooded skunks has not been reported previously (Aranda and Lopez-de Buen, 1999). This study was the first to confirm rabies in *M. macroura*; however, it is probable that rabies has been detected in hooded skunks previously, but that either the species of skunk was not recorded or was misidentified as a striped skunk.

Rabies in skunks in North America appears to have a 4–8-yr cycle (Gremillion-Smith and Woolf, 1988; Pybus, 1988). Trends are less clear in Arizona, and if a cycle exits, it may vary from 4 to 10 yr. Other studies have found rabies in skunks to peak in the winter or early spring (Heidt et al., 1982; Pool and Hacker, 1982; Rosatte and Gunson, 1984) or exhibit a bimodal trend, with peaks in the spring and fall (Gremillion-Smith and Woolf, 1988; Krebs et al., 2005). These peaks appear to correspond with increased transmission rates occurring during breeding and fall dispersal. Rabies in skunks in Arizona also peaked during the spring but showed little or no increase during fall months.

The number of rabid skunks was closely related to precipitation 1–2 yr prior to case detection. Precipitation is tied to primary productivity (Rosenzwieg, 1968), which may indicate that precipitationrelated changes in food supply during lactation may increase juvenile survival and ultimately skunk density. Higher density may increase rates of interaction and increase the probability of rabies virus transmission (Verts, 1967; Rosatte et al., 1986).

Additional research is needed on ecology of all four species of skunks in the Southwest, and more data are required on species identification of skunks submitted for rabies testing. If possible, voucher specimens should be prepared (Ruedas et al., 2000). Current protocols for collection of brain tissue usually result in destruction of the skull; however, species can be genetically typed using a small amount of muscle tissue (Dragoo et al., 2004).

Currently, effective oral rabies vaccines are not available for skunks, and trapvaccinate-release programs are not practical over large areas such as southeastern Arizona. Therefore, given the low numbers of rabid skunks and the cyclic nature of the disease in this state, the most effective and economical tool to reduce the threat of human exposure would be continued vaccination of pets and livestock.

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