

# COOPERATIVE RABIES MANAGEMENT PROGRAM NATIONAL REPORT 2006



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**United States Department of Agriculture  
Animal and Plant Health Inspection Service**

**COOPERATIVE RABIES MANAGEMENT PROGRAM  
NATIONAL REPORT 2006**

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## TABLE OF CONTENTS

EXECUTIVE SUMMARY .....	3
ALABAMA .....	9
ARIZONA .....	16
CALIFORNIA .....	22
FLORIDA.....	25
GEORGIA.....	31
KANSAS.....	35
KENTUCKY .....	39
LOUISIANA .....	42
MAINE .....	45
MARYLAND .....	52
MASSACHUSETTS.....	56
MICHIGAN .....	61
MISSISSIPPI .....	65
NEW HAMPSHIRE.....	68
NEW JERSEY .....	72
NEW YORK.....	75
NORTH CAROLINA.....	83
OHIO .....	87
PENNSYLVANIA .....	95
TENNESSEE.....	102
TEXAS.....	107
VERMONT .....	113
VIRGINIA.....	119
WEST VIRGINIA .....	124
WYOMING.....	131
NATIONAL WILDLIFE RESEARCH CENTER.....	134

## EXECUTIVE SUMMARY

Operational oral rabies vaccination (ORV) programs in the U.S. began in the early 1990s in New Jersey and Massachusetts with the goal of preventing the raccoon (*Procyon lotor*) variant of rabies from spreading to populated vacation areas of Cape May and Cape Cod, respectively. The United States Department of Agriculture, Animal and Plant Health Inspection Service, Wildlife Services (WS) program's initial involvement in cooperative ORV was in 1995 in south Texas to prevent canine rabies in coyotes (*Canis latrans*) from gaining a larger foot-hold in the U.S. One year later, an ORV program began in west-central Texas to prevent rabies in gray foxes (*Urocyon cinereoargenteus*). The following year, WS cooperated to implement ORV projects in Ohio and Vermont to prevent the spread of raccoon rabies. Wildlife Services' National Rabies Management Program (NRMP) continued to grow over the next 8 years and in 2006 included ORV programs targeting raccoon rabies in 16 eastern states and canine and gray fox rabies in Texas. In addition, WS continued pilot ORV projects targeting striped skunks (*Mephitis mephitis*) in Flagstaff, Arizona and feral dogs (*Canis familiaris*) on the Navajo Nation in Arizona. Overall in 2006, WS participated in coordinated ORV programs to distribute nearly 12.5 million baits over 216,090 km<sup>2</sup>, an area the size of Idaho (Table 1).

Table 1. Operational oral rabies vaccination (ORV) bait distribution and area baited by Wildlife Services and their cooperators in the United States, 2006.

State	Target species	ORV baits distributed	Area baited (km <sup>2</sup> )	Bait distribution methods
Alabama	Raccoon	1,063,010	9,738	Fixed-wing, ground
Arizona	Striped skunk	3,000	46	Ground
Arizona	Feral dog	373	55	Ground
Florida <sup>a</sup>	Raccoon	903,528	8,414	Fixed-wing, ground, helicopter, boat, bait station
Georgia	Raccoon	96,528	1,444	Fixed-wing, ground
Maine	Raccoon	49,680	747	Fixed-wing, ground
Maryland	Raccoon	99,645	766	Fixed-wing, ground, helicopter
Massachusetts	Raccoon	115,427	496	Ground, helicopter, bait station
New Hampshire	Raccoon	29,473	530	Fixed-wing, ground
New Jersey	Raccoon	45,600	480	Ground, helicopter
New York	Raccoon	1,579,706	17,908	Fixed-wing, ground, helicopter, boat, bait station
North Carolina	Raccoon	184,932	2,889	Fixed-wing, ground
Ohio	Raccoon	1,110,046	12,165	Fixed-wing, ground, helicopter
Pennsylvania	Raccoon	1,359,812	20,580	Fixed-wing, ground
Tennessee	Raccoon	770,618	12,257	Fixed-wing, ground
Texas	Coyote	772,000	33,123	Fixed-wing, ground, helicopter
Texas	Gray fox	2,035,763	58,190	Fixed-wing
Vermont	Raccoon	383,179	8,704	Fixed-wing, ground
Virginia	Raccoon	320,617	5,024	Fixed-wing, ground
West Virginia	Raccoon	1,503,365	22,534	Fixed-wing, ground
Total		12,426,302	216,090	

<sup>a</sup> Includes baits distributed by county officials in Broward County.

In the Northeast, WS continued cooperation with Cornell University, state agencies and international partners in New Brunswick, Quebec, and Ontario, Canada to try to prevent the northern and western spread of raccoon rabies. These ORV zones extended along a portion of the New Brunswick border with Maine, the Quebec border with New Hampshire and Vermont, and the Ontario border in northern and western New York (Figure 1). In June 2006, the first case of raccoon variant rabies was confirmed approximately 11 km (6.6 mi) north of the Vermont border by officials in Quebec. Prior to this case, the northernmost case of raccoon variant had been confirmed in January 2001 in a skunk in Coventry, Vermont (Lake Memphremagog basin) about 16 km (9.6 mi) south of the US-Canada Border. In response to this new case, Vermont WS increased their enhanced rabies

surveillance in the northern part of the state by improving existing and establishing new relationships with the Vermont Agency of Transportation, State Police, US Border Patrol, State game wardens, animal control officers, town health officers, local town officials, and farmers and landowners. Through this effort during the remainder of 2006, WS collected 139 animals (104 raccoons, 29 skunks, 5 red foxes, 1 coyote) from 8 counties within the ORV zones; 29 tested positive for rabies (21 raccoons and 8 skunks). This enhanced surveillance characterized the intensity and magnitude of the border rabies outbreak. Coordinating closely with Quebec, which responded immediately after the case with a modified point infection control program, WS was able to formulate meaningful targeted contingency actions for 2007.

The Appalachian Ridge ORV zone extended from Lake Erie in Ohio and Pennsylvania, south through West Virginia and western Virginia, to northeastern Tennessee and North Carolina in 2006 (Figure 1), where it interfaced with the high mountainous habitats that do not support robust raccoon populations based of WS densities indices. The Appalachian Ridge ORV zone, along with the Georgia-Alabama-Tennessee (GAT) ORV zone, baiting operations near Birmingham, Alabama, and natural barriers made up ORV programs targeting raccoons that reached from Lake Erie to the Gulf of Mexico.

In 2006, WS continued to participate in cooperative ORV projects in Massachusetts, New Jersey, eastern Maryland, Florida, and on Long Island, New York (Figure 1). These projects provided valuable information on ORV and surveillance strategies. Future contingency actions are planned to integrate trap-vaccinate-release (TVR) with ORV to restore Cape Cod to raccoon rabies free status. The current strategy is to systematically conduct control from the eastern part of the Cape back toward the Canal, with the objective of ultimately recreating an ORV zone west of the canal that is sufficient to prevent reemergence on Cape Cod. Cape May, New Jersey is the longest running operational ORV project in the U.S. and WS' role continues to be post-ORV evaluation when possible. The Maryland project is designed to investigate if ORV can be used to eliminate raccoon rabies from peninsulas on Chesapeake Bay. The Florida ORV program is an extension of the long-standing Pinellas County project, with a near-term goal of determining if rabies-free areas can be created and maintained, with the initial attention to Pinellas County. Raccoon rabies was detected on Long Island in August 2004. Enhanced surveillance, TVR and ORV were implemented around the initial focus. Planning for future actions in this area includes input from county, state, federal and university cooperators.

Wildlife Services assumes an important cooperative role with the Texas Department of State Health Services (TDSHS) and several other agencies and organizations in ORV efforts that began in Texas in 1995. The coyote program is a combination of surveillance, with reliance on maintaining a 64-kilometer (40-mile) wide ORV zone along the Rio Grande River (Figure 1), to prevent canine rabies from re-emerging in Texas coyotes from feral dogs in Mexico. Single cases were confirmed near Laredo in 2001 and 2004, but this ORV zone continues to be effective in preventing rabies spread. Since its establishment in 2000, the ORV zone maintained along the Rio Grande has been annually treated with approximately 700,000 vaccine-laden baits over more than 31,000 km<sup>2</sup> (12,000 mi<sup>2</sup>). Wildlife Services is also an important funding and operational partner with the TDSHS in conducting ORV efforts to contain a unique gray fox rabies variant in west-central Texas (Figure 1). In 2006, WS contributed over 2 million baits and assisted with bait distribution over more than 58,000 km<sup>2</sup> (22,000 mi<sup>2</sup>) to contain gray fox rabies in Texas. Wildlife Services also provides field expertise, infrastructure, and equipment to help obtain samples for continued monitoring and evaluating of ORV status targeting coyotes and gray foxes in Texas.

A pilot ORV study conducted in Flagstaff, Arizona that began in 2005 continued in 2006. The study was in response to the re-emergence of rabies in the striped skunk population that originated in big brown bats (*Eptesicus fuscus*). These studies represented the first operational attempts to field test the performance of Raboral V-RG<sup>®</sup> vaccine (Merial Limited, Athens, Georgia, USA) specifically targeting skunks. Over the 2-year study, only 8 of 54 skunks (14.8%) demonstrated a detectable rabies antibody response (titer  $\geq 0.05$  IU) and 7 of the 8 had a record of recent hand vaccination with Rabdomun<sup>®</sup>. In the eastern U.S., where raccoon variant rabies frequently spills over into striped skunks, there has been virtually no detectable antibody response related to large scale, multi-year ORV projects targeting raccoons. The significance of skunks infected with raccoon rabies remains unclear, but the apparent inability to orally vaccinate them with the currently licensed oral rabies vaccine and bait could potentially confound progress toward effective raccoon rabies control.

A pilot study continued from 2005 in the vicinity of Chinle, Arizona on the Navajo Nation to field test Raboral V-RG<sup>®</sup> and determine rabies titer levels in feral dogs that may have been exposed to the vaccine. These were the first field trials of their kind specifically targeting feral dogs in the U.S. Over the 2 years, 38 of 143 dogs (26.6%) showed a positive rabies antibody response. Coated sachet (CS) baits were used in all ORV field trials in Arizona in 2006.

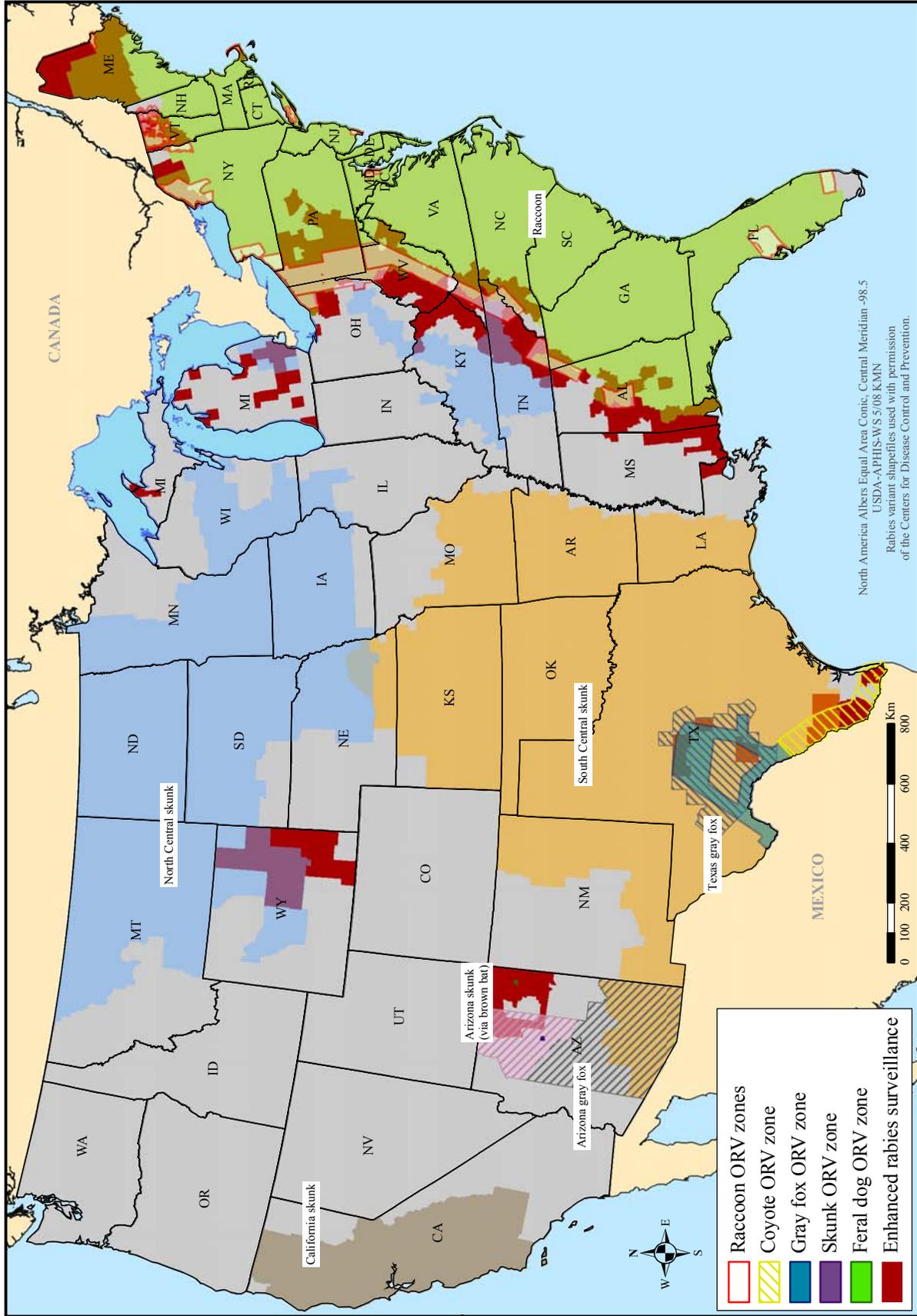


Figure 1. Terrestrial wildlife rabies variants with species-specific cooperative oral rabies vaccination (ORV) zones and Wildlife Services enhanced rabies surveillance counties in the United States, 2006.

In 2006, WS and cooperators continued to shift from fishmeal polymer (FMP) baits to CS's, with nearly 6 million CS baits distributed. At \$1.00/bait CS's are \$0.25 less expensive than FMP baits, less likely to cause damage from aerial distribution, more palatable to smaller carnivores like skunks, and perform generally at least as well as FMP baits based on field titer responses from Cornell University. The shift to CS's is currently viewed as only an interim management step until improved or new baits can be developed, licensed and produced.

Wildlife Services and cooperators continued contingency actions in Ohio to prevent the spread of raccoon rabies and restore the treatment areas to rabies free status. Contingency actions were also conducted in Massachusetts in continued response to rabies on Cape Cod, and in Chattanooga, Tennessee to bolster the existing ORV zone. Contingency actions often include an integration of ORV, TVR, and increased enhanced rabies surveillance. Enhanced surveillance is designed to complement public health surveillance and provide greater sensitivity in delineating the leading edge of rabies distribution, allowing for sound ORV decisions while maximizing the effective use of resources. Enhanced surveillance includes obtaining samples from: animals exhibiting behaviors suggestive of rabies, but not implicated in human or domestic animal exposures; road kills; other animals found dead; animals with wounds or lesions suggestive of rabies; and animals removed near locations where rabies has recently been confirmed. In 2006, WS continued to enhance rabies surveillance in most of the states conducting ORV, as well as emphasizing surveillance in adjacent states west of the raccoon ORV zone, including Michigan, Kentucky, Mississippi and Louisiana (Figure 1). In addition, Wyoming collected and submitted 254 animals for rabies testing (2 bats tested positive); Texas collected and tested 210 animals (3 positives); and Arizona collected and tested 29 animals (all negative). Overall during enhanced surveillance efforts in 2006, WS' cooperation led to the collection and submission of 7,281 samples for rabies testing that otherwise may not have been tested through the public health surveillance system (Table 2); 169 tested positive for rabies.

Table 2. Wildlife Services enhanced rabies surveillance and use of the direct rapid immunohistochemistry test as part of rabies management programs in the U.S., 2006.

<b>State</b>	<b>Enhanced surveillance animals</b>	<b>WS tested by dRIT</b>	<b>Rabid by dRIT</b>
Alabama	322	320 (99.3%)	0
Arizona	29	0	
Florida	0		
Georgia	105	105 (100%)	0
Kentucky <sup>b</sup>	230	230 (100%)	0
Louisiana <sup>b</sup>	34	34 (100%)	0
Maine	62	18 (29.0%)	1
Maryland	0		
Massachusetts	93	0	
Michigan <sup>b</sup>	94	94 (100%)	0
Mississippi <sup>b</sup>	106	106 (100%)	0
New Hampshire	0		
New Jersey	0		
New York	70	0	
North Carolina	105	105 (100%)	25
Ohio	609	609 (100%)	4
Pennsylvania	2,712	2,442 (90.0%)	47
Tennessee	1,573	1,573 (100%)	0
Texas	210	210 (100%)	3
Vermont	139	0	
Virginia	196	187 (95.4%)	1
West Virginia	338	338 (100%)	6
Wyoming	254	0	
<b>Total</b>	<b>7,281</b>	<b>6,400 (87.9%)</b>	<b>87</b>

<sup>a</sup> ORV not applied in this state.

Since 2005, 25 WS personnel have attended direct rapid immunohistochemistry test (dRIT) training at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia. The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals continue to be processed by public health experts at established local, state, or federal laboratories. In 2005 and 2006, WS implemented the dRIT in 16 states. In 2006, WS tested 6,400 (87.9%) animals from enhanced surveillance; 87 tested positive for rabies (Table 2).

RabID, a GIS-based surveillance mapping tool developed and implemented at the CDC, continued to undergo phased implementation in eastern states involved in raccoon rabies control. This tool provides nearly real-time access to spatial-temporal rabies distribution data that includes results from enhanced rabies surveillance submissions and dRIT testing. Currently, Alabama, Florida, Georgia, Kentucky, Ohio, Tennessee, Virginia, and West Virginia are routinely submitting data to the RabID system; Michigan, Mississippi, North Carolina and Pennsylvania have begun to submit data.

In all states conducting ORV, WS continues to take the lead on post-ORV monitoring to evaluate program effectiveness by collecting blood and tooth samples for determining rabies virus neutralizing antibody (VNA) levels and bait uptake (when appropriate) in raccoons, skunks, coyotes, and foxes. Density indexing is also used to characterize raccoon and skunk populations and to provide post-ORV serum samples for analysis. It is typical to report on 2005 serology in this 2006 report to allow time for laboratory analysis results. At the time of printing however, most states had received 2006 serology results as well.

In 2005, a mean positive antibody response (VNA  $\geq 0.05$  IU) of  $21 \pm 11\%$  was observed for 4,413 raccoon serum samples collected post-ORV (Table 3). Wildlife Services also collected sera from 16 skunks, 1 gray fox and 1 red fox during raccoon rabies management programs. One skunk (6%) and 1 gray fox (100%) had a detectable rabies VNA response. Texas collected serum samples from 115 coyotes and 141 gray foxes to evaluate ORV effectiveness targeting those species in south and west-central Texas, respectively; 29 coyotes (25%) and 80 gray foxes (57%) had a positive rabies VNA response. Arizona collected serum samples from 12 skunks and 40 dogs within the ORV pilot study areas of Flagstaff and Chinle, respectively. One skunk (8%) and 5 dogs (13%) had positive rabies VNA responses.

In 2006, a mean positive antibody response of  $32 \pm 12\%$  was observed for 2,819 raccoon serum samples collected post-ORV (Pennsylvania results were not available at the time of printing) (Table 3). Sera were also collected from 48 skunks, 2 gray foxes, and 1 red fox during raccoon rabies management programs. Two skunks (4%) and both gray foxes (100%) had a detectable rabies VNA response. Texas collected serum samples from 145 coyotes and 146 gray foxes to evaluate ORV efficacy targeting those species in south and west-central Texas, respectively; 49 (34%) coyotes and 74 (51%) gray foxes demonstrated positive rabies VNA responses (Table 3). Texas also collected sera from 19 coyotes, 12 skunks, 9 raccoons, and 8 bobcats during evaluation of the gray fox ORV program. Four coyotes (21%), 8 skunks (67%), 1 raccoon (11%), and 1 bobcat (13%) had detectable rabies antibodies. Arizona collected serum samples from 42 skunks and 103 dogs within the ORV pilot study areas of Flagstaff and Chinle, respectively (Table 3). Seven skunks (17%) and 33 dogs (32%) had positive rabies VNA responses. Over the 2-year skunk ORV study, 8 of 54 skunks had rabies antibodies, but 7 of the 8 had a recent record of hand vaccination.

The need for a bait-vaccine combination producing higher levels of rabies VNA in meso-carnivore species serving as rabies reservoirs remains the highest research priority and requires continued systematic research. In 2006, external research at Thomas Jefferson University in Philadelphia, Pennsylvania focused on the development of new, safe, and effective oral rabies vaccines, with canine adenovirus as a prospective vector for expression of the rabies glycoprotein gene.

Several pen, laboratory, and field studies were continued or initiated through WS' National Wildlife Research Center (NWRC) in Fort Collins, Colorado. Pen and laboratory studies were conducted at the NWRC and Colorado State University, while field studies were conducted in Alabama, Ohio, Pennsylvania, and Texas. Studies involved research on: bait development to better deliver Raboral V-RG® to raccoons and skunks; ecology of raccoons and gray foxes in rural and urban areas; better use of biomarkers to evaluate vaccine uptake by target and non-target wildlife; the evaluation of geographic barriers for wildlife dispersal that may affect the spread of rabies; and determining long-term efficacy of Raboral V-RG® in raccoons.

Table 3. Wildlife Services post-oral rabies vaccination (ORV) sampling efforts as part of rabies management programs in the U.S., 2005 and 2006<sup>a</sup>.

State	2005				2006			
	All post-ORV serum samples	Positive rabies antibody response ( $\geq 0.05$ IU)	4-12 <sup>b</sup> weeks post-ORV samples	Positive rabies antibody response ( $\geq 0.05$ IU)	All post-ORV serum samples	Positive rabies antibody response ( $\geq 0.05$ IU)	4-12 <sup>b</sup> weeks post-ORV samples	Positive rabies antibody response ( $\geq 0.05$ IU)
Raccoon <sup>c</sup>								
Alabama	398	91 (22.9%)	398	91 (22.9%)	387	105 (27.1%)	387	105 (27.1%)
Florida	548	96 (17.5%)	548	96 (17.5%)	271	32 (11.8%)	271	32 (11.8%)
Georgia	134	40 (29.8%)	134	40 (29.8%)	123	31 (25.2%)	123	31 (25.2%)
Maine	85	39 (45.9%)	85	39 (45.9%)	94	38 (40.4%)	92	36 (39.1%)
Maryland	210	41 (19.5%)	210	41 (19.5%)	176	56 (31.8%)	176	56 (31.8%)
Massachusetts	34	5 (14.7%)	25	2 (8.0%)	29	11 (37.9%)	1	0
New Hampshire	7	2 (28.6%)	7	2 (28.6%)	11	5 (45.5%)	11	5 (45.5%)
New Jersey	0		0		0		0	
New York	101	28 (27.7%)	101	28 (27.7%)	198	43 (21.7%)	198	43 (21.7%)
North Carolina	133	9 (6.8%)	133	9 (6.8%)	121	16 (13.2%)	121	16 (13.2%)
Ohio	838	88 (10.5%)	434	47 (10.8%)	453	163 (36.0%)	345	136 (39.4%)
Pennsylvania	677	25 (3.7%)	373 <sup>d</sup>	13 (3.5%)	not available at printing			
Tennessee	488	111 (22.7%)	488	111 (22.7%)	267	94 (35.2%)	267	94 (35.2%)
Vermont	239	38 (15.9%)	239	38 (15.9%)	189	39 (20.6%)	189	39 (20.6%)
Virginia	194	63 (32.5%)	194	63 (32.5%)	238	135 (56.7%)	238	135 (56.7%)
West Virginia	327	69 (21.1%)	275	51 (18.5%)	262	100 (38.2%)	212	76 (35.8%)
Total	4,413	745 (16.9%)	3,644	671 (18.4%)	2,819	868 (30.8%)	2,631	804 (30.6%)
Mean $\pm$ St. Dev.		21 $\pm$ 11%		21 $\pm$ 11%		32 $\pm$ 12%		29 $\pm$ 15%
Arizona (skunk)	12	1 (8.3%)	12	1 (8.3%)	42	7 (16.7%)	42	7 (16.7%)
Arizona (feral dog)	40	5 (12.5%)	n/a <sup>e</sup>	n/a	103	33 (32.0%)	n/a <sup>e</sup>	n/a
Texas (coyote)	115	29 (25.2%)	115	29 (25.2%)	146	74 (50.7%)	146	74 (50.7%)
Texas (gray fox)	141	80 (56.7%)	141	80 (56.7%)	145	49 (33.8%)	145	49 (33.8%)

<sup>a</sup> Most states report on 2005 serology in the 2006 report to allow for results to be returned from various labs. At the time of printing however, most states had serology results for the current year (2006) so they have been included here.

<sup>b</sup> Samples taken during optimal evaluation period of 4-12 weeks post-ORV bait distribution.

<sup>c</sup> Non-raccoon samples collected during raccoon rabies management programs are not represented in this table.

<sup>d</sup> Most samples collected during evaluation of a Spring Bait Study; 8 of 315 (2.5%) samples demonstrated positive rabies antibodies.

<sup>e</sup> In 2005, samples were collected 4-36 weeks post-ORV and 6-24 weeks post-ORV in 2006.

In 2007, WS and cooperators will continue to focus on implementing adequate enhanced rabies surveillance in conjunction with ORV to determine areas at risk of rabies spread and to monitor program success. Adjustment to ORV zones may occur as a result of improved surveillance information. Commitments will continue toward improvements to the existing bait-vaccine and development of new bait-vaccines that perform better and are efficacious in all carnivore reservoirs. Lastly, WS will continue to participate in formal meetings with counterparts from Canada and Mexico on the development of a North American Rabies Management Plan that identifies information exchange, enhanced surveillance, rabies control, and research as key needs toward meeting continental goals for rabies management.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM ALABAMA 2006

### BACKGROUND

Raccoon (*Procyon lotor*) rabies is thought to have entered Alabama in the late 1970s from Florida. The raccoon variant of the rabies virus has since been detected in most counties east and south of the Alabama-Coosa River system and is now considered enzootic there. Within the last 10 years, several confirmed raccoon rabies breaches of the Alabama and Coosa Rivers (Figure 1) have occurred in Mobile, Clarke, Dallas, Perry, and Autauga Counties, but the cases appeared to be fairly isolated and limited to 1 or 2 animals during each occurrence.

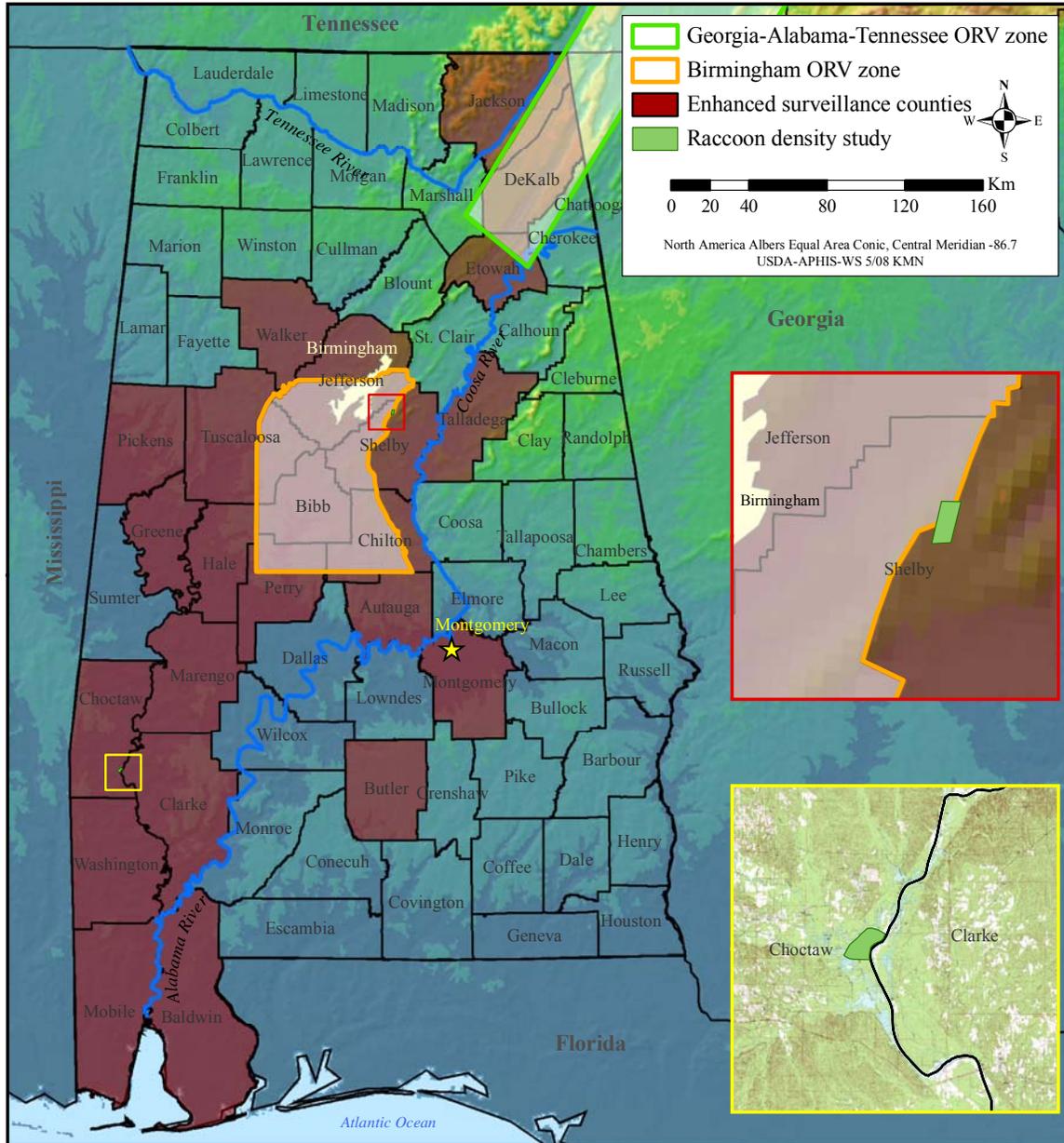


Figure 1. Wildlife Services cooperative rabies management program activities in Alabama, 2006.

In 2001, the Alabama Department of Public Health (ADPH) asked Wildlife Services (WS) and other cooperators to help determine the leading edge of the raccoon variant in the state in hopes of developing an effective oral rabies vaccination (ORV) program to keep raccoon rabies from spreading into western Alabama. In late 2001,

WS began conducting enhanced surveillance of road killed and trapped raccoons in counties west of, and bordering, the Alabama and Coosa Rivers.

Between 2002 and 2005, surveillance conducted by WS and the ADPH confirmed 16 cases of raccoon rabies in both domestic and wild animals in Autauga, Clarke, and Elmore Counties (where it had previously been detected) and Cherokee, DeKalb, Jefferson, and Shelby Counties (where it had never been detected), indicating that raccoon rabies might be on the move westward. As a result of earlier positives, in the fall of 2003, the ADPH and the Alabama Department of Agriculture and Industries (ADAI) cooperated with Alabama WS and WS offices in Georgia and Tennessee to initiate Alabama's first ever ORV effort in 5 northeast counties. Since then, Alabama WS has participated in 8 additional ORV baiting efforts in both northeast Alabama (as part of the Georgia-Alabama-Tennessee [GAT] zone) and central Alabama (Birmingham and Selma zones), all in response to newly confirmed positive cases that indicated raccoon rabies was potentially moving westward.

No new raccoon rabies cases were identified west of the Alabama-Coosa Rivers in 2006. However, Alabama WS continued surveillance efforts of sick and strange acting animals in counties west of the rivers, particularly in Jefferson and Shelby Counties, where 6 raccoon rabies cases were detected in 2004 and 2005.

## **ORV PROGRAM 2006**

### **Bait Distribution**

For the fourth consecutive year, WS participated in bait distribution efforts throughout central and northeastern Alabama; 1,063,010 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 9,738 km<sup>2</sup> (3,760 mi<sup>2</sup>) in 2006 (Figure 1). Since its program inception in 2003, WS has distributed 2,648,290 ORV baits in Alabama. Aircraft and pilots for all Alabama ORV programs in 2006 were provided by Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA), while WS provided ground support and served as navigators and flight crew in the planes.

*Birmingham (Spring and Fall).*--The objective of the 2006 Birmingham ORV zone, which was baited in the spring and fall, was to respond to previous raccoon rabies cases detected in Shelby County in 2004 and 2005 and in Birmingham in October 2005. Raccoon rabies had not been detected in Jefferson or Shelby Counties prior to these occurrences. From 4-7 April, 442,021 baits (326,041 fishmeal-coated sachets [CS] via fixed-wing aircraft and 115,980 fishmeal polymer [FMP] baits by hand baiting) were distributed over 6,537.6 km<sup>2</sup> (2,524.2 mi<sup>2</sup>) of Autauga, Bibb, Chilton, Dallas, Hale, Jefferson, Perry, Shelby, Tuscaloosa, and Walker Counties (Figure 1). The same area was baited again from 23-24 October with 441,448 baits (325,808 CS's via fixed-wing aircraft and 115,640 FMPs baits by hand baiting). Ground support for aerial baiting and ground bait distribution was provided by WS, Jefferson County Department of Health environmentalists, and volunteers from the Centers for Disease Control and Prevention (CDC).

*Georgia-Alabama-Tennessee.*--The objective of Alabama's 2006 GAT ORV zone was to continue to prevent the westward movement of raccoon rabies from northwest Georgia into Cherokee and DeKalb Counties. No new raccoon rabies cases had been detected in northeast Alabama since the first GAT program was initiated in 2003. From 16-23 October, 179,541 FMP baits (169,431 by air and 10,110 by hand) were distributed over 3,200.2 km<sup>2</sup> (1,235.6 mi<sup>2</sup>) of Cherokee, DeKalb, Etowah, Jackson, and Marshall Counties (Figure 1). Ground support for aerial baiting and ground bait distribution was provided by WS and volunteers from the CDC. Cooperators from Merial Limited also served as navigators and flight crew in the planes.

### **Enhanced Surveillance**

In 2006, WS continued to enhance rabies surveillance in 23 counties (Figure 1) by collecting abnormally behaving raccoons, road killed raccoons, nuisance raccoons, and raccoons found dead in unusual places in counties west of the Alabama and Coosa Rivers. The goal of this surveillance was to determine the leading edge of the raccoon variant in Alabama. Wildlife Services cooperated with animal control personnel, county health department environmentalists, and wildlife law enforcement officers to collect 322 animals for rabies testing (Table 1). All of these animals tested negative for rabies.

Table 1. Animals collected for rabies testing by Wildlife Services along the Alabama-Coosa River system and westward in Alabama, 2006 (no rabies positives).

County	Raccoon	Coyote	Gray fox	Red fox	Bobcat	Total
Autauga	4					4
Baldwin <sup>a</sup>	1					1
Bibb	4					4
Butler <sup>a</sup>	1					1
Chilton	8				1	9
Choctaw <sup>a</sup>	1					1
Clarke <sup>a</sup>	1		1	1		3
De Kalb	8		1			9
Etowah	1					1
Greene <sup>a</sup>	2					2
Hale	27	1				28
Jackson	6			3		9
Jefferson	59	1	1		1	62
Marengo <sup>a</sup>	8					8
Mobile <sup>a</sup>	92			1		93
Montgomery			1			1
Perry	3					3
Pickens <sup>a</sup>	2					2
Shelby	42					42
Talladega <sup>a</sup>	2					2
Tuscaloosa	25	1				26
Walker <sup>a</sup>	1	2	4			7
Washington <sup>a</sup>	4					4
Total	302	5	8	5	2	322

<sup>a</sup> ORV never applied in this county.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

During 2006, WS collected 322 animals for rabies testing (Table 1) and 320 were tested by WS using the dRIT (99.3%). Two samples were forwarded directly to the ADPH testing laboratory because of reports of human or animal exposure. Ten percent of all negatives were sent to the CDC for confirmation. The CDC (using the dFA test) had 100% agreement with the WS dRIT test results for negative samples. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Alabama.

### Population Monitoring

Wildlife Services conducted 2 raccoon density studies in 2006, 1 each in Shelby and Choctaw Counties (Figure 1 insets). The National Rabies Management Program standard protocol (50 traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights) was used during both studies. The Shelby County study took place in an urban/suburban area of Chelsea, Alabama just east of the Birmingham ORV zone. Some of the study area fell within the bait zone and some of it had never been baited. Given the close proximity of the density study to the Birmingham zone, raccoons captured during the study may have been moving in and out of the ORV zone. The Choctaw study was a replicate of a 2003 study conducted on the Choctaw National Wildlife Refuge and Wildlife Management Area. This is an ORV naïve area (never before treated with ORV) west of previously confirmed rabies cases.

Twenty-six and 35 unique raccoons were captured during the Shelby and Choctaw studies, respectively (Table 2). The Choctaw study had similar captures in 2003 when 41 unique raccoons were trapped. During both 2006 studies, blood serum samples were collected to test for the presence of rabies virus neutralizing antibodies (VNA) and first premolars were extracted for aging purposes. All but 1 raccoon in the 2 studies were released. One died under care and tested negative for rabies using the dRIT. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

Table 2. Index to raccoon densities in Shelby and Choctaw Counties of Alabama, 2006.

	Shelby County	Choctaw County
Time of study	21-31 August	5-15 September
Macrohabitat	Urban/suburban	Forested
Target trap nights	500	500
Unique raccoons	26	35
Recaptured raccoons	11	19
Non-target captures	56	28
Area (km <sup>2</sup> )	2.85	2.87
Raccoon density index <sup>a</sup>	9.1	12.2

<sup>a</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

### Post-ORV Monitoring

*Georgia-Alabama-Tennessee.*--Post-ORV sampling for Alabama's 2006 GAT ORV zone was initiated 5 weeks post-ORV on 28 November. Cage traps were used to capture 133 unique raccoons from DeKalb County. All raccoons were immobilized, processed and released.

*Birmingham.*--Post-ORV sampling for the spring and fall 2006 Birmingham ORV zones were initiated 4 and 5 weeks post-ORV on 9 May and 1 December, respectively. During spring post-bait sampling 131 raccoons and 2 gray foxes were captured and during fall post-bait trapping 123 raccoons were captured. Animals were captured using cage traps in Bibb, Chilton, Jefferson, Shelby, and Tuscaloosa Counties. Two animals were found dead in the traps during the spring post-bait evaluation, and 254 were immobilized, processed and released.

### Raccoon Movements Study

Field work for a telemetry study initiated in January 2004 to monitor raccoon habitat use and movements relative to the Alabama River concluded in December 2005. As many as 217 raccoons were captured and radio-collared from 4 counties as part of this project. Manuscripts are being prepared for publication, and analyses indicated that the Alabama River and managed pine habitat along the river affected the movements of raccoons and may have decreased the likelihood of rabies establishing to the west of the river.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 141 opossums (*Didelphis virginiana*), 38 domestic/feral cats (*Felis catus*), 2 Eastern box turtles (*Terrapene carolina*), 2 gray foxes (*Urocyon cinereoargenteus*), 1 cotton rat (*Sigmodon hispidus*), and 1 feral/free-ranging dog (*Canis familiaris*).

Non-target animals that were captured and euthanized by WS in 2006 included: 42 opossums, 1 coyote (*Canis latrans*), and 1 gray fox. All animals euthanized by WS in 2006 were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

### Rabies Laboratory Cooperation

Wildlife Services' ORV program in Alabama cooperates with the ADPH Bureau of Clinical Laboratories and the CDC.

*Alabama Department of Public Health Bureau of Clinical Laboratories.*--The ADPH tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure

to the rabies virus). The ADPH confirmed 82 cases of rabies in Alabama in 2006: 56 raccoons, 17 bats (*Chiroptera* spp.), 7 foxes, and 2 cats. For more information about the ADPH please visit: <http://www.adph.org/>

*Centers for Disease Control and Prevention.*--The CDC tests animal brainstems for rabies as part of enhanced surveillance (specimens not involved in an exposure and usually collected by WS). The CDC also analyzes wildlife blood serum samples (submitted by WS) for levels of rabies VNA.

In 2006, the CDC tested 21 wildlife brainstem samples submitted by Alabama WS. This was a 91.1% decrease from the 235 brainstems submitted by Alabama WS in 2005. This decline represents the full implementation of the dRIT procedure by Alabama WS. The 21 brainstems submitted tested negative using dRIT, and were forwarded to CDC for confirmation/validation of the testing technique. Alabama WS also submitted 585 blood serum samples for rabies VNA analysis to the CDC in 2006. This represented a 45.4% increase from the 319 samples submitted by WS in 2005. This increase in samples reflects the addition of a new bait zone that was baited twice in 2006. The Alabama ORV program anticipates a similar number of brainstem and serum sample submissions to CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## **ORV PROGRAM 2005 and 2006 – EVALUATION**

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, some of the current year's (2006) evaluation data were available so they have been included here as well.

### **Serology, Tetracycline Biomarker, and Age Results**

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and teeth are analyzed to determine animal age and bait uptake. Fishmeal polymer baits contain a chemical biomarker (tetracycline) that stains teeth/bone and can be detected under microscope; fishmeal coated sachet baits do not contain this biomarker. Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, during the Alabama cooperative rabies management program, WS live-trapped 483 unique raccoons; 401 were trapped 4-7 weeks post-2005 ORV bait distribution and 82 were trapped in an ORV naïve area prior to the 2005 Birmingham ORV distribution (Table 3). Blood and tooth samples were collected from most of these animals and serum samples were sent to the CDC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA). Overall, 91 of the 398 (22.9%) serum samples collected post-ORV demonstrated a presence of rabies VNA (titer  $\geq 0.05$  IU), while 2 of the 82 (2.4%) samples collected in ORV naïve areas showed rabies antibodies. One of these 2 raccoons was actually trapped in an area of Chilton County that had been baited during the March 2005 Selma ORV campaign which may explain the presence of rabies antibodies. The other raccoon was trapped in an ORV naïve area of Tuscaloosa County approximately 28 miles from the nearest ORV campaign (2005 Selma zone). It is not known if this antibody response may be attributed to an actual exposure to the rabies virus, or if it is the result of the raccoon eating an ORV bait and then traveling a long distance to where it was trapped. Both raccoons were trapped in October 2005 prior to the first baiting in the Birmingham ORV zone.

Table 3. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Alabama, 2005.

	<b>Birmingham ORV naïve<sup>a</sup></b>	<b>Selma post-ORV</b>	<b>GAT post-ORV</b>	<b>Birmingham post-ORV</b>
Sample collection timeframe	October	5-12 April	28 Nov.-4 Dec.	6-15 December
Last ORV date (and aerial bait type) <sup>b</sup>	n/a <sup>a</sup>	5 March (FMP)	25 October (FMP)	30 October (CS)
Weeks post-ORV	n/a <sup>a</sup>	4-5	5-6	5-7
Unique raccoons	82	160	121	120
<b>Serology</b>				
Testable blood samples	82	158	120	120
Positive rabies antibody response ( $\geq 0.05$ IU)	2 (2.4%) <sup>c</sup>	38 (24.1%)	29 (24.2%)	24 (20.0%)
<b>Tetracycline</b>				
Testable tooth samples <sup>d</sup>	n/a <sup>a</sup>	148	125	135
Presence of tetracycline biomarker		24 (16.2%)	29 (23.2%)	13 (9.6%) <sup>e</sup>

<sup>a</sup> Samples collected in an ORV naïve area (never before treated with ORV) prior to initial ORV.

<sup>b</sup> CS=coated sachet; FMP=fishmeal polymer.

<sup>c</sup> When mapped, 1 of these raccoons was actually trapped in an area of Chilton County that had been baited during the March 2005 Selma ORV campaign.

<sup>d</sup> Samples include teeth collected from road killed animals in zone.

<sup>e</sup> Fishmeal polymer baits (which contain tetracycline) were distributed by hand in areas too populated to bait by air and likely account for the tooth samples that showed the presence of biomarker.

In 2006, WS live-trapped 448 unique raccoons and 2 gray foxes during post-ORV and ORV naïve trapping activities. Serology data on the 61 raccoons captured during 2 density studies were not available at the time this report was printed. Tetracycline data for all samples were also pending at the time of this report. Overall, 105 of the 387 (27.1%) raccoon serum samples collected post-ORV demonstrated a presence of rabies VNA, while both (100%) of the gray fox samples showed rabies antibodies.

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Alabama, 2006.

	<b>Birmingham spring post-ORV</b>	<b>GAT post-ORV</b>	<b>Birmingham fall post-ORV</b>
Sample collection timeframe	9-12 May	28-30 November	1-15 December
Last ORV date (and aerial bait type) <sup>a</sup>	7 April (CS)	23 Oct. (FMP)	24 Oct. (CS)
Weeks post-ORV	4-5	5	5-7
Unique raccoons	131	133	123
<b>Serology</b>			
Testable blood samples	131	133	123
Positive rabies antibody response ( $\geq 0.05$ IU)	29 (22.1%)	31 (23.3%)	45 (36.6%)
<b>Tetracycline</b>			
Presence of tetracycline biomarker	pending	pending	pending

<sup>a</sup> CS=coated sachet; FMP=fishmeal polymer.

*Age Results.*--In 2005, 739 raccoon teeth were aged using premolars of live-captured animals and canines of animals found dead or euthanized. These samples were collected from raccoons during the raccoon movement study, enhanced surveillance, ORV-naïve trapping, and post-ORV trapping (Figure 2). Age class distribution was typical with the majority of raccoons being young of the year juveniles (<1 y.o.) or 1-year olds. Three raccoons  $\geq 10$  y.o. were captured: 2 at 10 y.o. and 1 at 12 y.o. Age results from 2006 were pending at the time of this report.

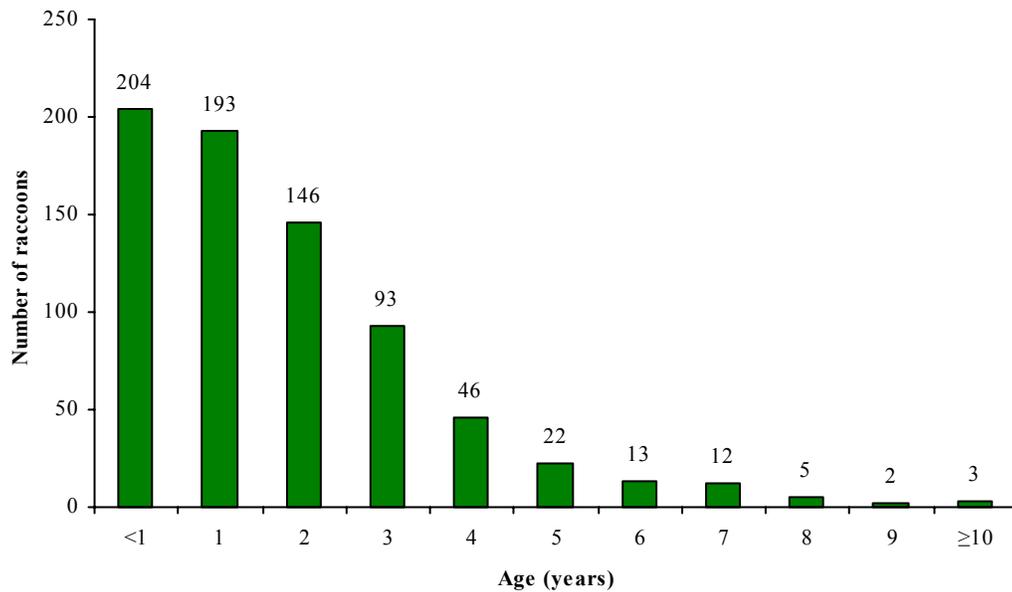


Figure 2. Age class distribution of 739 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in Alabama, 2005.

## SUMMARY

In 2006, WS completed its sixth year of cooperative participation in rabies management in Alabama. Work emphasized surveillance of raccoon rabies west and north of the Alabama and Coosa Rivers as well as ORV naïve and post-ORV monitoring and evaluation. Oral rabies vaccination efforts were continued in Birmingham and its surrounding counties and in northeast Alabama in an attempt to stop the westward movement of the raccoon variant from northwest Georgia and southeast Tennessee. Future ORV baiting strategies in Alabama will continue to be directed towards halting the spread of raccoon rabies into western Alabama and beyond. These ORV zones will be tied to a national planning effort to contain the disease and explore strategies to eliminate the raccoon variant of the rabies virus from North America.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM ARIZONA 2006

### BACKGROUND

The striped skunk (*Mephitis mephitis*) and gray fox (*Urocyon cinereoargenteus*) are the primary reservoirs of terrestrial rabies in Arizona. The South Central skunk variant of the rabies virus typically occurs in the southeastern counties of Arizona. The Arizona gray fox variant generally runs through the eastern counties and into the central part of the state, below the Mogollon Rim. A bat variant of the rabies virus, common in big brown bats (*Eptesicus fuscus*), occurs throughout the state. In 2001, a skunk was infected with a bat variant causing an unprecedented rabies outbreak in Flagstaff (Coconino County). By year's end, this rare spillover had infected 19 rabid skunks and was the first documented event of a bat variant of the rabies virus being transmitted and maintained in skunks. This spillover reoccurred in late 2004 and again in 2005, prompting a trap-vaccinate-release (TVR) campaign by Wildlife Services (WS) in Flagstaff. This campaign continued opportunistically throughout 2006 (Figure 1).

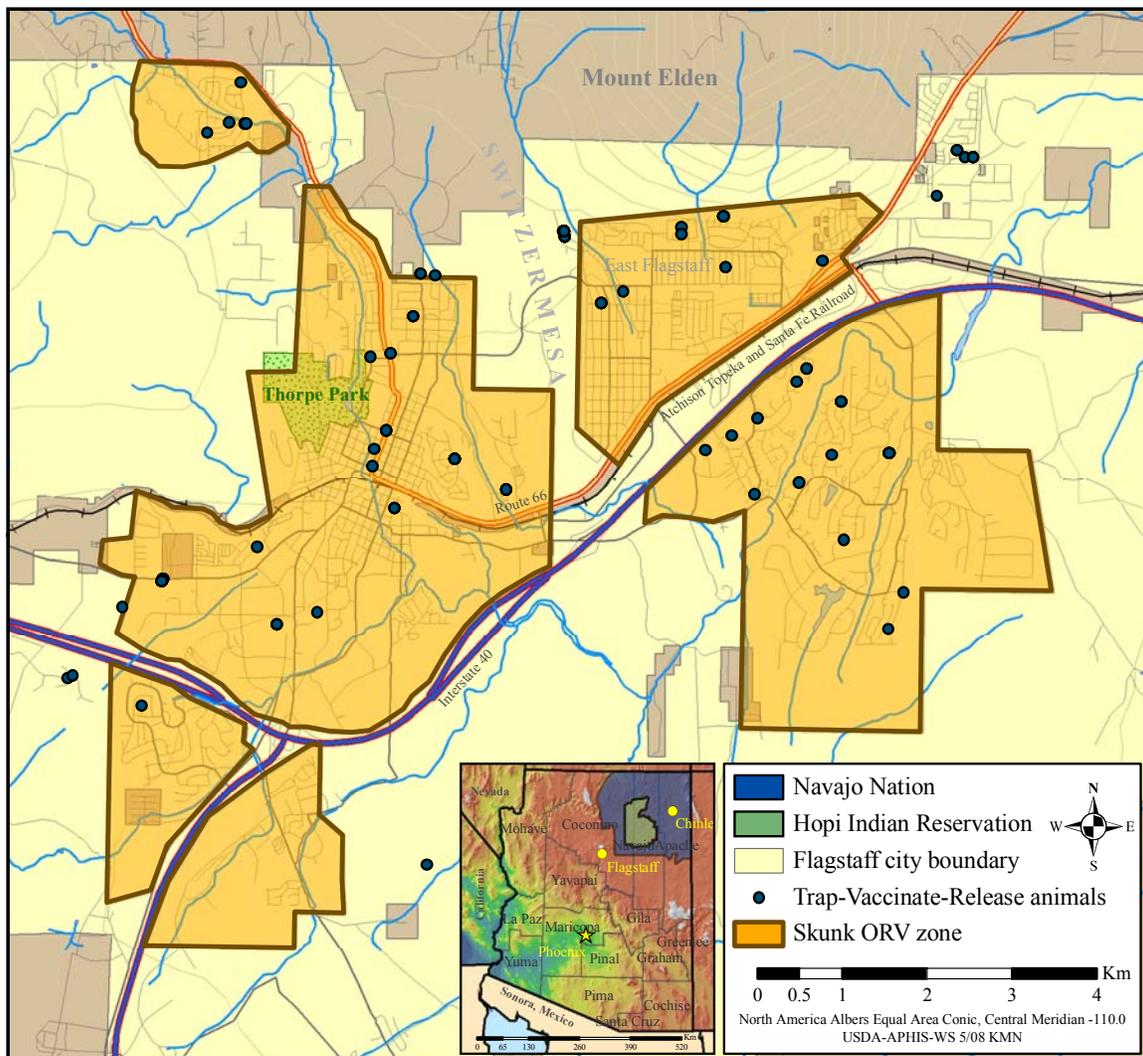


Figure 1. Wildlife Services cooperative rabies management program activities in Arizona, 2006.

In 2006, the Arizona Department of Health Services (ADHS) tested 2,499 animals and confirmed 140 cases of rabies, a 17% decrease in positive cases from 2005. The 2006 rabies cases occurred in 11 of the 15 counties in

Arizona (Table 1). For more information on animals tested from Arizona in 2006 by the ADHS please visit: [www.azdhs.gov/phs/oids/vector/rabies/stats.htm](http://www.azdhs.gov/phs/oids/vector/rabies/stats.htm)

Table 1. Animals confirmed positive for the rabies virus by the Arizona Department of Health Services in Arizona, 2006.

County	Bat	Skunk	Fox	Coyote	Other	Total
Cochise	3	2				5
Coconino	4					4
Gila	3		8	1 <sup>a</sup>	1 (mt. lion) <sup>a</sup>	13
Greenlee		1	4			5
Maricopa	9					9
Navajo	2					2
Pima	66	11	2		2 (bobcats) <sup>a</sup>	81
Pinal	3		6			9
Santa Cruz	2	2	2		1 (cat) <sup>b</sup>	7
Yavapai	3				1 (bobcat) <sup>a</sup>	4
Yuma	1					1
Total	96	16	22	1	5	140

<sup>a</sup> Arizona gray fox variant of rabies..

<sup>b</sup> South Central skunk variant of rabies.

## RABIES MANAGEMENT PROGRAM 2006

### Operational Assistance

Wildlife Services assisted with a localized outbreak of the Arizona gray fox rabies variant in Greenlee County. Arizona WS personnel found 38 dead gray foxes along the Eagle Creek drainage and reported the die-off to local animal control. A localized population reduction campaign resulted in the removal of 3 foxes from the area with 1 fox submitted for rabies testing to the ADHS lab. This fox along with 3 others submitted from the same drainage tested positive for rabies in 2006.

Arizona WS also assisted with a localized outbreak of rabies in gray foxes at a public botanical park in Pinal County. A patron of the park was chased and bitten by a fox which tested positive for rabies. The patron received post-exposure-prophylaxis after the incident. Wildlife Services conducted a localized population reduction in the park and removed 2 hooded skunks (*Mephitis macroura*), 1 raccoon, 1 ringtail (*Bassariscus astutus*), and 1 gray fox. All animals were submitted for testing to the ADHS and the fox tested positive for rabies.

Wildlife Services also cooperated with the Southeastern Cooperative Wildlife Disease Study on a project designed to determine the prevalence (or presence) of *Trypanosoma cruzi* in wildlife species in Arizona and Georgia. *T. cruzi* is a protozoan parasite and is the causative agent for Chagas disease. Excess serum from skunks euthanized during ORV post-bait trapping campaigns and other operational projects was used for this collaboration. Three of 34 skunks, 1 of 4 raccoons, and 1 of 1 ringtail tested were sero-positive for *T. cruzi*. A presentation entitled “*Trypanosoma cruzi* in mesomammals from Arizona and Georgia” by Yabsley, EM Brown, K Wenning, and DM Roellig was presented at the Society for Tropical Veterinary Medicine 9<sup>th</sup> Biennial Conference in Merida, Mexico in June, 2006.

In addition, WS cooperated with USDA, APHIS, International Services employees to assist with the Mexico/U.S. Joint Vampire Bat capture exercise in San Luís Potosí, Mexico. Eight WS employees from the National Rabies Management Program, the National Wildlife Disease Program and the National Wildlife Research Center attended the exercise. Captured vampire bats were treated with vampiricide to assist with control efforts associated with the bovine paralytic rabies campaign.

### Hand Vaccination (Trap-Vaccinate-Release [TVR])

In a continuing effort to reduce the number of confirmed rabies cases in Arizona, and particularly Flagstaff, WS cooperated with the Centers for Disease Control and Prevention (CDC), the ADHS, the Coconino County Health Services (CCHS) and numerous local nuisance wildlife control operators to trap, vaccinate (using Rabdomun®, [Schering-Plough Animal Health Corporation, Summit, New Jersey, USA]), and release 186 animals

in 2006 (Figure 1). These hand vaccinated animals included: 162 striped skunks (plus 2 recaptures vaccinated twice), 23 raccoons (*Procyon lotor*), and 1 gray fox. Animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

A presentation summarizing the TVR efforts entitled “Trap-Vaccinate-Release to Manage Rabies Outbreaks in Striped Skunks in Flagstaff, Arizona, USA” was presented at the *Rabies in the Americas* conference in Brasilia, Brazil in October 2006.

### **Enhanced Surveillance**

Arizona WS personnel attended dRIT training in November 2005 at the CDC in Atlanta, Georgia and implemented the test on 13 April 2006. Wildlife Services tested 29 animals for field surveillance in 2006 including 24 feral/free-ranging dogs (*Canis familiaris*), 2 rabbits (*Sylvilagus audubonii*), 1 domestic cow (*Bos taurus*), 1 domestic goat (*Capra aegagrus hircus*) and 1 American badger (*Taxidea taxus*). All dRIT samples tested negative and 10% of the samples were sent to the CDC for confirmation. The CDC (using the dFA test) had 100% agreement with the WS dRIT results. All dRIT surveillance testing was conducted in Chinle, Arizona on the Navajo Nation. Wildlife Services will continue to use the dRIT in 2006 to enhance surveillance of suspect rabid animals in Arizona. Additionally, one Arizona WS biologist attended dRIT training in December 2006 at the CDC in Atlanta, Georgia.

### **RABIES-RELATED RESEARCH STUDIES 2006**

Rabies control in skunks (via ORV or other means) is not as well studied or documented as it is in raccoons, coyotes (*Canis latrans*), and foxes. Additionally, rabies control in feral/domestic dogs is most commonly achieved via hand vaccination by veterinarians or other trained professionals. Prior to 2005, an oral rabies vaccine had never been used in a feral dog population in the U.S. Consequently, WS conducted 5 rabies-related research projects in Arizona in 2005. Two of those studies were continued (skunk and feral dog ORV), the skunk telemetry study was completed, and a study on ORV titers in captive dogs is reported on for the first time in 2006. The 2 skunk studies occurred in Flagstaff and the 2 dog studies took place on the Navajo Nation in northeastern Arizona (Figure 1).

#### **Skunk Telemetry Study**

The unprecedented 2001 rabies outbreak in Flagstaff (and its re-emergence in 2004 and 2005) involving the spillover and perpetuation of a bat variant of the rabies virus in skunks led to many unanswered questions about skunk biology and ecology. In winter 2003-04, WS and the National Wildlife Research Center, in cooperation with the Northern Arizona University (NAU), began a behavioral research project to better understand striped skunk population dynamics for management and control of rabies outbreaks in Arizona. In 2006, WS completed the study, designed to answer the following questions:

- 1) What and where are urban skunk den locations and what are the urban skunks denning behaviors?
- 2) What is the interspecific contact at and around den locations?
- 3) Is rabies spread likely due to skunk-to-skunk interactions in dens and can skunks spread rabies to other wildlife or domestic animals at or around den sites?
- 4) What are striped skunk home range sizes in Flagstaff’s urban environment and do they overlap?
- 5) Could disease spread have been propagated by translocation?
- 6) What are Flagstaff’s urban skunk seasonal and daily movements?
- 7) What is striped skunk density within Flagstaff’s east and west urban matrices?

*Study Area.*--Flagstaff’s urban environment is naturally divided into an east and west region by Switzer Mesa (a mixed ponderosa and grassland environment). The eastern study site covered approximately 5.6 km<sup>2</sup> and was bound to the west by Switzer mesa, to the north by Mount Elden, and to the south by potential barriers to dispersal (Route 66, the Atchison Topeka and Santa Fe Railroad, and Interstate 40). The western study area covered approximately 5.5 km<sup>2</sup> and was bound to the west by Thorpe Park, to the east by Switzer Mesa, and to the south by Route 66 and the Atchison Topeka and Santa Fe Railroad (Figure 1).

*Methods.*--Striped skunks were live-trapped, marked with numbered ear tags, and fit with radio collars to determine home range sizes, den locations, and nocturnal and season movements. Den sites were monitored with automated, infrared 35 mm and digital cameras to document potential inter- and intraspecific contact at den sites.

*Results.*--This study began in winter 2003-04. Based on radio tracking data collected from January 2004-December 2005, we determined that skunk nightly rates of travel were highest during the post breeding season (May-July) and lowest during winter months (November-February). There was no difference between the sexes for nightly travel rates.

Home range sizes (determined by 95% kernel estimates) were largest in the spring and summer (1.3 km<sup>2</sup> for males and 1.1 km<sup>2</sup> for females) and smallest in the fall and winter months (0.7 km<sup>2</sup> for males and 0.4 km<sup>2</sup> for females). Amount of home range overlap also varied, ranging from 9-25% among males, 25-33% among females, and 26% among male-female pairs depending on the season. Because all animals in the environment were not radio collared, these overlap estimates are conservative. Comparing our results to those in published literature, we found that urban skunks have smaller home ranges and move less distance than those in rural areas. This could be due to a greater availability of food resources in urban areas.

Skunks used non-urban areas surrounding the urban study sites. Twenty of 40 radio collared animals were documented crossing the urban-wildland interface at least once during tracking efforts. Radio collared female skunks utilized the non-urban areas more often from March-July while males utilized them steadily throughout the year.

Twenty-one photo monitoring sites were used to document sequential and simultaneous use by skunks, other wildlife and domestic animals. Sixteen of the 21 sites had animals documented either sequentially or simultaneously. At one site we recorded 4 different skunks, 1 raccoon, and 1 domestic cat within a night while another site yielded 4 different skunks and 2 raccoons within a single night. Each of our sites was monitored for only 1-2 weeks, therefore these results provide only a limited perspective and likely underestimate the potential for intraspecific transfer of rabies among skunks and spill-over into other species within the urban environment.

From this study, we developed 4 basic management recommendations: 1) translocation of skunks by private or public animal control agencies appears to be the major means of disease spread from one part of the city to another and therefore should be strongly discouraged; 2) anthropogenic actions (i.e., placing pet food in areas accessible by wildlife or providing den sites under building or decks) should be discouraged since they attract both wildlife and domestic animals to the same location and may allow for transfer of rabies between individuals more rapidly; 3) many homeowners are reluctant to change their behaviors so rabies vaccination programs should focus on areas where anthropogenic actions occur; and 4) given that skunks move between urban and wildlife areas, vaccination programs should be performed in 2 pulses (separated by several weeks) to maximize animals encountering vaccine.

### **Skunk Oral Rabies Vaccine Study**

This study was first conducted in 2005 to field test an oral rabies vaccine and determine rabies titer levels in skunks that may have been exposed to the vaccine and skunks not exposed to the vaccine. This was the first field trial of its kind specifically targeting skunks. The study was conducted again in 2006.

*Study Areas.*--The 2006 ORV study area was an expansion of the 2005 ORV zone and included all of the major residential areas of Flagstaff, Arizona and the Fox Glen Continental area of Flagstaff where historical rabies outbreaks in the skunk population occurred (Figure 1). The approximately 45 km<sup>2</sup> study site also offered a mix of urban and wild land habitats.

*Methods.*--On 17 April 2006, approximately 30 individuals from WS, the CDC, the ADHS, the CCHS, the City of Flagstaff, and the NAU distributed 3,000 fishmeal coated sachet (CS) baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) by hand at a density of approximately 75 baits/km<sup>2</sup> throughout the ORV study area. The CS's contained microencapsulated tetracycline in the bait coating which served as a biomarker.

Five weeks post-bait distribution, WS began live-trapping skunks by placing cage traps in areas of suitable skunk habitat (e.g., travel corridors, natural washouts, culverts, etc.) throughout the study areas. Captured animals were anesthetized with a 5:1 ketamine:xylozine mixture injected intramuscularly. Blood was collected from each animal and serum samples were sent to the CDC to be analyzed for rabies virus neutralizing antibodies (VNA). Each animal was euthanized and the head sent to the ADHS laboratory to test the brain for the rabies virus using the

dFA test. The lower mandible of each captured animal was sent to Johnston Biotech (Sarnia, Ontario Canada) for tetracycline analysis and aging. Animals euthanized by WS in 2006 were done so in accordance with the 2000 American Veterinary Medical Association's Panel on Euthanasia recommendations.

*Results.*--Over the 2-year study, 54 striped skunks and 2 raccoons were captured from the vaccine study sites. Forty-six (85.2%) of the skunks and 1 raccoon had no detectable rabies antibody response (rabies titers <0.05 International Units [IU]), while the remaining 8 skunks (14.8%) and 1 raccoon had positive antibody responses (rabies titers  $\geq$ 0.05 IU). However, much of the antibody response could be attributed to concurrent hand vaccination efforts that took place in Flagstaff as part of the TVR efforts. All but 1 of the skunks with a detectable rabies antibody response ( $\geq$ 0.05 IU) had a record of recent hand vaccination with Rabdomun®. All 56 animals were confirmed negative for rabies using the dFA test.

Twelve of the animals tested had record of tetracycline biomarker, some with multiple lines visible indicating that multiple baits were encountered. However, of those 12 animals, only 2 had a positive antibody response and both of those animals had recent record of hand vaccination with Rabdomun®. Additionally, there were 7 animals with no record of the tetracycline biomarker, but with positive antibody responses. Given these results it was concluded that the current bait/vaccine formulation is not effective for field use for striped skunks in Flagstaff, Arizona. Therefore, WS will no longer continue this skunk ORV study in Flagstaff.

A presentation summarizing the results of this study without the biomarker data entitled "Effectiveness of Oral Rabies Vaccine (ORV) to Manage Rabies in Striped Skunks (*Mephitis mephitis*)" was presented at the *Rabies in the Americas* conference in Brasilia, Brazil in October 2006. A manuscript will be developed for publication in a peer-review journal in late 2007.

### **Feral Dog ORV Bait Study**

In April 2006, WS conducted a second field trial using CS baits containing Raboral V-RG® on feral free-ranging dogs on the Navajo Nation in conjunction with the CDC and the Navajo Nation Veterinary Services. This study followed an ORV placebo bait study on the Navajo Nation in April 2004. The 2004 study used 3 different baits with placebo vaccine (CS, fishmeal polymer [FMP], and dog food polymer [DFP]) to determine bait acceptance among feral dogs. Coated sachet baits were accepted more often than the other baits. More importantly, the placebo vaccine was taken nearly 4 times as often in CS than in FMP baits and twice as often in CS than DFP baits. The greater acceptance of placebo vaccine in CS baits during the 2004 study led to the initial ORV field trial in 2005 using CS baits containing Raboral V-RG® vaccine. This was the first field trial of its kind specifically targeting feral dogs in the U.S. The 2006 study was a continuation to field test an oral rabies vaccine and determine rabies titer levels in feral dogs that may have been exposed to the vaccine. In 2006, a microencapsulated tetracycline biomarker was added to the fishmeal coating of the bait to determine if dogs sampled during post-bait round-ups were exposed to the CS's.

*Study Areas.*--Sites in and around Chinle, Arizona on the Navajo Nation were chosen as the areas to distribute ORV baits (Figure 1 inset).

*Methods.*--Groups of 2-4 people, which included at least 1 representative from the Navajo Nation, hand baited as many dogs as possible. Each dog was presented a CS bait from a person's hand or a CS bait was tossed in a dog's direction if he was suspicious of eating out-of-hand. Personnel recorded numbers and sex of each dog, and how the dogs reacted to the bait: ignored the bait; took it and discarded it; swallowed it whole; chewed it but spilled the vaccine; or chewed it and received a vaccine dose. In total, 373 CS baits were distributed to 373 dogs (142 males, 103 females, 128 not recorded) with 276 of those dogs potentially receiving a vaccine dose.

Starting 6 weeks after baiting and continuing for 6 months post-ORV bait distribution, the Navajo Nation Animal Control (NNAC) collected 103 feral dogs during their routine dog control "round-ups" (all from the baited area). All the dogs were bled following euthanasia by the NNAC and serum samples from each dog were sent to the CDC to be analyzed for rabies VNA.

*Results.*--Serum samples were collected from 103 dogs rounded-up in the ORV baited area; 33 of the 103 samples (32.0%) had detectable levels of rabies antibodies with 10 having a titer >0.49 IU. Eighteen tooth/jaw samples were collected and analyzed for the presence of the tetracycline biomarker. Four of the 18 samples (22.2%) were positive, but only 1 of the positive biomarker samples corresponded to a positive antibody response. The 2006

serology results were more promising than the 2005 results where 5 of 40 samples (12.5%) had detectable levels of rabies antibodies.

### **Oral Rabies Vaccine Titer Study with Captive Dogs on the Navajo Nation**

In 2005, CS baits containing Raboral V-RG® vaccine were administered to 6 captive dogs to quantify the serological response following a single exposure to the vaccine. None of the animals developed a positive rabies antibody response (titer  $\geq 0.05$  IU) as determined by the RFFIT. In 2006 the study was expanded to quantify serological response following administration of the raccoon field dose of Raboral V-RG® over single and multiple days to a mixed population of captive dogs. Additionally, this study aimed to quantify the amount of human exposure risk to the vaccinia virus associated with animals recently exposed to Raboral V-RG®.

*Study Areas.*--All feral dogs were obtained during round-ups conducted by the NNAC from areas in and around Chinle, Arizona.

*Methods.*--To quantify the serological response following administration of CS baits containing Raboral V-RG®, 24 dogs were placed into one of four groups. Each group consisted of 6 dogs and received one of the following treatment regimes: Raboral V-RG® given once, Raboral V-RG® given twice (once each on 2 consecutive days), Raboral V-RG® given 3 times (once each on 3 consecutive days), and no treatment (control). Sera were collected from each animal at the start of the study and again on day 30 to determine rabies antibody levels. One dog in group 2 died of natural causes prior to the completion of the study so the sample size for that was decreased by 1 (n=5).

Whole blood was collected into a serum separator tube via vena puncture and centrifuged at a minimum of 2,000 rpm for 10 minutes to obtain serum. Sera were frozen immediately in a  $-70^{\circ}\text{C}$  where they were stored until shipped to the CDC for analysis. Because Raboral V-RG® contains live vaccinia virus, there is a potential for human exposure if the vaccinia persists in the oral cavity following administration of the CS bait. To determine how long vaccinia is present following administration of the sachet, each dog had its mouth swabbed beginning the day the last sachet was administered and continuing for 5 days.

Approximately 3 hours after administering the final Raboral V-RG® sachet, a swab of the animal's tonsils was taken. Swabs were collected using sterile cotton-tipped swabs which were placed in BHI media and frozen at  $-70^{\circ}\text{C}$ . Swabs were collected beginning the day of the final CS administration and continuing for a total of 5 days.

*Results.*--No dogs in group 1 (receiving 1 CS) developed detectable rabies VNA by day 30 after receiving the ORV. Three of 5 (60%) dogs in group 2 (receiving 2 CS's, 1 each on 2 consecutive days) developed detectable rabies VNA. Three of 6 (50%) dogs in group 3 (receiving 3 CS's, 1 each on 3 consecutive days) developed detectable rabies VNA. Results are still pending from the oral swabs.

Although more research is necessary, our results indicate that the raccoon field dose of Raboral V-RG® is ineffective at producing VNA in dogs after only a single dose. There is potential that multiple, consecutive doses could prove to be effective, however it is unlikely that feral/free-ranging dogs could be vaccinated successfully given the obstacles to administering vaccines to the same dogs on multiple days. However, should a product become available with a higher dosage of vaccine or different packaging, it could prove effective for use with feral/free-ranging dogs.

### **SUMMARY**

The Arizona WS program continues to be unique in that it is currently the only program specifically targeting skunks and feral dogs with ORV. In the coming year, WS anticipates continued ORV bait distribution to suppress rabies in the feral dog population on the Navajo Nation. WS will also begin new research projects to gain information on vaccine effectiveness with Mexican gray wolves (*Canis lupus baileyi*) and to better understand the rabies exposure rates experienced by free-flying bats throughout Arizona. Wildlife Services will continue to provide support and respond to requests for assistance with rabies surveillance and management in Arizona. Wildlife Services looks forward to continuing a strong cooperative relationship with state and local agencies, while providing federal leadership in wildlife rabies management.

**WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM  
CALIFORNIA 2006**

**BACKGROUND**

California has 2 variants of the rabies virus maintained in striped skunks (*Mephitis mephitis*) and bats (*Chiroptera* spp.). Rabies has remained enzootic in the skunk population since 1945. The skunk variant of rabies occurs in all areas of the state north of the Tehachapi mountain range and west of the Sierra Nevada crest (Figure 1). Rabies was first identified in a California bat in 1953 and has remained enzootic in the population since its detection. Rabies in raccoons (*Procyon lotor*), opossums (*Didelphis virginiana*), and various canids (*Canidae* spp.) occasionally occurs, but is not maintained in these species within the state. Rabies in these species likely represents a spillover from enzootic skunk or bat variants. From 1997-2006, the California Department of Health Services (CDHS) confirmed 2,788 cases of rabies in animals throughout the state (Figure 1).

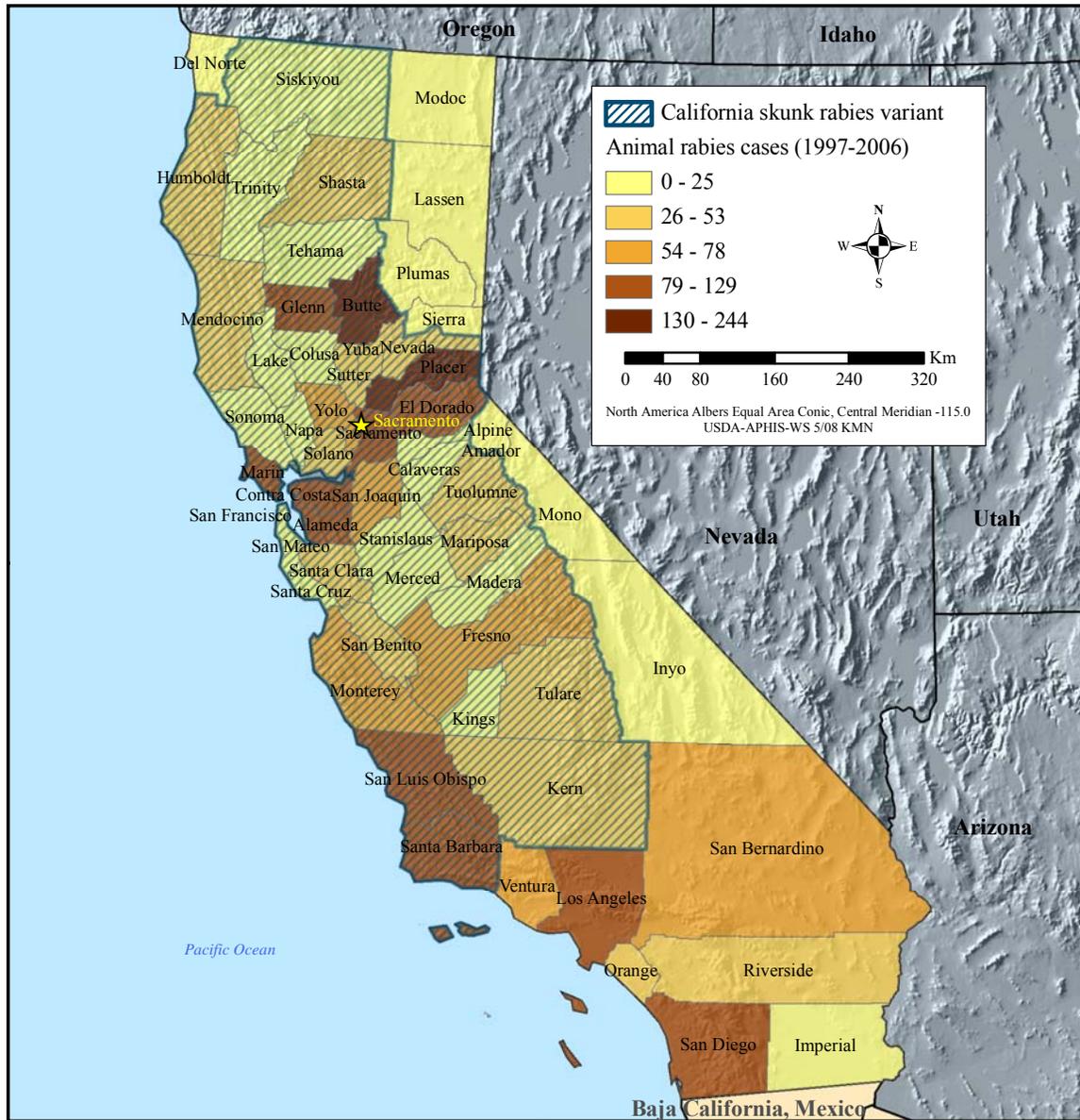


Figure 1. California skunk rabies variant distribution and animal rabies cases in California, 1997-2006.

## **RABIES MANAGEMENT PROGRAM 2006**

### **Bait Distribution**

Currently, there is no oral rabies vaccine (ORV) licensed for use in skunks in the United States. In 2004, Wildlife Services (WS), in conjunction with its National Wildlife Research Center (NWRC) conducted placebo ORV bait research projects. This was part of a larger project primarily conducted in Arizona, Louisiana, Texas, and Wyoming. The goal was to compare various formulations of placebo baits and determine bait acceptance levels in skunks. The optimal bait formulation derived from these studies may eventually be used to deliver an ORV to skunks. Several vaccines are currently being evaluated in the laboratory and may be ready for field testing in the future. California WS participated in these past bait trials in an effort to obtain U.S. Department of Agriculture licensing for ORV in skunks.

### **Enhanced Surveillance**

California WS removes animals from locations where human-animal interactions are high. These areas represent sites of potential human exposure to rabid animals where they occur. In 2006, WS personnel removed 4,447 skunks from throughout the state. Nearly 48% of the skunks removed were from 5 counties (Butte, El Dorado, Placer, San Luis Obispo, and Santa Barbara) identified as having a high prevalence of skunk rabies. All animals captured or taken by WS were evaluated for signs and symptoms of disease in the field prior to release or removal. Any animal exhibiting illness or unusual behavior was transported to the local health authority for testing. Unfortunately, there was no record kept of how many animals were submitted to local health laboratories, but WS did receive reports from some of these labs indicating that no rabies positives were detected in our samples. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

### **Rabies Laboratory Cooperation**

Rabies has been a legally required reportable disease under the California Code of Regulations, Title 17, Section 2500 since 1921. The CDHS, Veterinary Public Health Section is responsible for the surveillance, prevention, and control of rabies in California. During 2006, only animals that exhibited behavior/circumstances consistent with rabies or were directly involved in a potential exposure incident were tested for rabies by the CDHS. For a full listing of reported rabies by county and species in California for 2006 and the last 10 years please visit: <http://www.cdph.ca.gov/healthinfo/discond/Pages/rabies.aspx>

In 2006, the CDHS confirmed 201 cases of rabies in animals and 1 case in a human. The animal rabies cases included: 158 bats, 40 skunks, 2 foxes, and 1 horse (*Equus caballus*). The human case was identified in a patient who had recently emigrated from the Philippines and was associated with a dog bite received before coming to the U.S. (Blanton 2006).

In a 10-year period from 1997-2006, the CDHS reported 2,788 cases of animal rabies throughout the state. Infected skunks and bats represented 96.7% of all reported cases (1,065 and 1,630, respectively). Cases of rabies were also reported in: 43 foxes, 18 domestic/feral dogs (*Canis familiaris*), 15 domestic/feral cats (*Felis catus*), 5 opossums, 3 raccoons, 2 cows (*Bos taurus*), 2 horses, 2 wolf-dog hybrids, 1 goat (*Capra aegagrus hircus*), 1 rabbit (*Sylvilagus audubonii*), and 1 sheep (*Ovis aries*). The CDHS also reported 6 cases of rabies in humans during this same 10-year period. Bat variants of rabies have been involved in most cases of human rabies infection. Currently, 11 of 24 bat species in California are listed as State Species of Special Concern. Due to the special status of such a large number of bat species, WS refers bat nuisance calls to the California Department of Fish and Game.

Rabies cases by species and county in California for 2006 are within the average range of cases for the last 10-year period. Low numbers of cases in 10 counties may reflect a small sample size for submissions. Wildlife Services lacks an agreement to remove skunks in the majority of these counties.

## **SUMMARY**

Wildlife Services personnel in California remain vigilant for the signs and symptoms of disease in their wildlife management activities. In addition to skunk removal activities and the possibility of future ORV studies, WS continues to educate the public about safety when living or working near wildlife through technical assistance

and outreach activities. California WS has begun negotiations with the CDHS to increase the number and species of wild animals being tested for rabies. Specifically, WS is asking for testing of any unusually behaving animal. In addition, based on analysis by the CDHS, WS will provide samples from presumed healthy skunks and gray foxes (*Urocyon cinereoargenteus*) in select counties starting in 2007.

#### **LITERATURE CITED**

Blanton JD, CA Hanlon, and CE Rupprecht. 2007. Rabies surveillance in the United States during 2006. *Journal of the American Veterinary Medical Association* 231 (4):540-556.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM FLORIDA 2006

### BACKGROUND

In 1947, raccoon (*Procyon lotor*) rabies was first documented in Florida and is now considered enzootic statewide. During the late 1970s raccoon rabies was translocated by raccoon hunters from Florida to the mid-Atlantic States, where it began to spread throughout the eastern United States. In 1995, Pinellas County Animal Services initiated a county-wide oral rabies vaccination (ORV) program to combat an explosive outbreak of rabies in raccoons. This program continues today and over 600,000 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) have been distributed in the county since 1995 (Figure 1).

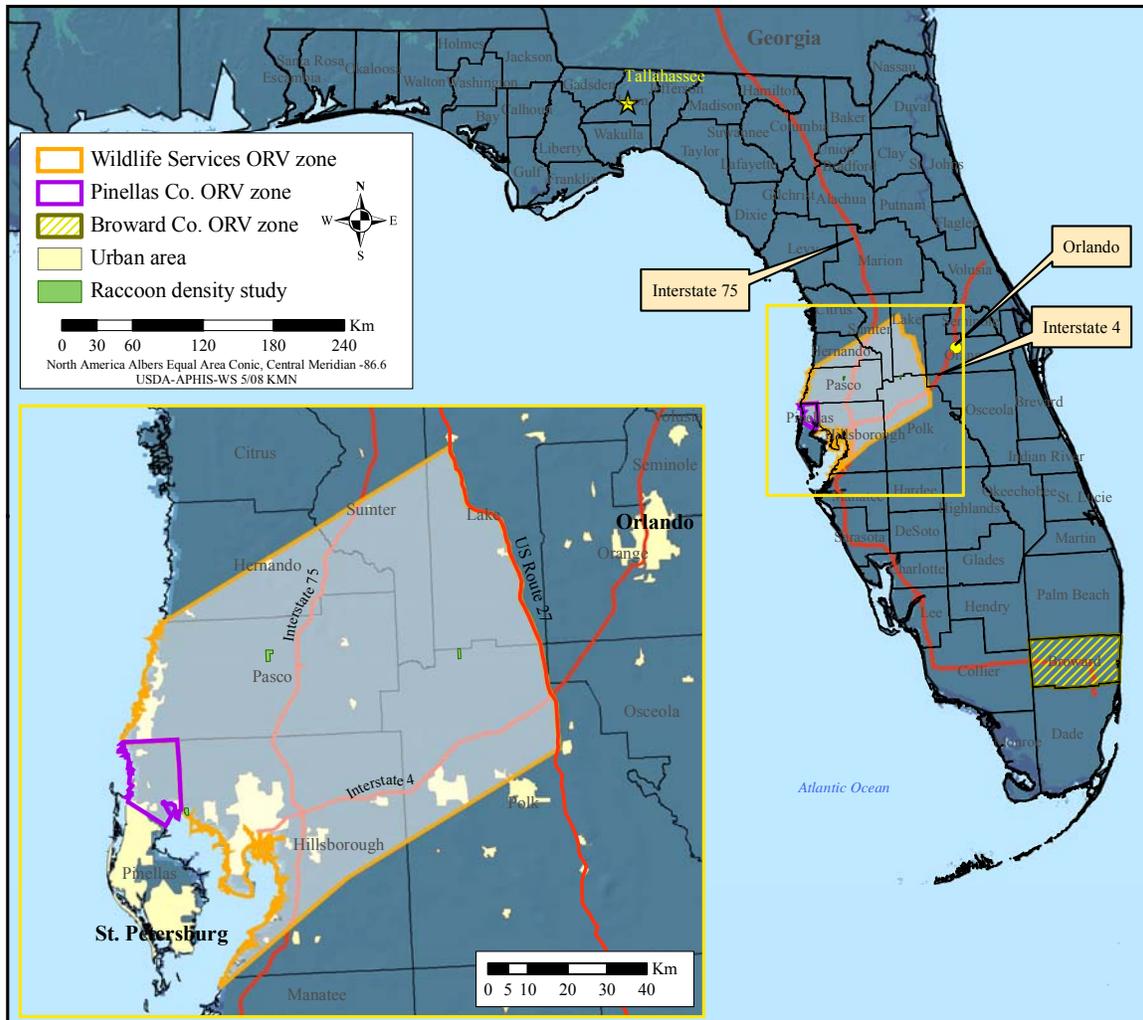


Figure 1. Wildlife Services cooperative rabies management program activities in Florida, 2006.

Wildlife Services (WS) began an ORV program on the Gulf coast in central Florida in 2003. The objective of the Florida WS Cooperative Rabies Management Program was to expand on the success of the Pinellas County ORV Program by establishing a vaccination zone in areas of high human population along the Interstate 4 corridor (Figure 1). The ORV program in Florida constitutes an important southern component in WS' National Rabies Management Program (NRMP).

In 2006, WS worked cooperatively with the Florida Department of Agriculture and Consumer Services (FDACS), Division of Animal Industry; the Florida Department of Health (FDH); the Florida Fish and Wildlife Conservation Commission; the Southwest Florida Water Management District; and the Florida Park Service on the

Florida ORV Program. In addition, WS gained the support of many county and city agencies to aid with the planning and the implementation of 2006 ORV bait distribution efforts.

## **ORV PROGRAM 2006**

### **Bait Distribution**

For the fourth consecutive year in 2006, WS participated in bait distribution efforts throughout central Florida; 663,528 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 7,318.1 km<sup>2</sup> (2,825.6 mi<sup>2</sup>). The ORV zone included portions of Hernando, Hillsborough, Lake, Pasco, Pinellas, Polk, and Sumter Counties. During 14-16 February 2006 bait distribution efforts, 196,018 fishmeal-coated sachet (CS) baits were distributed by fixed-wing aircraft, while 467,246 fishmeal polymer (FMP) baits were distributed by hand and by helicopter (16,920 of those in Pinellas County). Another 264 baits (144 CS and 120 FMP) were placed in bait stations in Pasco County.

In 2006, aerial bait distribution occurred at a rate of 100 baits/km<sup>2</sup> across the entire bait zone (Figure 1). Aircraft and pilots for the 2006 ORV program were provided by Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA). Helicopters and pilots were provided by Hillsborough County Mosquito Control, Pasco County Mosquito Control, and Polk County Sheriffs Department. Ground and aerial baiting support were provided by WS, the FDACS, and county and municipal agencies. Since its program inception in 2003, WS has distributed 2,468,756 ORV baits in Florida.

In 2005, a 5-year ORV program initiated by the Broward County Commission began as part of their overall Rabies Prevention Campaign to limit the spread of raccoon rabies in the county. The effort is led by the Broward County Animal Care and Regulation, and Parks and Recreation Divisions. Over 200,000 baits are distributed throughout the county annually (Figure 1), via ground and helicopter distribution. In 2006, WS provided video and demonstration training to Broward County officials on ORV bait handling and placement techniques for hand distribution.

### **Enhanced Surveillance**

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Florida WS personnel plan to attend dRIT training late in 2007 at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia.

### **Population Monitoring**

Wildlife Services conducted 3 raccoon density studies in 2006 using the NRMP standard protocol of 50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights; 1 each in Hillsborough, Pasco, and Polk Counties (Figure 1 inset). All studies coincided with post-ORV trapping. Blood samples were collected from 59 unique raccoons (Table 1) and sent to the CDC for rabies virus neutralizing antibody (VNA) testing. One raccoon was found dead near a trap during these studies. The raccoon was not tested for rabies. All remaining raccoons (58) were immobilized, processed and released. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

Table 1. Index to raccoon densities in Polk and Pasco Counties in Florida, 2006.

	Pasco	Hillsborough	Polk
Macrohabitat	Pasture	Tidal-influenced wetland	Pasture
Trap nights	500	500	500
Unique raccoons	4	33	22
Recaptured raccoons	0	3	1
Total raccoons	4	36	23
Trap success <sup>a</sup>	0.8%	6.6%	4.4%
Non-target captures	8	17	8
Area (km <sup>2</sup> )	3.64	1.64	1.93
Raccoon density index <sup>b</sup>	1.1	20.1	11.4

<sup>a</sup> Trap success = (unique raccoons ÷ trap nights) x 100.

<sup>b</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

### Post-ORV Monitoring

In April 2006, WS conducted post-ORV trapping throughout the ORV zone and collected 212 raccoon tooth and blood serum samples (in addition to the 59 samples collected during density studies). Tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA) for age and tetracycline biomarker analysis, while serum samples were sent to the CDC for rabies VNA testing. All raccoons (212) were immobilized, processed and released.

### ORV Bait Uptake Study

Florida WS started testing the uptake of FMP and CS baits in late 2005. These studies were replicated in early- to mid-2006. Tracking stations were designed with specifications from Rick Engeman of WS' National Wildlife Research Center in Fort Collins, Colorado to mimic the distribution of baits by fixed-wing aircraft. Each station was made by tilling up a 1 m<sup>2</sup> (10.8 ft<sup>2</sup>) piece of soil to remove dense ground vegetation. One ORV bait was placed in the middle of each sandy plot. Two study zones were established, 1 each on the east and west sides of I-75. The west study zone in 2006 consisted of 6 sites, each containing 24 stations, totaling 144 stations in this zone; while the east study zone consisted of 5 sites, each containing 24 stations, totaling 120 stations in this zone. (One of the east sites was removed from the study due to a vegetation project by the landowner.) Half of the stations in each zone were baited with FMP baits, while the other half were baited with CS baits. The stations in the zone west of I-75 were monitored for 1 week in February, while the stations east of I-75 were monitored for 1 week in June. Fishmeal polymer baits were taken more often than CS baits and raccoons took the most bait in both study zones (Table 2). These data were consistent with bait uptake data in the 2005 study as well. In 2006, all FMP baits were taken in the east study and raccoon numbers at the east bait stations were slightly higher than in the other studies. This could be explained by young of the year moving from the dens at this time of the year.

Table 2. Results of an oral rabies vaccine bait uptake study in Florida, 2006.

Bait type	Stations east of I-75		Stations west of I-75	
	FMP <sup>a</sup>	CS <sup>a</sup>	FMP	CS
Bait stations	48	72	72	72
Baits taken	48 (100%)	57 (79.2%)	65 (90.3%)	49 (68.1%)
Baits taken by raccoons	39 (81.3%)	43 (59.7%)	37 (51.4%)	32 (44.4%)

<sup>a</sup> FMP=fishmeal polymer; CS=coated sachet.

### Non-target Captures

In 2006, non-targets were marked with spray paint at the base of the tail, enabling WS personnel to identify the number of unique animals in an area. Non-target animals captured included: 69 opossums (*Didelphis virginiana*), 6 nine-banded armadillos (*Dasyus novemcinctus*), 5 domestic/feral cats (*Felis catus*), 1 gopher tortoise

(*Gopherus polyphemus*), and 1 Eastern fox squirrel (*Sciurus niger*). The 5 feral cats were euthanized during post-ORV sampling at landowners' requests.

### Rabies Laboratory Cooperation

Wildlife Services' ORV program in Florida cooperates with the FDH Laboratory (FDHL) and the CDC.

*Florida Department of Health Laboratory.*--The FDHL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by Animal Control Officers). The FDHL tested 3,594 animals for the rabies virus in 2006 (Table 3), representing a 5.1% decrease from the number of samples tested statewide in 2005. Animals were submitted from all 67 counties throughout the state, including the 7 ORV counties and 9 adjacent counties: Charlotte, Citrus, De Soto, Hardee, Highlands, Lee, Manatee, Osceola, and Sarasota. Of the animals tested statewide, 40.0% came from within or adjacent to the ORV zone, representing a 7.8% decrease from the number of samples tested within or adjacent to the ORV zone in 2005.

Raccoons, skunks (*Mephitidae* spp.), foxes, coyotes (*Canis latrans*), and bobcats (*Lynx rufus*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 81.1% of the animals tested for rabies in Florida in 2006 are reported by WS as "other." For a full listing of animals tested from Florida in 2006 by the FDHL please visit: <http://www.doh.state.fl.us/Environment/community/rabies/rabies-charts.htm>

Table 3. Animals tested for rabies by the Florida Department of Health Laboratory via the public health surveillance system in Florida, 2006.

	Statewide	Within and adjacent to Florida ORV zone
Raccoons	585	182 (31.1%)
Skunks	12	6 (50.0%)
Foxes	79	21 (26.6%)
Coyotes	3	1 (33.3%)
Bobcats	2	1 (50.0%)
Other <sup>a</sup>	2,913	1,227 (42.1%)
Total	3,594	1,438 (40.0%)

<sup>a</sup> Other animals included: alpaca, armadillos, bats, bears, cats, coatimundi, cougars, cows, deer, dogs, ferrets, goats, gophers, guinea pigs, horses, impalas, mice, monkeys, opossums, otters, panthers, pigs, rabbits, rats, rhinos, squirrels, and wolf-dog hybrids.

*Centers for Disease Control and Prevention.*--The CDC tests animal brainstems for rabies as part of enhanced surveillance (specimens not involved in an exposure and usually collected by WS). The CDC also analyzes wildlife blood serum samples (submitted by WS) for levels of rabies VNA.

Five enhanced surveillance brainstem samples from Florida WS were submitted to the CDC for rabies testing in 2006; all were feral cats from within the ORV zone that were not involved in a human or domestic animal exposure incident. All 5 cats tested negative for rabies.

The CDC also analyzed 276 blood serum samples for rabies VNA submitted by Florida WS in 2006. This represented a 63.9% decrease from the 626 samples submitted by WS in 2005. The Florida ORV program anticipates similar numbers of submissions to this laboratory in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/rabies/>

### ORV PROGRAM 2006 – EVALUATION

Florida's 2006 ORV bait distribution occurred in February and 2006 program evaluation data (serology, tetracycline, and age results) were available at the time of this report.

## Serology, Tetracycline Biomarker, and Age Results

Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

Serum samples from 271 raccoons were collected 5-13 weeks following the 2006 ORV bait distribution in Florida, which included 3 density studies (Table 4). Of these samples, 11.8% demonstrated a positive rabies VNA response. This was down from a 17.5% positive response following the 2005 ORV bait distribution. In addition, tooth samples were collected from 248 raccoons for tetracycline analysis (sent to Matson's Laboratory LLC). Of these samples, 11.7% indicated a presence of tetracycline. This was down from 28.6% presence of tetracycline following the 2005 ORV bait distribution.

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected during post-ORV and population monitoring evaluation in Florida, 2006.

	Post-ORV	Pasco	Hillsborough	Polk
Unique raccoon captures	212	4	33	22
<b>Serology</b>				
Testable blood samples	212	4	33	22
Positive rabies antibody response ( $\geq 0.05$ IU)	20 (9.4%)	0	4 (12.1%)	8 (36.3%)
<b>Tetracycline</b>				
Testable tooth samples	196	4	26	22
Presence of tetracycline biomarker	23 (11.7%)	2 (50.0%)	2 (7.7%)	2 (9.1%)

*Age Results.*--In 2006, 248 teeth were collected from raccoons during the 3 density studies and post-ORV trapping; however, only 219 were able to be aged (Figure 2). Yearlings and 2-year-old raccoons dominated the age class distribution in Florida.

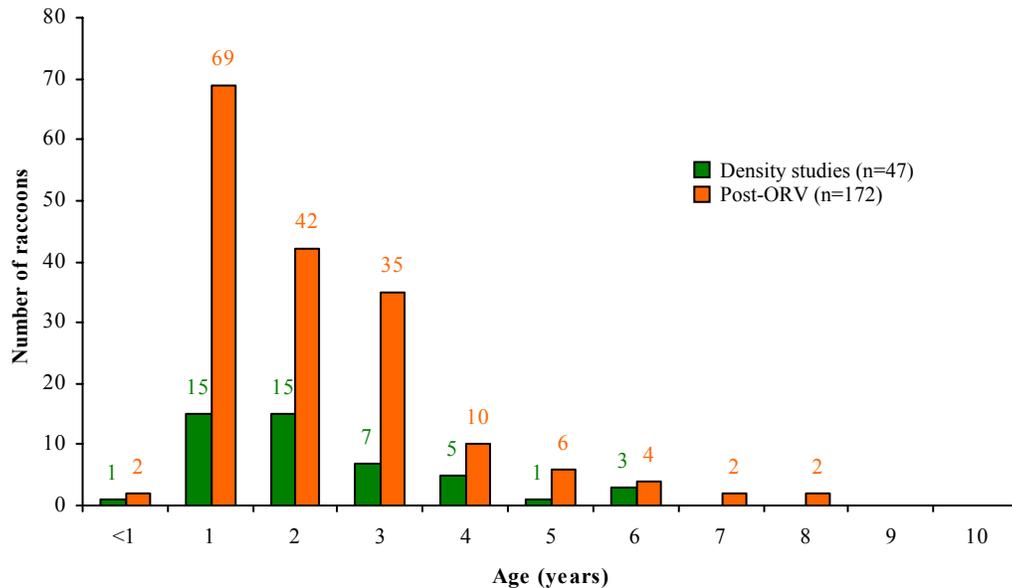


Figure 2. Age class distribution of 219 raccoon tooth samples collected during post-ORV trapping and density studies in Florida, 2006.

## **SUMMARY**

During 2006, WS completed its fifth year of cooperative participation in rabies management in Florida. The focus of activities this year was ORV bait distribution and resuming raccoon density studies in the bait zone area. Since WS' involvement in the Florida cooperative rabies management program began in 2003, over 2.4 million ORV baits have been distributed. In 2007, Florida's baiting efforts will continue to be an extension of the successful Pinellas County ORV program. Enhanced surveillance will be increased in the coming year.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM GEORGIA 2006

### BACKGROUND

Raccoon (*Procyon lotor*) rabies was first documented in Florida in 1947. The disease spread northward, entering South Georgia during the 1960s. Raccoon rabies is now enzootic throughout the state. The Georgia Wildlife Services (WS) oral rabies vaccination (ORV) program began in April 2003. Initially, through the use of enhanced surveillance, Georgia's program was designed to help determine the leading edge of the raccoon rabies variant within the state. During the summer of 2003, it was determined that the distribution of ORV baits would occur in Georgia during November 2003, forming the Georgia-Alabama-Tennessee (GAT) ORV zone (Figure 1).

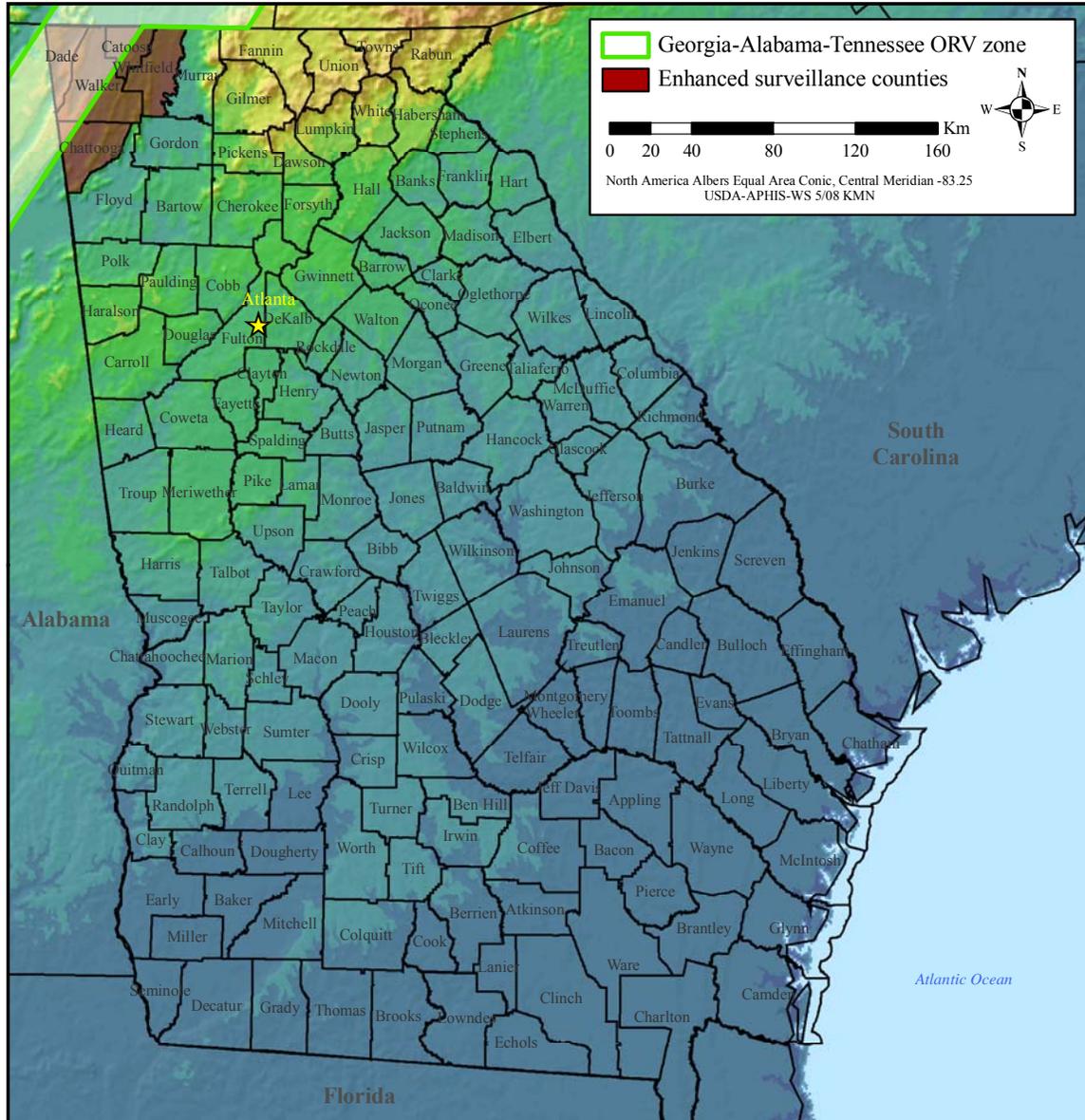


Figure 1. Wildlife Services cooperative rabies management program activities in Georgia, 2006.

Since that time, WS' major cooperators on the Georgia rabies management program have been the: Georgia Department of Human Resources Public Health Division (GDHR), the Georgia Department of Natural Resources, the Georgia Department of Agriculture, and the Centers for Disease Control and Prevention (CDC). In 2006, additional support on the local level was provided by Walker County Animal Control, and the City of Chickamauga

Police Department. These cooperators participated in various aspects of the ORV program including providing public information, assisting with aerial and ground bait distribution, and enhancing rabies surveillance.

## ORV PROGRAM 2006

### Bait Distribution

For the fourth consecutive year, WS participated in bait distribution efforts in northwestern Georgia; 96,528 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 1,443.7 km<sup>2</sup> (557.4 mi<sup>2</sup>). The ORV zone included portions of Catoosa, Chattooga, Murray, Walker, and Whitfield Counties, and all of Dade County (Figure 1). During 16-23 October bait distribution efforts, 69,528 fishmeal polymer (FMP) baits were distributed by fixed-wing aircraft, while 27,000 FMP baits were distributed by hand in areas too populated to bait by air.

Since its program inception in 2003, WS has distributed 385,644 ORV baits in Georgia.

### Enhanced Surveillance

In 2006, WS continued enhanced rabies surveillance in the northwestern part of the state by collecting 105 samples from road killed, abnormally behaving, and nuisance animals submitted by local animal control agencies in 5 counties (Figure 1). All animals were tested for rabies by Georgia WS using the Direct Rapid Immunohistochemistry Test (dRIT) and included the following species: raccoon, striped skunk (*Mephitis mephitis*), gray fox (*Urocyon cinereoargenteus*), bobcat (*Lynx rufus*), domestic/feral dog (*Canis familiaris*), domestic/feral cat (*Felis catus*), mink (*Mustela vison*), and red fox (*Vulpes vulpes*) (Table 1). None of the 105 animals tested were positive for the rabies virus.

Table 1. Animals collected for rabies testing by Wildlife Services in northwestern Georgia, 2006 (no rabies positives).

County	Raccoon	Skunk	Gray fox	Bobcat	Dog	Cat	Mink	Red fox	Total
Catoosa	6	2	1	1					10
Chattooga	5			1					6
Dade	19	1						1	21
Walker	45	3	1	1	1	1	2		54
Whitfield	12	2							14
Total	87	8	2	3	1	1	2	1	105

### Post-ORV Monitoring

In December 2006, WS conducted post-ORV trapping in Catoosa, Dade, and Walker Counties. Cage traps were used over 1,110 trap nights to capture 123 unique raccoons and collect tooth and blood serum samples from all of them. Nineteen raccoons were euthanized and 1 was found dead in a trap. All 20 raccoons tested negative for the rabies virus using the dRIT. All remaining raccoons (103) were immobilized, processed and released. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 65 opossums (*Didelphis virginiana*), 52 domestic/feral cats, 9 striped skunks, 1 eastern cottontail rabbit (*Sylvilagus floridanus*), and 1 eastern woodrat (*Neotoma floridana*). One gray fox was captured and euthanized.

### Rabies Laboratory Cooperation

Wildlife Services' ORV program in Georgia cooperates with the GDHR Public Health Laboratory (PHL) and the CDC.

*Georgia Department of Human Resources Public Health Laboratory.*--The PHL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure to the rabies virus). In 2006, the PHL tested 1,320 animal submissions and confirmed 268 cases of rabies statewide (154 raccoons [57%]), resulting in 310 human post-exposure treatments. The numbers of animals submitted from the 4 ORV counties are shown in Table 2. The 3 animals that tested positive for rabies within the ORV bait zone were located on the eastern edge of the zone. For more information on historical animal rabies cases in Georgia please visit: <http://health.state.ga.us/epi/disease/rabies.asp>

Table 2. Submissions to the Georgia Department of Human Resources Public Health Laboratory for rabies testing from counties treated with oral rabies vaccine in Georgia, 2006.

County	Submissions	Rabies Positive
Catoosa	6	0
Chattooga	11	0
Dade	0	0
Walker	32	3 (1 raccoon, 1 cat, 1 mule) 9.4%
Total	49	3 (1 raccoon, 1 cat, 1 mule) 6.1%

*Centers for Disease Control and Prevention.*--The CDC tests 10% of the negative and all of the positive animal brainstems that are tested by Georgia WS using the dRIT as part of enhanced rabies surveillance (specimens not involved in an exposure and collected by WS). The CDC also analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA).

In 2006, the CDC tested 11 negative wildlife brainstem samples submitted by Georgia WS for confirmation and validation of the dRIT results. Georgia WS also submitted 123 blood serum samples for rabies VNA analysis to the CDC in 2006. This represented an 8.2% decrease from the 134 samples submitted by WS in 2005. The Georgia ORV program anticipates similar numbers of brainstem and serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, some of the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; coated sachet (CS) baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

Serum samples from 134 raccoons were collected 4-7 weeks following the 2005 ORV bait distribution in Georgia. Of these samples, 40 (29.8%) demonstrated a positive rabies VNA response (titer  $\geq 0.05$  IU). This was up from a 25.4% positive response following the 2004 ORV bait distribution. In addition, 124 tooth samples were collected from raccoons and submitted to Matson's Laboratory LLC (Milltown, Montana, USA) for tetracycline biomarker analysis and aging. Biomarker analysis indicated that 28 (22.6%) of the teeth showed presence of tetracycline. Age results were not available at the time of printing.

Serum samples from 123 raccoons were collected 7-8 weeks following the 2006 ORV bait distribution in Georgia. Of these samples, 31 (25.2%) demonstrated a positive rabies VNA response. This was fairly consistent with the previous 2 years' antibody response following ORV bait distribution. In addition, 102 tooth samples were collected from raccoons and submitted to Matson's Lab for biomarker analysis and aging. Twenty-four (23.5%) of the teeth showed presence of tetracycline. Age results were not available at the time of printing.

## **SUMMARY**

During 2006, WS completed its fourth year of cooperative participation in rabies management in Georgia. Work emphasized ORV bait distribution, enhanced surveillance of raccoon rabies, and post-ORV monitoring and evaluation in northwestern Georgia. Since WS' involvement in the Georgia cooperative rabies management program began in 2003, nearly 400,000 ORV baits have been distributed and the program has continued to receive positive support from both cooperators and the general public.

Future ORV baiting strategies in Georgia will continue to be directed towards halting the spread of raccoon rabies into the western U.S. The Georgia ORV zone will continue to be tied to national and international planning efforts to contain the disease and explore strategies to eliminate the raccoon variant of the rabies virus from North America.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM KANSAS 2006

### BACKGROUND

Kansas has 1 terrestrial variant of the rabies virus in striped skunks (*Mephitis mephitis*) and another variant in bats (*Chiroptera* spp.). The South Central skunk variant is enzootic over nearly all of the state. In 2006, Kansas confirmed 83 cases of rabies in the state; striped skunks accounted for 53 (63.9%) of those cases. Despite only a 3.6% increase from the 80 statewide cases reported in 2005, the need for more information on rabies epidemiology in Kansas remains high. In 2003, the Kansas Wildlife Services (WS) program, Kansas State University College of Veterinary Medicine (KSUCVM) Rabies Lab and the Fort Riley Military Installation (FRMI) began a cooperative effort to collect information on the striped skunk population found on the FRMI in northeast Kansas (Figure 1).

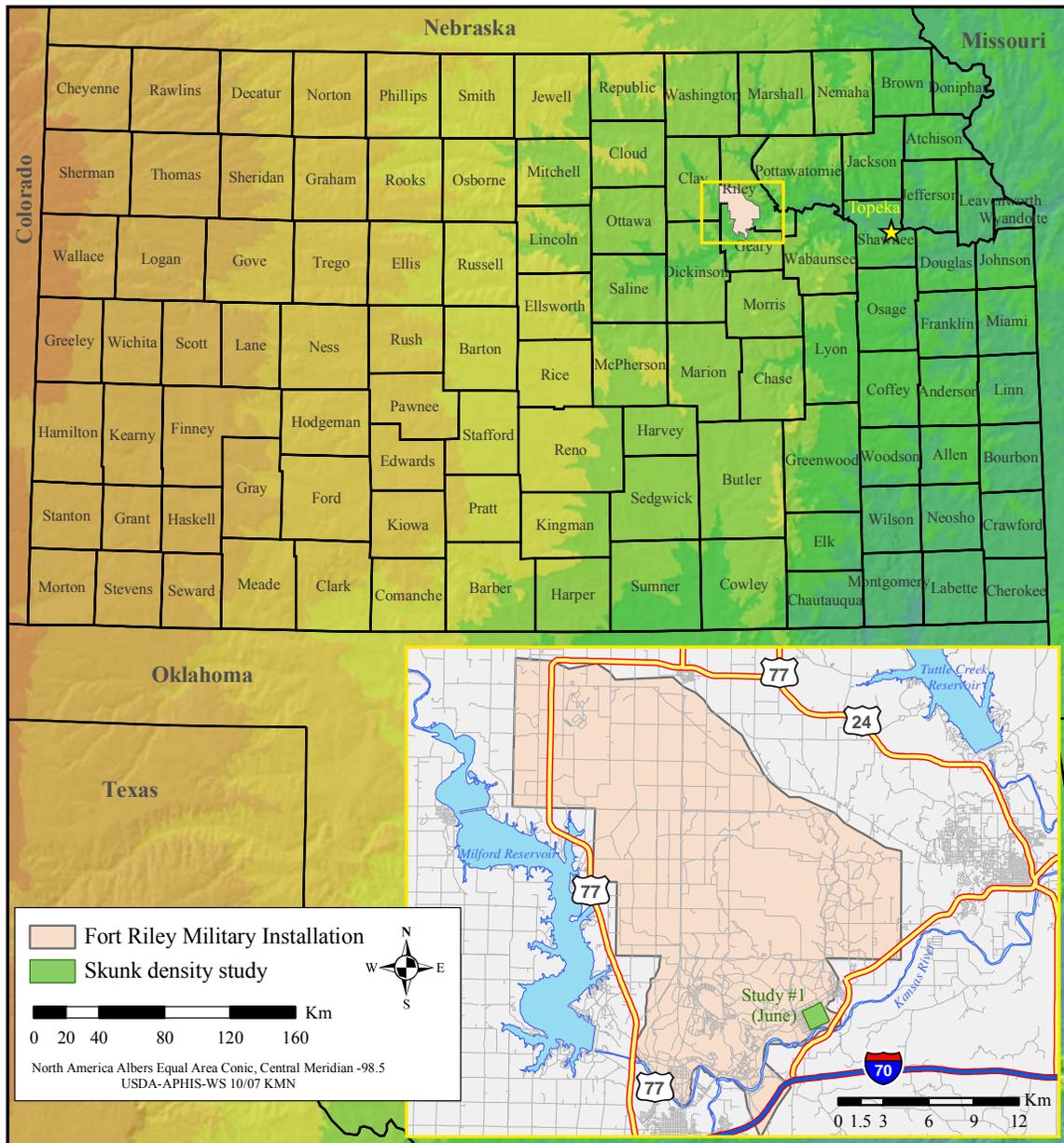


Figure 1. Wildlife Services cooperative rabies management program activities in Kansas, 2006.

Wildlife Services has historically had a full-time Wildlife Biologist stationed at the FRMI who regularly removed several skunks each year while responding to routine nuisance animal complaints on the installation. Due to WS' existing presence at the fort, and the nature of the land available to sample both urban and rural animal populations, the FRMI is an ideal location to monitor rabies and conduct density studies of striped skunks and raccoons (*Procyon lotor*). In 2006, the WS biologist was reduced to part-time due to Department of Defense (DOD) funding constraints allowing for only 1 skunk density study to be completed.

## **RABIES MANAGEMENT PROGRAM 2006**

### **Bait Distribution**

There is currently no oral rabies vaccination (ORV) bait distribution program in Kansas.

### **Enhanced Surveillance**

As nationwide research continues to find a more effective oral rabies vaccine for use in skunks, the Kansas WS program has focused much of its efforts on enhancing rabies surveillance to better define the distribution and prevalence of the virus on the FRMI. With a population of anywhere between 40,000-50,000 soldiers, families and civilians, the FRMI is considered the ninth largest "city" in the state. Citizens of Fort Riley report all nuisance animal problems including sick or strange acting animals to either the Military Police or a Public Works service order hotline. These reports are then funneled to the WS biologist stationed there. This arrangement allows for excellent indirect rabies surveillance. Due to the unique situation at the FRMI described above, and a reporting system already in place, the WS biologist stationed there has access to most potentially diseased animals. Over the last several years, WS has collected 1-3 rabid striped skunks annually.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Currently, WS is not planning training or implementation of the dRIT because the KSUCVM is meeting enhanced surveillance testing needs.

### **Population Monitoring**

Wildlife Services has conducted several striped skunk density studies in Kansas by modifying the National Rabies Management Program (NRMP) protocol for raccoon density studies. Each study has consisted of 50 cage traps set over a target study area of 3 km<sup>2</sup> and baited with dry cat food. Traps were not relocated during the 10-day studies (the raccoon protocol calls for traps to be relocated every 2-3 days if they have not captured a unique raccoon).

During 2003-2005, WS conducted 7 skunk density studies (3 in the rural training areas of Fort Riley and 4 in a heavily populated urban area on the FRMI). Indices to skunk density across the 7 studies were similar regardless of habitat type (approximately 2-3 skunks/km<sup>2</sup>). In 2006, WS conducted 1 skunk density study on a site with similar suburban habitats located near the Kansas River (Figure 1). This study was a repeat of one conducted in 2005. While 9 unique striped skunks were captured in 2006, 10 unique raccoons were also captured during the study (Table 1). All skunks and raccoons were euthanized in 2006 at the request of the FRMI. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations. The number of raccoon captures has been consistently high, relative to skunk captures, since studies began in 2003 (149 raccoons vs. 56 skunks). This could be due in part to the NRMP density study protocol, which was originally designed for raccoons. As skunk research and density studies continue, there may be a need for a dedicated skunk density protocol.

Table 1. Index to skunk density on the Fort Riley Military Installation, Riley County, Kansas, 2006.

	Study #1 (June)
Macrohabitat	Suburban
Trap nights	500
Unique skunks	9
Unique raccoons	10
Trap success of skunks <sup>a</sup>	1.8%
Non-target captures	5
Area (km <sup>2</sup> )	1.94
Skunk density index <sup>b</sup>	4.6

<sup>a</sup> Trap success = (unique skunks ÷ trap nights) x 100.

<sup>b</sup> Skunk density index (skunks/km<sup>2</sup>) = unique skunks ÷ area.

### Other Rabies Management Program Activities

There are hundreds of buildings on the FRMI that both humans and resident big brown bats (*Eptesicus fuscus*) occupy. Historically, nuisance bats were excluded and/or relocated from buildings and only bats involved in a human exposure were euthanized for rabies testing. In 2004, in cooperation with a DOD biologist, WS initiated a basic monitoring program to determine the prevalence of rabies in the local bat population on the FRMI. In 2005, WS continued to remove a select few bats from various buildings and euthanize them for rabies testing; there were no positives. Due to the overwhelming amount of negatives WS discontinued rabies testing of bats during 2006.

### Non-target Captures

Non-target animals captured and euthanized by WS in 2006 included: 2 opossums (*Didelphis virginiana*) and 3 feral cats (*Felis catus*). All animals were euthanized at the request of the DOD for nuisance complaint reasons.

### Rabies Laboratory Cooperation

The KSUCVM operates the state's public health rabies surveillance laboratory. They test animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by the public) and enhanced surveillance (specimens not involved in an exposure and often submitted by WS).

The KSUCVM Rabies Lab tested 1,210 animals for the rabies virus in 2006 from Kansas (Table 2); the Lab serves as the rabies testing facility for Nebraska as well. The number of samples tested in 2006 represented a 4.4% decrease from the number of samples tested in Kansas in 2005. Animals were submitted from 83 of 105 (79.0%) counties throughout the state, including Geary and Riley Counties (where the FRMI is located).

Skunks, raccoons, foxes (*Canidae* spp.), coyotes (*Canis latrans*), and bobcats (*Lynx rufus*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance in support of ORV. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 85.9% of the animals tested for rabies in Kansas in 2006 are reported by WS as "other." For a full listing of animals tested from Kansas in 2006 by the KSUCVM Rabies Lab please visit: [www.vet.ksu.edu/depts/rabies/kansas.htm](http://www.vet.ksu.edu/depts/rabies/kansas.htm)

Table 2. Animals tested for rabies by the Kansas State University College of Veterinary Medicine Rabies Lab in Kansas, 2006 (percent positive in parentheses).

	<b>Rabies Negative</b>	<b>Rabies Positive</b>	<b>Total Tested</b>
Skunks	31	53	84 (63.1%)
Raccoons	69	0	69 (0%)
Foxes	5	0	5 (0%)
Coyotes	11	1	12 (8.3%)
Bobcats	0	0	0 (0%)
Other <sup>a</sup>	1,011	29	1040 (2.8%)
Total	1,127	83	1,210 (6.9%)

<sup>a</sup> Other animals included: bat, beaver, camel, cat, chipmunk, cow, deer, dog, goat, gopher, guinea pig, horse, kangaroo, llama, mole, mouse, muskrat, opossum, rabbit, rat, rodents, sheep, shrew, squirrel, and woodchuck.

## SUMMARY

During 2006, WS completed its fourth year of cooperative participation in rabies management in Kansas, and specifically on the FRMI. Cooperators at Fort Riley and the KSUCVM continue to support the efforts made by WS to enhance rabies surveillance in skunks. In 2007, WS plans to upgrade the FRMI biologist to a 9-month position and resume rabies surveillance with 2 skunk density studies per year.

# WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM KENTUCKY 2006

## BACKGROUND

Kentucky has 1 terrestrial variant of the rabies virus in striped skunks (*Mephitis mephitis*) and another variant in bats (*Chiroptera* spp.). The North Central skunk variant is enzootic over most of the state. In 2002, Wildlife Services (WS) began participating in a cooperative rabies management program in Kentucky as an integral part of the National Rabies Management Program to stop the westward spread of the raccoon (*Procyon lotor*) variant of the rabies virus. In an effort to obtain baseline information on raccoon populations in Kentucky and actively search for raccoon rabies west of the Appalachian Ridge (AR) oral rabies vaccination (ORV) zone, WS began conducting raccoon density studies and enhancing rabies surveillance in the eastern counties bordering Ohio, Virginia, and West Virginia. Since 2002, WS has continued to collect road killed animals to enhance rabies surveillance in eastern Kentucky (Figure 1). To date, no positive cases of raccoon rabies have been documented in the state.

Activities conducted by WS are in cooperation with the Kentucky Department of Fish and Wildlife Resources (KDFWR), the Kentucky Department for Public Health (KDPH), and the Centers for Disease Control and Prevention (CDC).

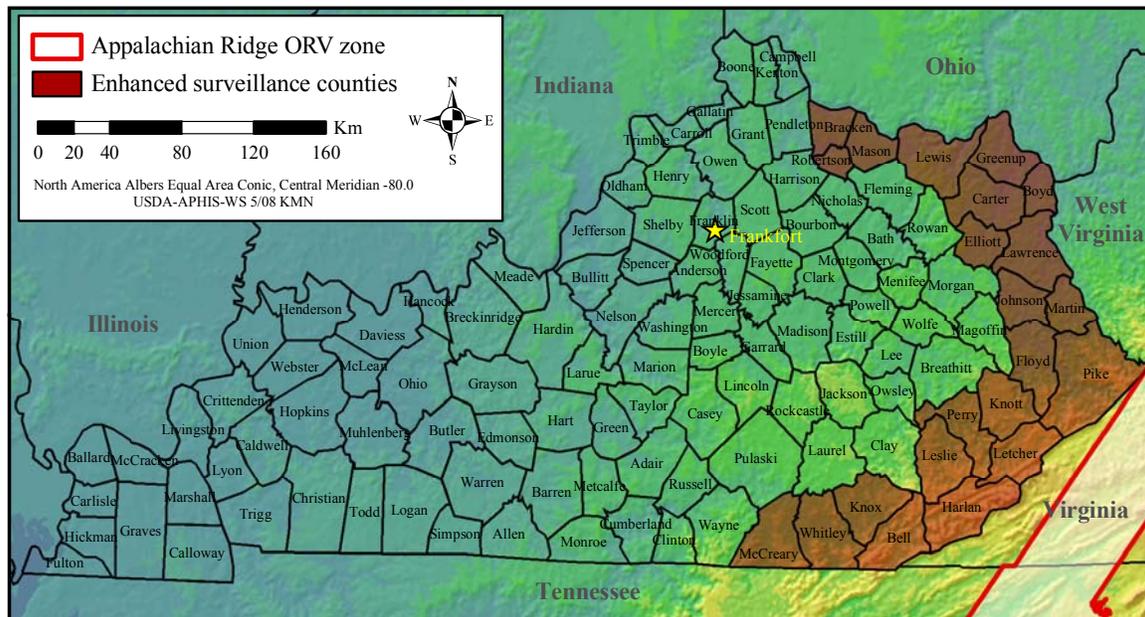


Figure 1. Wildlife Services cooperative rabies management program activities in Kentucky, 2006.

## RABIES MANAGEMENT PROGRAM 2006

### Bait Distribution

There is currently no ORV bait distribution program in Kentucky.

### Enhanced surveillance

In 2006, WS conducted enhanced surveillance for the detection of raccoon rabies in 22 counties in eastern Kentucky (Figure 1). This created a surveillance corridor along the borders with Ohio, West Virginia, Virginia, and Tennessee. In addition to cooperating with the KDFWR, WS personnel recruited state and local agencies to collect suspect animals for testing. An emphasis was placed on raccoons, striped skunks, gray foxes (*Urocyon cinereoargenteus*), red foxes (*Vulpes vulpes*), coyotes (*Canis latrans*) and bobcats (*Lynx rufus*) that exhibited strange behavior, were found dead in unusual places, or were fresh road killed animals. Nuisance and fur-trapped animals were also collected. Wildlife Services purchased small chest freezers and strategically located them within

14 of these counties for cooperators to store these suspect animals for testing. These efforts resulted in the collection and testing of 230 animals: 122 raccoons, 34 coyotes, 33 red foxes, 24 gray foxes, 9 bobcats, and 8 striped skunks. All samples tested negative for the rabies virus.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Kentucky WS personnel attended dRIT training in February 2006 at the CDC in Atlanta, Georgia. A Biosafety 2 (BSL-2) laboratory at the Louisville Metro Department of Health and Wellness was secured and fitted to conduct this test. During 2006, WS tested all 230 animals collected during enhanced surveillance efforts using the dRIT; they all tested negative. Ten percent of the negatives were sent to the CDC for confirmation. Using the dFA test, the CDC had 100% agreement with the WS dRIT results. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Kentucky.

### Rabies Laboratory Cooperation

Wildlife Services' rabies surveillance program in Kentucky cooperates with the KDPH, Division of Laboratory Services (DLS) in Frankfort, the Breathitt Veterinary Center (BVC) at Murray State University in Hopkinsville, and the CDC.

*Kentucky Department for Public Health, Division of Laboratory Services and the Breathitt Veterinary Center.*--Both the DLS and the BVC are responsible for testing animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure). Positive samples from both labs are then sent to the CDC for rabies variant typing.

The DLS and BVC tested 1,126 animal brainstem samples for the rabies virus in 2006 (Table 1). This represents a 20.4% increase from the number of samples tested in 2005. The 2006 samples were submitted from 114 of 120 counties in Kentucky and from 15 counties within the designated enhanced rabies surveillance zone.

Raccoons, skunks, foxes, coyotes, and bobcats are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 86.9% of the animals tested for rabies in Kentucky in 2006 are reported by WS as "other." For more information on rabid animals by county in 2006 please visit: <http://chfs.ky.gov/dph/epi/rabies.htm>

Table 1. Animals tested for rabies by the Kentucky Department for Public Health, Division of Laboratory Services and the Breathitt Veterinary Center at Murray State University in Kentucky, 2005.

	Statewide	Within Wildlife Services' enhanced rabies surveillance zone
Raccoons	95	3 (3.2%)
Skunks	26	0
Foxes	15	0
Coyote	12	0
Bobcats	3	2 (66.7%)
Other <sup>a</sup>	975	59 (6.1%)
Total	1,126	62 (5.5%)

<sup>a</sup> Other animals included: alpaca, bats, beaver, cats, cattle, chipmunks, deer, dogs, ferret, goats, groundhogs, hamsters, horses, mink, moles, mice, muskrats, opossums, rabbits, rats, squirrels, and a wolf.

*Centers for Disease Control and Prevention.*--The Kentucky rabies surveillance program did not submit brainstem samples to the CDC in 2006 due to implementation of the dRIT; WS expects this to be the case in 2007 as well. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## **SUMMARY**

During 2006, WS completed its fifth year of participation in cooperative rabies management efforts in Kentucky. The focus of activities continued to be collection of road killed or suspect rabid animals to enhance rabies surveillance west of the existing AR ORV zone (Figure 1).

In 2007, WS will increase enhanced surveillance for the raccoon variant of rabies in eastern Kentucky by continuing to recruit state and local entities to collect unusual acting and road killed animals. Surveillance efforts will continue to be concentrated in 22 counties bordering Ohio, West Virginia, Virginia, and Tennessee.

# WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM LOUISIANA 2006

## BACKGROUND

To date, there have been no reported cases of the raccoon (*Procyon lotor*) variant of rabies in Louisiana, although other variants including bat (*Chiroptera* spp.) and striped skunk (*Mephitis mephitis*) do occur. In an effort to detect possible entry of raccoon rabies into the state, Wildlife Services (WS) conducted surveillance of road killed and trapped animals in Washington and St. Tammany Parishes during April, May, and June of 2006 (Figure 1).

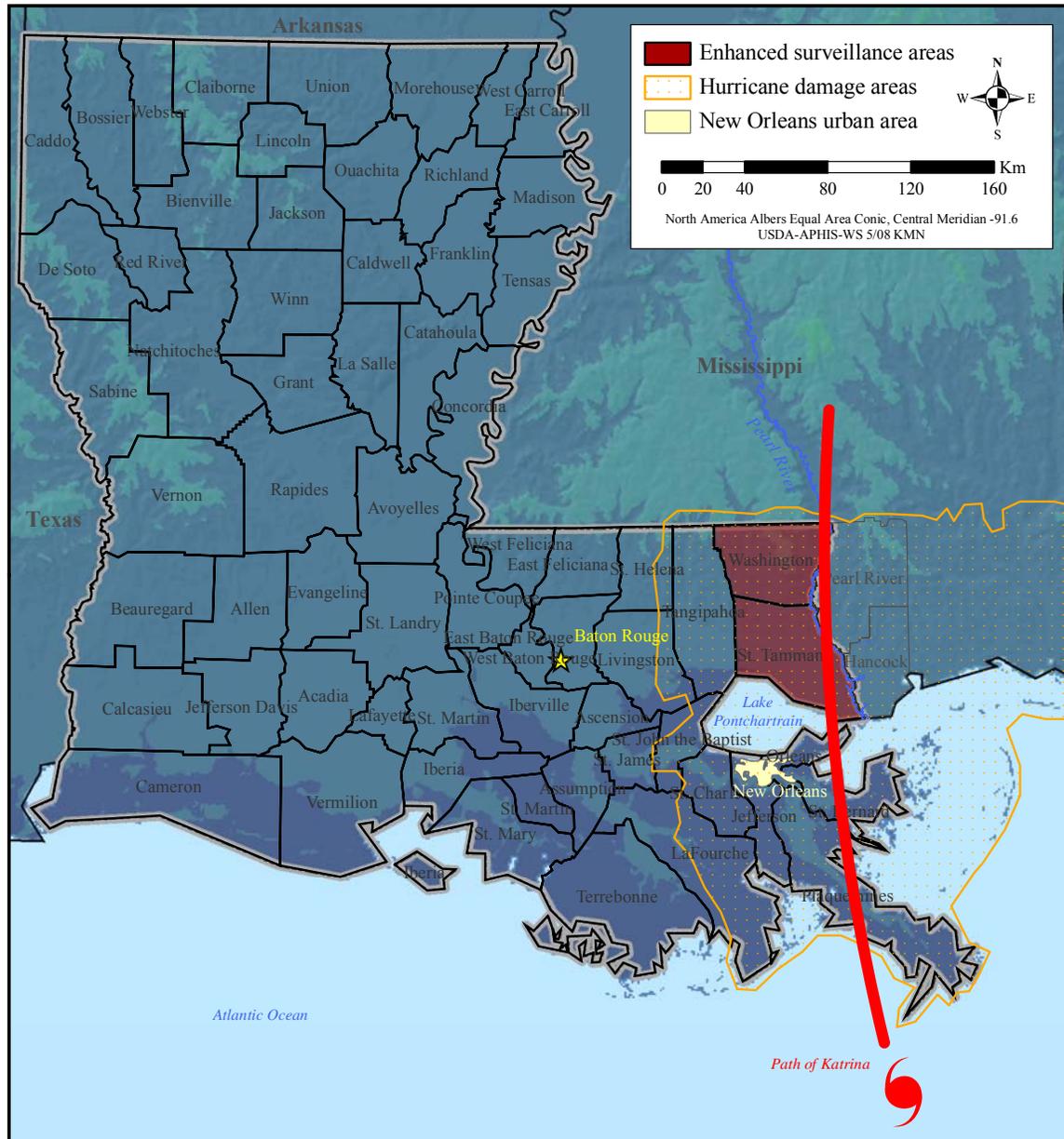


Figure 1. Wildlife Services cooperative rabies management program activities in Louisiana, 2006.

## RABIES MANAGEMENT PROGRAM 2006

### Bait Distribution

There is currently no oral rabies vaccination (ORV) bait distribution program in Louisiana.

### Enhanced Surveillance

From April-June 2006, a WS specialist completed road and highway surveys in Washington and St. Tammany Parishes to collect road killed raccoons and other small carnivores to enhance rabies surveillance. During those surveys, no road kills were collected by WS, but St. Tammany Parish Animal Control (STPAC) collected 29 raccoons and 1 gray fox (*Urocyon cinereoargenteus*), mostly from urban nuisance projects. Very few road killed raccoons were observed and none were processed due to advanced deterioration or excessive damage. Surveillance was suspended at the end of June due to a lack of success trapping and virtually no raccoon sign (possibly due to the combination of a distemper outbreak and hurricane damage from 2005). In addition to animals collected from STPAC, 4 raccoons caught on a damage management project in Ouachita Parish were also collected. All 34 animals were tested by WS using the Direct Rapid Immunohistochemistry Test (dRIT) and they were all negative for rabies. Four samples (11.8%) were sent to the CDC for confirmation in August 2006. Louisiana WS will continue to use the dRIT in 2007 and all positives, 10% of all negatives, and all indeterminate samples will be sent to the CDC for confirmation and strain typing. Additionally, Louisiana WS plans to have 2 more personnel trained in dRIT procedures.

### Rabies Laboratory Cooperation

Louisiana regulations require that testing for rabies be conducted when human exposures are involved. Testing for other exposures is not mandatory. The Louisiana Department of Health and Hospitals (LDHH) is responsible for carrying out mandates related to required rabies testing. The LDHH and WS have expressed interest in continued and expanded cooperation on rabies issues; however, the agencies do not jointly conduct any rabies projects at this time.

During 2006, the LDHH tested 678 animals for the rabies virus (Table 1). Seven animals (2 skunks and 5 bats from outside of WS' enhanced rabies surveillance zone) tested positive. These rabid animals did not have the raccoon variant of the rabies virus.

Raccoons, skunks, foxes (*Canidae* spp.), and coyotes (*Canis latrans*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 94.0% of the animals tested for rabies in Louisiana in 2006 are reported by WS as "other." For more information on historical rabies cases in Louisiana please visit: <http://www.dhh.louisiana.gov/offices/page.asp?id=249&detail=7488>

Table 1. Animals tested for rabies by the Louisiana Department of Health and Hospitals for public health surveillance in Louisiana, 2006.

	Statewide	Within Wildlife Services' enhanced rabies surveillance zone <sup>a</sup>
Raccoons	29	5 (17.2%)
Skunks	7	0
Foxes	3	0
Coyotes	2	0
Other <sup>b</sup>	637	43 (6.8%)
Total	678	48 (7.1%)

<sup>a</sup> St. Tammany and Washington Parishes.

<sup>b</sup> Other animals included: bats, cats, cattle, dogs, ferrets, goats, horses, minks, moles, opossums, rodents, sheep, and squirrels.

## **SUMMARY**

In 2006, WS completed its fourth year of participation in cooperative rabies management efforts in Louisiana by continuing enhanced rabies surveillance work in 2 gulf coast parishes. A rabies specialist, using the dRIT, managed to test 30 usable samples from the enhanced surveillance zone despite the lack of target species' presence and ongoing hurricane recovery. Surveillance was not conducted in Pearl River and Hancock Counties in Mississippi due to ongoing hurricane recovery (Figure 1).

Wildlife Services plans for 2007 include: continued road and highway surveillance, intensifying trapping/reconnaissance efforts to locate and sample possible concentrations of raccoons, training 2 more individuals in the dRIT procedures, and hiring a Wildlife Disease Biologist to coordinate and intensify surveillance efforts. These planned activities should aid in the effort to detect possible entry of the raccoon variant of rabies into Louisiana and southwestern Mississippi.

**WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM  
MAINE 2006**

**BACKGROUND**

The raccoon (*Procyon lotor*) variant of the rabies virus was first confirmed in a striped skunk (*Mephitis mephitis*) in 1994 (York County) and has been enzootic in wildlife populations in Maine since 1995 (Table 1). Since then, the disease has progressed steadily from southern Maine to central Maine and continues to slowly progress north. During 2006 there were 127 cases of rabies confirmed in Maine (Figure 1) and over the last decade, raccoons have consistently accounted for approximately half of all rabies cases in the state (Table 1). To reduce the spread of rabies, Wildlife Services (WS) continues efforts to vaccinate targeted raccoon populations in eastern Maine and monitor the disease through enhanced rabies surveillance along the front line of documented rabies cases. Since 2003, WS has been able to maintain an oral rabies vaccination (ORV) zone connected to the New Brunswick, Canada trap-vaccinate-release (TVR) zone as a barrier to the advancement of rabies across the U.S.-Canada border (Figure 1).

Table 1. Confirmed positive rabies cases in Maine, 1994-2006.

Year	Total rabies cases (all species)	Rabid raccoons
1994	10	0
1995	43	41 (95%)
1996	131	53 (40%)
1997	244	130 (53%)
1998	248	143 (58%)
1999	208	116 (56%)
2000	139	73 (53%)
2001	85	34 (40%)
2002	67	37 (55%)
2003	82	38 (46%)
2004	69	37 (54%)
2005	61	37 (61%)
2006	127	59 (46%)
Total	1,514	798 (53%)

**ORV PROGRAM 2006**

**Bait Distribution**

Maine WS participated in ORV baiting activities for the fourth consecutive year in 2006 by distributing 49,680 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) over 747.3 km<sup>2</sup> (288.5 mi<sup>2</sup>) of northeastern Maine (Figure 1). On 24 August 2006, baiting activities were based out of the Millinocket Municipal Airport in Millinocket, Maine and 49,440 fishmeal polymer (FMP) baits were distributed by air, while 240 FMP baits were distributed by hand within the Danforth and Vanceboro city limits. Fixed-wing aircraft were provided by the Ontario Ministry of Natural Resources, while WS personnel served as navigators and flight crew in the planes. Ground baiting was provided by WS. Since its program inception in 2003, WS has distributed 469,017 ORV baits in Maine.

The 2006 ORV zone included portions of Penobscot and Washington Counties and averaged 16 km (10 mi) wide and 40 km (25 mi) long along the Maine-New Brunswick, Canada, border. Maine’s ORV zone is a northern continuation of New Brunswick’s TVR zone. Together, the international effort to help stop the northward and eastward spread of rabies spans approximately 100 continuous miles (160 km) of the U.S.-Canada border. Raccoon rabies was first confirmed in neighboring New Brunswick in 2001, but since March 2002 New Brunswick has maintained a “rabies-free” status due to the implementation of a large-scale TVR program. To date, this program has resulted in more than 4,000 raccoons and striped skunks being trapped, vaccinated, and released in New Brunswick. The documented movement of rabies provides Maine an important role in helping to prevent the re-introduction of raccoon rabies into New Brunswick, Canada.

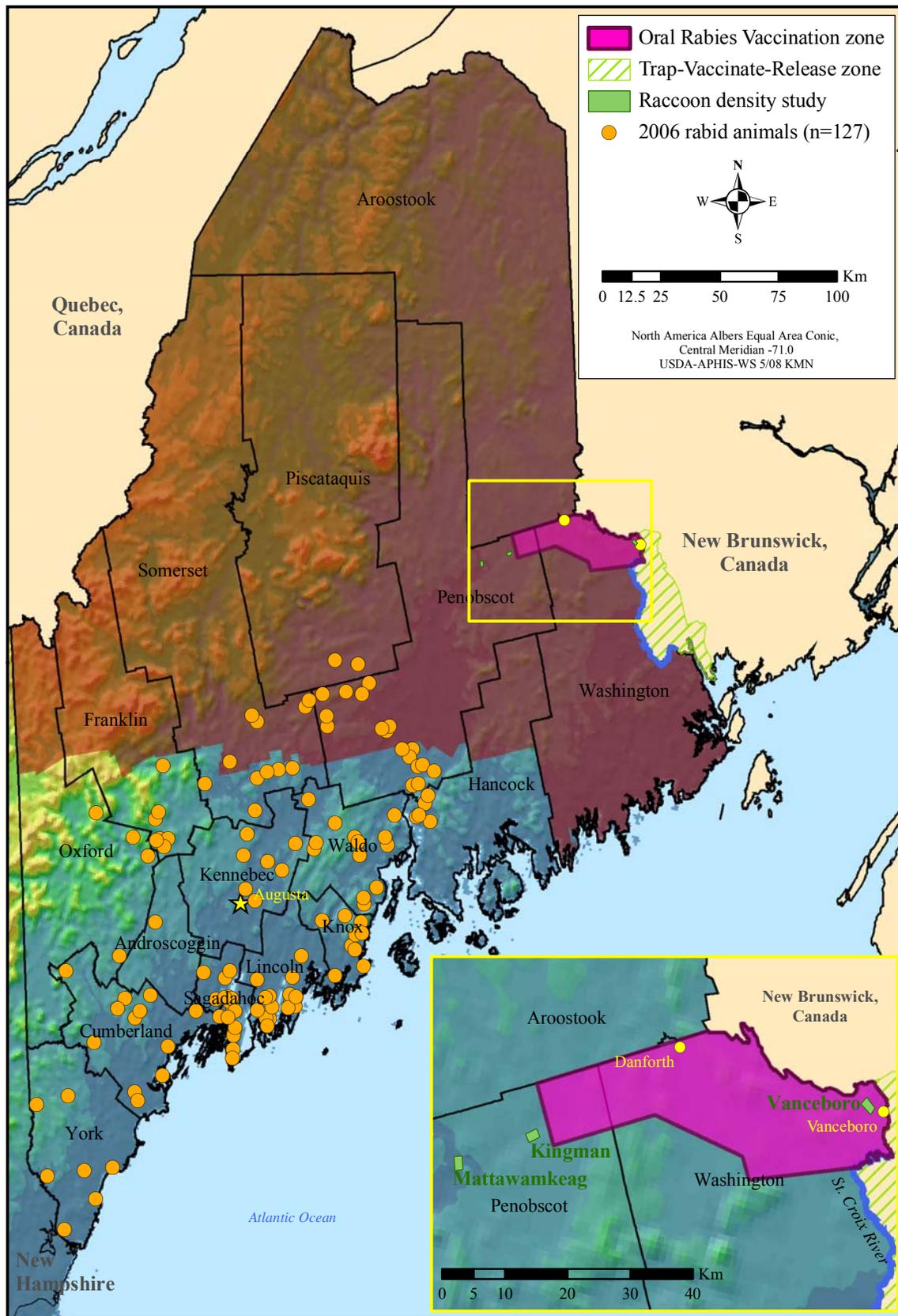


Figure 1. Wildlife Services cooperative rabies management program activities in Maine, 2006.

## Enhanced Surveillance

In 2006, Maine WS continued to conduct enhanced rabies surveillance throughout northern Maine and along the Canadian border in areas where raccoon rabies was emerging or had not yet been documented. Surveillance methods included road kill surveys and the collection of sick or suspicious acting animals that had no contact with humans or domestic animals. During 2006, WS provided logistics and funding to collect and submit 44 animals (33 raccoons, 6 striped skunks, 3 red foxes [*Vulpes vulpes*], 1 muskrat [*Ondatra zibethicus*], and 1 woodchuck [*Marmota monax*]) from northern Maine to the Maine Department of Health and Human Services, Health and Environmental Testing Laboratory (MDH HETL). All 44 animals tested negative for rabies.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turn around and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

In addition to the 44 enhanced surveillance animals submitted to the MDH HETL, WS collected and tested 18 animals (12 raccoons, 4 skunks, 1 red fox, and 1 moose [*Alces alces*]) using the dRIT. One skunk tested positive for rabies using the dRIT. The skunk and 10% of all negatives were sent to the CDC for confirmation and strain typing. The CDC (using the dFA test) had 100% agreement with the WS dRIT results for positive and negative samples. They also typed the positive skunk as raccoon rabies variant. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Maine.

## Population Monitoring

From 2002 to 2005, WS conducted 9 raccoon density studies in northeastern Maine using the National Rabies Management Program (NRMP) standard protocol (50 live traps over a target study area of 3 km<sup>2</sup> for 10 consecutive nights). During those studies, the index to raccoon density averaged 4 raccoons/km<sup>2</sup> (with a low of 1 raccoon/km<sup>2</sup> in Codyville to a high of 6 raccoons/km<sup>2</sup> in Hodgdon, both in 2002). The NRMP low density protocol (which involves 5 consecutive nights of trapping instead of 10) was established in 2005 to eliminate futile effort in areas where raccoon densities are believed to be  $\leq 1$  raccoon/km<sup>2</sup>.

In 2006, WS conducted 3 raccoon density studies in the towns of Mattawamkeag, Kingman, and Vanceboro, Maine (Figure 1 inset). All 3 studies were conducted during the summer prior to annual ORV bait distribution and the indexes to density were similar to previous studies in Maine, ranging from approximately 1-3 raccoons/km<sup>2</sup> (Table 2). The Mattawamkeag and Kingman studies were conducted just west of the current bait zone in ORV naïve areas (areas that had not previously been treated with ORV). During the 3 studies, 20 raccoons and 8 skunks were trapped, processed, and released. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

Table 2. Index to raccoon densities in 3 northern Maine townships, 2006.

	<b>Mattawamkeag</b>	<b>Kingman</b>	<b>Vanceboro</b>
Time of study	18-28 July	18-28 July	15-20 Aug.
Macrohabitat	Forested	Forested	Forested
Target trap nights	500	500	250
Unique raccoons	10	8	2
Recaptured raccoons	3	3	0
Non-target captures <sup>b</sup>	20	6	1
Area (km <sup>2</sup> )	2.96	2.77	2.98
Raccoon density index <sup>a</sup>	3.4	2.9	0.7

<sup>a</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

<sup>b</sup> Non-target captures include skunks (Mattawamkeag=3; Kingman=4; Vanceboro=1).

## Post-ORV Monitoring

During 25 September and 13 October 2006, WS conducted post-ORV trapping in Penobscot and Washington Counties. Cage traps were used over 2,073 trap nights to capture 92 raccoons and 22 skunks. All raccoons and skunks were immobilized, processed and released, and tooth and blood serum samples were collected from each animal.

## Non-target Captures

Non-target animals captured and released by WS in 2006 included: 30 domestic cats (*Felis catus*), 7 fishers (*Martes pennanti*), 5 red squirrels (*Tamiasciurus hudsonicus*), 2 snowshoe hares (*Lepus americanus*), and 1 pine marten (*Martes americana*).

## Rabies Laboratory Cooperation

Wildlife Services' ORV program in Maine cooperates with the MDH HETL and the New York State Department of Health's Rabies Laboratory at the Wadsworth Center (WC).

*Maine Department of Health and Human Services, Health and Environmental Testing Laboratory.*--The HETL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure) at no cost to the public. The HETL has provided in-kind services to test enhanced rabies surveillance samples (specimens not involved in an exposure and submitted by WS) over the last 5 years. The lab is available 24 hours a day for specimen drop off and results are routinely available the same day the test is conducted. The HETL tested 812 animals statewide in 2006 and confirmed 127 cases of rabies in Maine (Table 3).

Raccoons, skunks, foxes, and coyotes (*Canis latrans*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 74.6% of the animals tested for rabies in Maine in 2006 are reported by WS as "other." For more information on current and historical animal rabies cases in Maine (by year, county, town, and species) please visit:

<http://www.maine.gov/dhhs/etl/rabies/rabies.htm>

Table 3. Animals tested for rabies by the Maine Department of Health and Human Services, Health and Environmental Testing Laboratory, 2006.

	Number tested	Number rabies positive
Raccoons	115	59 (51.3%)
Skunks	73	43 (58.9%)
Foxes	16	2 (12.5%)
Coyotes	2	0
Other <sup>a</sup>	606	23 (3.8%)
Total	812	127 (15.6%)

<sup>a</sup> Other animals included: bats; beaver; bobcat; chipmunk; deer; ermine; fisher; mink; mole; mouse; muskrat; opossum; otter; porcupine; rabbit; rat; squirrel; weasel; woodchuck; and domestic alpaca, cats, cows, dogs, donkey, ferret, goat, hamster, horse, lamb, llama, pig, rabbit, sheep, and wolf hybrid.

*New York State Department of Health's Rabies Laboratory at the Wadsworth Center.*--The WC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA). In 2006, Maine WS submitted 134 blood serum samples (112 raccoons and 22 skunks) for rabies VNA analysis to the WC. The Maine ORV program anticipates similar numbers of serum sample submissions to the WC in 2007.

The timeliness of test results from both laboratories enhances rabies management planning and program analysis concurrent with real-time program implementation. Wildlife Services has had an efficient and cooperative relationship with both laboratories since 2003, and they remain critical to the surveillance and monitoring phases of the ORV program in Maine.

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and teeth are analyzed to determine animal age and bait uptake. Fishmeal polymer baits contain a chemical biomarker (tetracycline) that stains teeth/bone and can be detected under microscope; fishmeal coated sachet baits do not contain this biomarker. Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, during the Maine cooperative rabies management program, WS live-trapped 100 unique raccoons; 85 were trapped 4-7 weeks post-2005 ORV bait distribution, 1 was trapped in an area that had not been baited since 2003, and 14 were trapped in an ORV naïve area. Blood and tooth samples were collected from most of these animals and serum samples were sent to the WC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA). One raccoon from the ORV naïve Medway density study showed a presence of rabies VNA (Table 4); a 2 y.o. male captured approximately 38.5 km (24 mi) from the 2004 ORV zone. Similarly, the raccoon from the T2R4 density study (last baited in 2003) also showed a positive rabies antibody response. He was a 3 y.o. male that was captured approximately 15 km (9.5 mi) from the 2004 ORV zone. These raccoons may have taken baits in 2004, retained their rabies VNA levels for nearly a year, and traveled large distances over that time; or they may have had elevated titers because of exposures to the rabies virus.

Four skunks were also trapped and processed during post-ORV activities in 2005. None of the samples had detectable rabies VNA, but 1 tooth showed presence of tetracycline. Two of the skunks were aged and were young of the year animals (<1 y.o.).

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Maine, 2005.

	Medway density study <sup>a</sup>	T2R4 density study <sup>b</sup>	Post-ORV monitoring
Sample collection timeframe	2-12 Aug. 05	18-23 Aug. 05	26 Sep.-14 Oct. 05
Last ORV date (and bait type) <sup>c</sup>	n/a <sup>a</sup>	24 Aug. 03 (CS)	27 Aug. 05 (FMP)
Weeks post-ORV	n/a <sup>a</sup>	103-104	4-7
Unique raccoons	14	1	85
		<b>Serology</b>	
Testable blood samples	14	1	84
Positive rabies antibody response (≥0.05 IU)	1 (7.1%)	1 (100%)	38 (45.2%)
		<b>Tetracycline</b>	
Testable tooth samples	14	1	79
Presence of tetracycline biomarker	0	0	55 (69.6%)

<sup>a</sup> Study was conducted in an ORV naïve area (never before treated with ORV).

<sup>b</sup> Study was conducted prior to annual ORV bait distribution activities, but in an area that had been previously baited.

<sup>c</sup> CS=coated sachet; FMP=fishmeal polymer.

In 2006, WS live-trapped 112 unique raccoons; 92 were trapped 4-7 weeks post-2006 ORV bait distribution, 2 were from an area that was last baited in 2005, and 18 were trapped in an ORV naïve area (Table 5). Blood and tooth samples were collected from most of these animals and serum samples were sent to the WC, while tooth samples were sent to Matson's Lab. The Mattawamkeag study site was only 16 km (10 mi) from the 2005 ORV zone which may explain why 30% of the raccoons captured during that study showed a presence of rabies VNA.

Twenty skunks were also trapped and processed during post-ORV activities in 2006. Two (10.0%) of the samples had detectable rabies VNA, and 8 teeth showed presence of tetracycline. The 2 skunks with a positive antibody response were also trapped and vaccinated in New Brunswick during their TVR efforts. One of the positive antibody skunks was also positive for tetracycline, which may be due to exposure or consumption of an

ORV bait in Maine containing Raboral V-RG®. Eighteen of the skunks were aged: 15 at <1 y.o., 1 at 1 y.o., and 2 at 3 y.o.

Table 5. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Maine, 2006.

	Mattawamkeag density study <sup>a</sup>	Kingman density study <sup>a</sup>	Vanceboro density study <sup>b</sup>	Post-ORV monitoring
Sample collection timeframe	18-28 Jul. 06	18-28 Jul. 06	15-20 Aug. 06	25 Sep.-13 Oct. 06
Last ORV date (and bait type) <sup>c</sup>	n/a <sup>a</sup>	n/a <sup>a</sup>	27 Aug. 05 (FMP)	24 Aug. 06 (FMP)
Weeks post-ORV	n/a <sup>a</sup>	n/a <sup>a</sup>	50-51	4-7
Unique raccoons	10	8	2	92
<b>Serology</b>				
Testable blood samples	10	8	2	92
Positive rabies antibody response (≥0.05 IU)	3 (30.0%)	0	2 (100%)	36 (39.1%)
<b>Tetracycline</b>				
Testable tooth samples	10	8	2	90
Presence of tetracycline biomarker	0	0	1 (50.0%)	44 (48.9%)

<sup>a</sup> Study was conducted in an ORV naïve area (never before treated with ORV).

<sup>b</sup> Study was conducted prior to annual ORV bait distribution activities, but in an area that had been previously baited.

<sup>c</sup> CS=coated sachet; FMP=fishmeal polymer.

*Age Results.*--In 2005 and 2006, 93 and 108 raccoon teeth, respectively, were aged using premolars of live-captured animals (Figure 2). Age results are typical, with the populations dominated by animals ≤1 y.o.

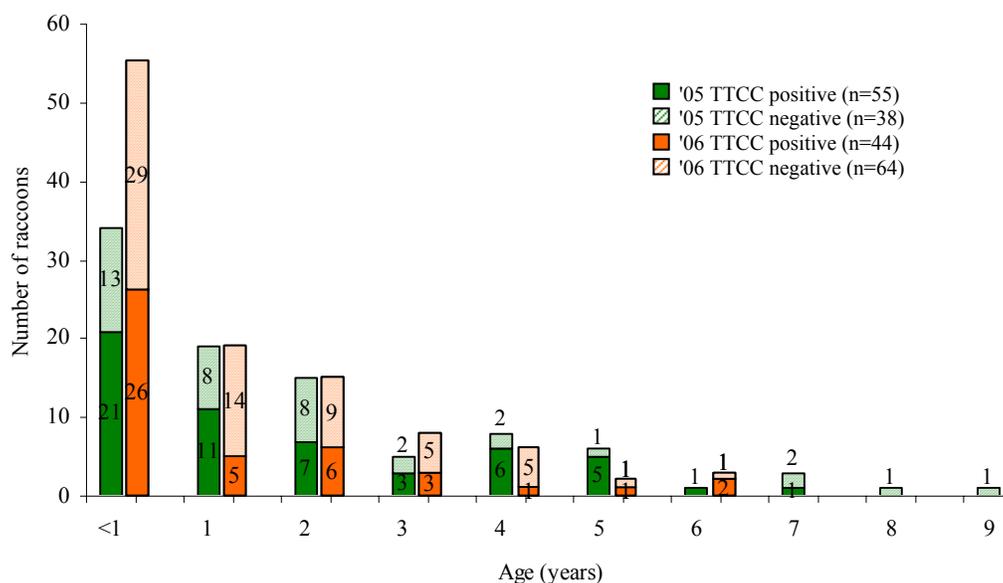


Figure 2. Age class distribution and presence of tetracycline (TTCC) biomarker of 93 and 108 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in Maine, 2005 and 2006.

## SUMMARY

In 2006, WS completed its fifth year of participation in a cooperative rabies management program in northeastern Maine. The placement of Maine’s ORV zone is a northern continuation of neighboring New Brunswick, Canada’s trap-vaccinate-release (TVR) zone. Together, the international effort to stop the northward and eastward spread of rabies covers approximately 100 km (62 mi) of the Maine and New Brunswick border. Since March 2002, after implementing their large scale TVR program, New Brunswick has remained free of raccoon rabies. In 2007, WS will: fully implement the dRIT to enhance rabies surveillance in northern Maine; provide

technical assistance to the public by relating rabies virus information; and coordinate with New Brunswick to plan ORV strategies for preventing the spread of raccoon rabies along, and across, the international border and ultimately eliminate terrestrial rabies in Maine.

# WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM MARYLAND 2006

## BACKGROUND

In 1981, raccoon (*Procyon lotor*) rabies first entered Maryland in Allegany County. It quickly spread and is now present throughout the state. In 1982, raccoon rabies was first documented in Anne Arundel County. In 2000, Anne Arundel County reported 43 cases of rabies, a downward trend from the 97 and 73 cases reported in 1997 and 1998, respectively. From 1996-1998 an average of 18 cases of rabies was reported from the Annapolis Peninsula alone.

In October 1998, the Anne Arundel County Department of Health (AACDH) initiated an oral rabies vaccination (ORV) program on the Annapolis Peninsula. Fishmeal polymer (FMP) baits, containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA), have been distributed each year on the Annapolis Peninsula (94 km<sup>2</sup>) since October 1998, on Gibson Island (4 km<sup>2</sup>) since 2000, and on the Broadneck Peninsula (88 km<sup>2</sup>) since 2001. With the assistance of Wildlife Services (WS) in 2003, ORV efforts expanded to include the entire area (1,080 km<sup>2</sup>) of Anne Arundel County, Maryland (Figure 1). This is a cooperative effort between WS and the AACDH. Wildlife Services provides the major source of funds for project implementation.

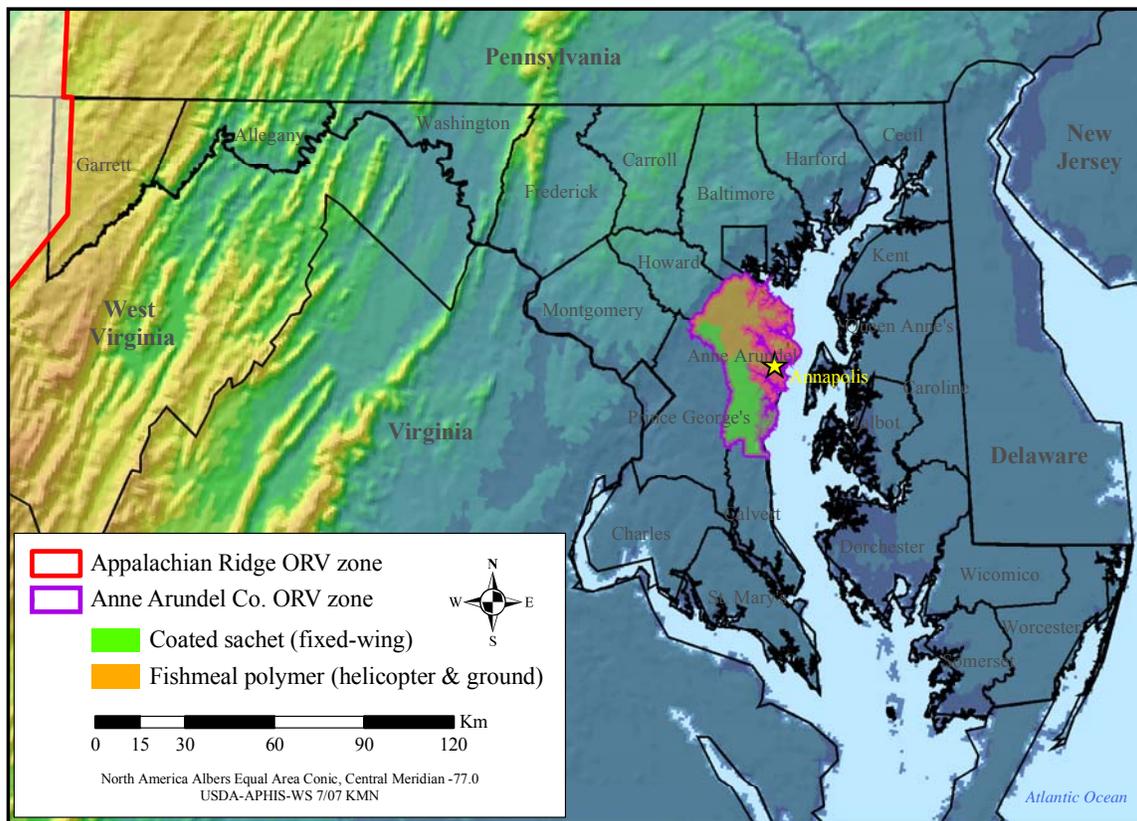


Figure 1. Wildlife Services cooperative rabies management program activities in Maryland, 2006.

In 2003, the Appalachian Ridge (AR) ORV project expanded the eastern boundary from West Virginia into Garrett County, Maryland. To date, WS has distributed 90,933 FMP baits in Garrett County as part of the AR ORV zone that extends from Lake Erie to Tennessee.

## **ORV PROGRAM 2006**

### **Bait Distribution**

For the fourth consecutive year, WS participated in bait distribution efforts in eastern (Anne Arundel County) and western (Garrett County) Maryland; 99,645 baits were distributed over 1,408 km<sup>2</sup> (548 mi<sup>2</sup>) in 2006 (Figure 1).

*Appalachian Ridge.*--The Maryland portion of the AR ORV zone was baited during the greater AR south campaign. From 7-16 August, WS distributed 18,501 FMP baits by fixed-wing aircraft over 328 km<sup>2</sup> (126.6 mi<sup>2</sup>) in Garrett County. The fixed-wing aircraft and pilots were provided by Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA), while WS provided ground support and served as navigators and flight crew in the plane.

*Anne Arundel County.*--In 2006, all of Anne Arundel County (1,080 km<sup>2</sup> [421 mi<sup>2</sup>]) was baited with 81,144 ORV baits: 51,120 FMP and 30,024 fishmeal coated sachets (CS) (Figure 1). The CS baits were distributed by fixed-wing aircraft on 12 September. From 7-29 September 39,335 FMP baits were distributed by helicopter. From 7-8 September and 11-22 September ground teams distributed 11,785 FMP baits by hand. The fixed-wing aircraft and pilots were provided by Dynamic Aviation and the helicopter and pilots were provided by the Anne Arundel County Police Department. Baiting efforts and support were provided by WS and the AACDH.

### **Enhanced Surveillance**

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Maryland WS personnel may schedule dRIT training in the near future at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia.

### **Post-ORV Monitoring**

On 10 October 2006, WS initiated post-ORV trapping to collect blood and tooth samples to evaluate and monitor program success in Anne Arundel County. Over approximately 840 trap nights, WS captured and released 176 unique raccoons. Blood samples were collected from all 176 raccoons, while 158 tooth samples were collected. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

### **Non-target Captures**

Non-target animals captured and released by WS in 2006 included: 63 opossums (*Didelphis virginiana*) and 12 domestic/feral cats (*Felis catus*).

### **Rabies Laboratory Cooperation**

Wildlife Services' ORV program in Maryland cooperates with the Maryland Department of Health and Mental Hygiene's Laboratories Administration (MDH) and the CDC.

*Maryland Department of Health and Mental Hygiene's Laboratories Administration.*--The MDH tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure to the rabies virus). The MDH confirmed 414 cases of rabies in Maryland in 2006. The number of animals tested for rabies statewide was unknown at the time of printing, but 18 and 15 animals were confirmed positive from Anne Arundel and Garrett Counties, respectively. Raccoons represented 65.7% of the animals that tested positive for rabies in Maryland in 2006 (61% and 87% in Garrett and Anne Arundel Counties respectively).

For more information on rabies cases by county and species in Maryland (from 2002-2006) please visit: [http://edcp.org/vet\\_med/rabies.html](http://edcp.org/vet_med/rabies.html)

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA). In 2006, the CDC analyzed 176 raccoon blood serum samples for rabies VNA submitted by Maryland WS. This represented a 17% decrease from the 212 samples submitted by WS in 2005. The Maryland ORV program anticipates 170-200 sample submissions to this laboratory in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and teeth are analyzed to determine animal age and bait uptake. Fishmeal polymer baits contain a chemical biomarker (tetracycline) that stains teeth/bone and can be detected under microscope; fishmeal coated sachet baits do not contain this biomarker. Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

During the evaluation phase of the Maryland cooperative rabies management program, WS live-trapped 210 raccoons, 1 gray fox, and 1 red fox following the 2005 ORV bait distribution and 176 raccoons following 2006 bait distribution in Anne Arundel County. Samples were not collected from the AR ORV zone in Garrett County in either year. Serum samples were sent to the CDC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA). Rabies antibody response in raccoons was down in 2005 (19.5%) from 2004 (23.7%) but up in 2006 (31.8%), while presence of tetracycline was up in 2005 (33.2%) from 2004 (25.9%) but down in 2006 (20.3%) (Table 1). The gray fox from 2005 showed a presence of tetracycline and a positive rabies antibody response of  $\geq 0.56$  IU, while the red fox was negative for both tetracycline and rabies antibody presence.

Table 1. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Anne Arundel County, Maryland, 2005-2006.

	Post-ORV 2005	Post-ORV 2006
Sample collection timeframe	7 Sep.-2 Dec. 05	11 Oct.-17 Nov. 06
Last ORV date (and aerial bait type) <sup>a</sup>	8 Aug.-26 Sep. (FMP/CS)	7-29 Sep. (FMP/CS)
Weeks post-ORV	4-6	5-7
Unique raccoons	210	176
	<b>Serology</b>	
Testable blood samples	210	176
Positive rabies antibody response ( $\geq 0.05$ IU)	41 (19.5%)	56 (31.8%)
	<b>Tetracycline</b>	
Testable tooth samples	190	158
Presence of tetracycline biomarker	63 (33.2%)	32 (20.3%)

<sup>a</sup> CS=coated sachet; FMP=fishmeal polymer.

*Age Results.*--In 2005 and 2006, 188 and 157 raccoon teeth were aged using premolars of live-captured animals from the ORV zone in Anne Arundel County (Figure 2). The gray and red foxes from 2005 were both juveniles <1-year old.

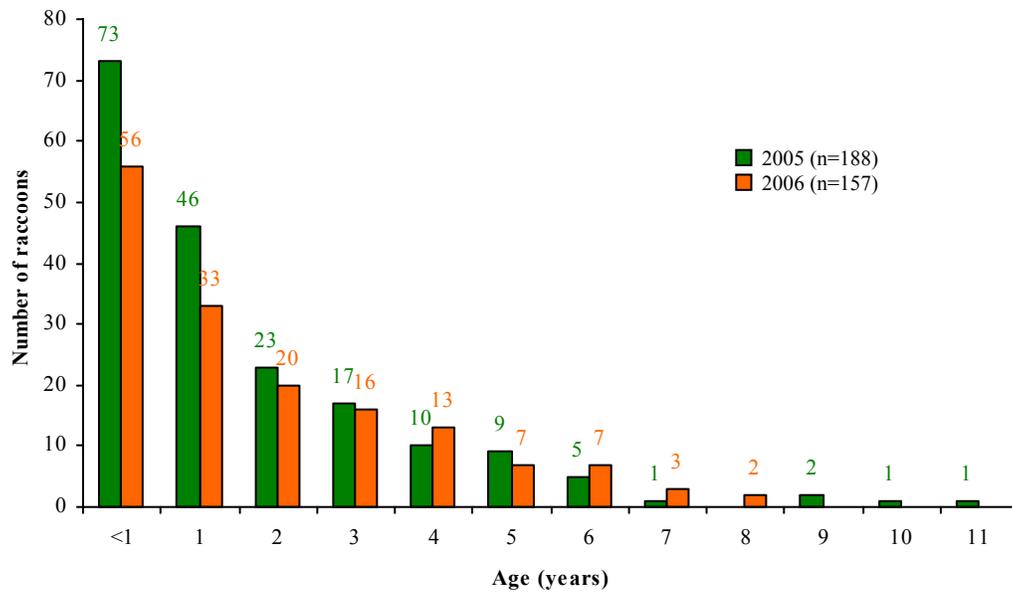


Figure 2. Age class distribution of 345 tooth samples collected by Wildlife Services during the evaluation phases of the cooperative rabies management program in Anne Arundel County, Maryland, 2005-2006.

## SUMMARY

The fall of 2006 marked the fourth year of the eastward expansion of the AR ORV program in Maryland, where WS distributed 18,501 FMP baits in Garrett County. The fall of 2005 also marked the sixth year of WS cooperative participation in the AACDH ORV program. During the 3 years prior to the beginning of the AACDH ORV program (1995-97), an average of 19 rabid animals were reported from the Annapolis Peninsula alone. Since 1998, with the intervention of 361,321 FMP baits, only 18 rabid raccoons have been reported from the Annapolis Peninsula, indicating the success of the Anne Arundel County ORV program.

In 2007, WS plans to continue its cooperative role in ORV bait distribution in Anne Arundel and Garrett Counties, and follow-up post-ORV surveillance trapping to evaluate the success of ORV in Maryland.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM MASSACHUSETTS 2006

### BACKGROUND

Wildlife Services (WS) continues to support the Cape Cod Oral Rabies Vaccination program (CCORV) in southeastern Massachusetts. This cooperative project among Tufts University (TU), the Massachusetts Department of Public Health (MDPH), and the Barnstable County Department of Health and the Environment began in 1994 to reduce the incidence of terrestrial rabies in a 420-712 km<sup>2</sup> area adjacent to the Cape Cod canal and to prevent the spread of rabies to Cape Cod, a heavily populated tourist destination south of Boston. Full time assistance from WS began in 2001 and has typically included bait purchase and distribution, membership on the Massachusetts state and Barnstable County Rabies Advisory Committees, surveillance trapping, and ORV-related wildlife research.

In 2004, the raccoon (*Procyon lotor*) variant of rabies was detected on the ocean-side of the canal and the CCORV operational area now includes all townships on Cape Cod (Barnstable County) (Figure 1). The raccoon variant of rabies was detected in Provincetown in April 2006 and by year's end had been confirmed in 50 animals (48 raccoons and 2 skunks) on the Cape.

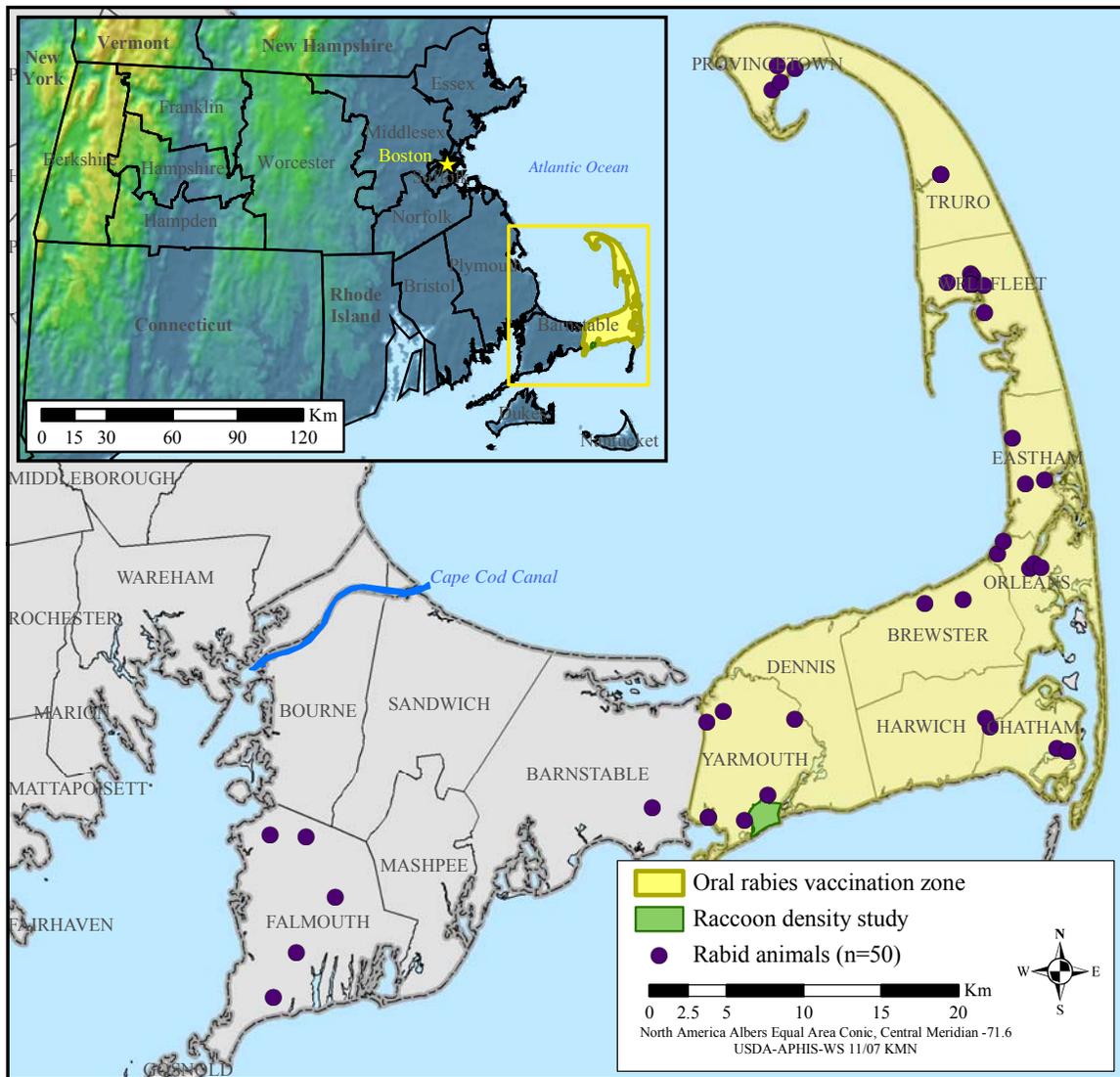


Figure 1. Wildlife Services cooperative rabies management program activities in Massachusetts, 2006.

## ORV PROGRAM 2006

### Bait distribution

For the sixth consecutive year, WS participated in the CCORV program. During 2 campaigns (early summer and late fall), WS and CCORV cooperators distributed 115,427 ORV baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) over approximately 495.9 km<sup>2</sup> (191.5 mi<sup>2</sup>) of Cape Cod (Figure 1). Aircraft and pilots for both CCORV campaigns in 2006 were provided by the U.S. Coast Guard, while WS served as flight crew in the helicopters. Community-based volunteers were an integral part of ground operations in 2006, distributing baits in residential areas by vehicle and on foot. Since its program inception in 1994, CCORV cooperators and WS have distributed 716,317 baits in Massachusetts.

*Early Summer Campaign.*--From 1-9 May 2006, the area of Yarmouth eastward through Provincetown was baited with 55,391 ORV baits (114 baits/km<sup>2</sup>): 5,714 fishmeal-coated sachets (CS) via helicopter; 10,362 CS and 39,315 fishmeal polymer (FMP) via ground operations. This area was baited again in the fall 2006 (see *Late Fall Campaign* below).

*Late Fall Campaign.*--From 24 October-8 November, the *Early Summer Campaign* was repeated. This area was baited with 60,036 ORV baits (122 baits/km<sup>2</sup>): 7,201 CS and 619 FMP via helicopter; 12,517 CS and 38,181 FMP via ground operations. In addition, 25 bait stations containing 1,518 FMP baits were implemented for 2 weeks during the fall campaign in South Yarmouth on a 3 km<sup>2</sup> area that was also used for a density study.

### Enhanced Surveillance

In March 2004, WS and cooperators implemented an enhanced rabies surveillance program that continued through 2006 to track the rabies epizootic on Cape Cod for planning purposes, epidemiological data collection, and reduction of municipal infrastructure-based variation in specimen submissions. Primary activities of the CCORV enhanced surveillance program included the collection, preparation, and transportation of samples to the MDPH Laboratory Institute (LI) for rabies testing. These specimens were collected via public (residential) reports of sick or strange behaving animals to municipal officials, nuisance wildlife trapping, and road kill surveys. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

Wildlife Services (with cooperator assistance) submitted 93 specimens (59 raccoons, 11 skunks [*Mephitis mephitis*], 9 coyotes [*Canis latrans*], 9 red foxes [*Vulpes vulpes*], 4 Virginia opossums [*Didelphis virginiana*], 2 woodchucks [*Marmota monax*], 1 gray fox [*Urocyon cinereoargenteus*], and 1 muskrat [*Ondatra zibethicus*]) to the MDPH LI as part of an enhanced rabies surveillance program on Cape Cod. Six of these specimens were obtained off-Cape in the historic ORV zone (3 raccoons, 2 skunks, and 1 gray fox). Seven specimens (5 raccoons, 1 coyote, and 1 muskrat) were not tested because the samples were unsuitable. Of the 64 raccoons and skunks tested, 26 (40.6%) tested positive for rabies (Table 1); no other positives were reported among WS surveillance specimens. Of all specimens tested, 25 raccoons and 1 skunk tested positive for rabies.

Table 1. Prevalence of rabies in Wildlife Services raccoon and skunk surveillance specimens tested by the Massachusetts Department of Public Health, Laboratory Institute from various sources on Cape Cod, Massachusetts, 2006.<sup>a</sup>

Source	Raccoons submitted	Raccoons suitable for testing	Rabies positive raccoons <sup>b</sup>	Skunks submitted	Skunks suitable for testing	Rabies positive skunks <sup>b</sup>
Complaint-based: residential property <sup>c</sup>	26	23	19 (82.6%)	4	4	1 (25.0%)
Rehab center/veterinary clinic	16	16	1 (6.3%)	3	3	0
Road killed	5	5	1 (20.0%)	0	0	0
Trapped <sup>d</sup>	2	2	0	1	1	0
Complaint-based: commercial property	1	1	0	0	0	0
Other <sup>e</sup>	2	1	1 (100%)	0	0	0
Unknown	6	5	3 (60.0%)	3	3	0
Total	58	53	25 (47.2%)	11	11	1 (9.1%)

<sup>a</sup> Does not include specimens submitted directly by municipal governments.

<sup>b</sup> Percentage = (# rabies positive ÷ # animals suitable for testing) x 100.

<sup>c</sup> Result for 1 raccoon obtained from a residential property is unavailable; specimen omitted from Table 1.

<sup>d</sup> Live trapping to monitor for rabies bait-uptake. At no time was trapping undertaken with the aim of collecting surveillance specimens.

<sup>e</sup> Includes forested and recreation areas.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

An adequate space was not available for implementation of the dRIT during 2006. Efforts are currently underway to locate a new office/warehouse space which will accommodate the dRIT process. During 2006, WS collected 4 raccoons that will be tested by WS using the dRIT once adequate laboratory space is obtained. Once full-scale dRIT use is underway, all positives, 10% of all negatives, and all indeterminate samples will be sent to the Centers for Disease Control and Prevention (CDC) for confirmation and strain typing.

### Population Monitoring

In 2006, WS conducted a raccoon density study in South Yarmouth (Figure 1 inset) in collaboration with the Yarmouth Department of Natural Resources (YDNR). The National Rabies Management Program standard protocol of 50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights was used. From 15-25 July, WS captured 20 unique raccoons for an index to density of 6.5 raccoons/km<sup>2</sup>. In addition, WS trapped 8 recaptured raccoons, 15 unique skunks, and 19 non-targets. Target animals were chemically immobilized, and hand vaccinated against rabies with Imrab® 3 (Merial Limited). Blood sera and a first premolar tooth were collected from each target animal for ORV program evaluation. Other morphological characteristics were also recorded. One raccoon and 1 skunk died in captivity. One strange-acting raccoon was euthanized by the YDNR. All 3 specimens tested negative for rabies.

### Post-ORV Monitoring

In 2006, rabies surveillance efforts again took precedence over post-ORV monitoring as WS tracked the rabies epizootic through the outer portions of Cape Cod to Provincetown. Therefore, post-ORV trapping was conducted secondarily to the surveillance effort. During 2006, in 175 post-ORV monitoring trap nights, 15 raccoons and 3 unique skunks were captured and released on Cape Cod. Seven raccoons were recaptured after our density study in South Yarmouth, which is described below. Captured target animals were chemically immobilized, and hand vaccinated (with Imrab® 3) against rabies. Blood sera and a first premolar tooth were collected from each target animal for ORV program analysis. Other morphological characteristics were also recorded. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

## Non-target Captures

Non-target animals captured and released by WS in 2006 included: 32 opossums, 3 domestic/feral cats (*Felis catus*), 2 Eastern box turtles (*Terrapene carolina*), and 1 bird (Class *Aves*).

## Rabies Laboratory Cooperation

Wildlife Services in Massachusetts cooperates with the MDPH LI and the CDC in support of the ORV program on Cape Cod. The LI processes specimens from all counties in Massachusetts in addition to cooperating with the CCORV program.

*Massachusetts Department of Public Health Laboratory Institute.*--The LI tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by Animal Control and Public Health Officials) and enhanced surveillance (specimens not involved in an exposure and usually submitted by WS from Barnstable and Plymouth Counties or municipal officials statewide). In 2006, the LI received 2,910 specimens for rabies testing, representing a 14.1% decrease from the number of samples received statewide in 2005. Animals were submitted from all 14 counties throughout the state and 232 tested positive for rabies in 2006 (Table 2). Of the animals tested statewide, 261 (9.0%) came from within the current ORV zone (Barnstable County), representing a 57.1% decrease from the number of samples submitted from Barnstable County in 2005. Although the number of raccoons submitted from Barnstable County in 2006 (300) was down considerably when compared to 2005 (552), the percentage of raccoons testing positive in 2006 (45.0%) was up from 2005 (37.5%).

Raccoons, skunks, foxes, and coyotes are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 82.2% of the animals submitted for rabies testing in Massachusetts in 2006 are reported by WS as "other." For a more detailed listing of rabid animals from Massachusetts in 2006 (and prior years dating back to 1992) please visit: <http://www.mass.gov/dph/cdc/epii/rabies/rabies.htm>.

Table 2. Animals submitted for rabies testing to the Massachusetts Department of Public Health Laboratory Institute in Massachusetts, 2006.

	Submissions	Rabies Positive
Raccoons	300	135 (45.0%)
Skunks	154	40 (26.0%)
Foxes	52	9 (17.3%)
Coyotes	12	1 (8.3%)
Other <sup>a</sup>	2,392	47 (2.0 %)
Total	2,910	232 (8.0%)

<sup>a</sup> Other animals included: bats, cats, dogs, woodchucks, and a river otter.

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA) in support of the CCORV program. Massachusetts WS submitted 44 blood serum samples for rabies VNA analysis to the CDC in 2006. Wildlife Services anticipates an increase in the number of serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>.

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, some of the current year's (2006) evaluation data were available so they have been included here as well.

## **Serology, Tetracycline Biomarker, and Age Results**

Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and teeth are analyzed to determine animal age and bait uptake. Fishmeal polymer baits contain a chemical biomarker (tetracycline) that stains teeth/bone and can be detected under microscope; fishmeal coated sachet baits do not contain this biomarker. Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, during the CCORV program, WS collected serum samples from 34 raccoons and 1 skunk from March through September. The serum samples were sent to the CDC and 5 raccoons (14.7%) and the lone skunk demonstrated a positive rabies antibody response ( $\geq 0.05$  IU). Tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA), but biomarker and age results were unavailable at the time of printing.

In 2006, during the evaluation phase of the Massachusetts cooperative rabies management program, WS collected serum samples from 29 raccoons and 15 skunks from July through September. The serum samples were sent to the CDC and 11 raccoons (37.9%), but none of the skunks, demonstrated a positive rabies antibody response. Tooth samples were sent to Matson's Lab, but biomarker and age results were unavailable at the time of printing.

## **SUMMARY**

In 2006, WS completed its sixth year of cooperative participation in rabies management on Cape Cod, Massachusetts. Wildlife Services continued to work with cooperators to develop optimal rabies control strategies. In addition, epizootic front delineation, enhanced rabies surveillance behind the front to track epizootic intensity, and public information campaigns were priorities of the CCORV program in 2006. A 3 km<sup>2</sup> study area located in South Yarmouth was also used for a raccoon density study and for implementing the experimental use of bait stations in an attempt to optimize ORV bait delivery.

In 2007, the CCORV goal is to implement and evaluate strategies for restoring Cape Cod to raccoon rabies-free status and creating a new, appropriate ORV zone on the west side of the Cape Cod Canal to prevent rabies from spreading back onto the Cape.

## **WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM MICHIGAN 2006**

### **BACKGROUND**

To date, there have been no reported cases of the raccoon (*Procyon lotor*) variant of rabies in Michigan, although other variants (including bat [*Chiroptera* spp.] and striped skunk [*Mephitis mephitis*]) do occur. Given its close proximity to both Ohio and Ontario, Canada however, where raccoon rabies is known to exist, the state of Michigan established a multi-agency rabies working group in 1997. The goals of the group are to: educate the public; provide expertise and recommendations on rabies issues; conduct research; prevent an increase in the disease and risk for existing variants; and prevent the establishment of new variants (raccoon rabies in particular). The Michigan rabies working group developed a contingency plan for the introduction of raccoon rabies, should it occur. The plan was adopted by Wildlife Services' (WS) National Rabies Management Program (NRMP) and has been implemented in areas where raccoon rabies approached or breeched an oral rabies vaccination (ORV) zone, or was confirmed where it was not formerly known to exist. The Michigan rabies working group has also developed an enhanced surveillance system (ESS) to aid in the early detection of raccoon rabies. Through guidance and training from the NRMP, biologists from WS and the Michigan Departments of Natural Resources (MDNR) and Community Health (MDCH) began implementing the ESS in 2005. Therefore, 2006 marked the second year that raccoon rabies enhanced surveillance activities took place in Michigan.

### **RABIES MANAGEMENT PROGRAM 2006**

#### **Bait Distribution**

There is currently no ORV bait distribution program in Michigan.

#### **Enhanced Surveillance**

In 2006, a total of 94 enhanced surveillance specimens were collected in 19 counties (Figure 1) and processed by WS. Animals included: 76 raccoons, 12 gray foxes (*Urocyon cinereoargenteus*), 4 striped skunks, and 2 red foxes (*Vulpes vulpes*) (Table 1). All animals were displaying signs of a neurological disorder and were collected by cooperators, such as nuisance wildlife control operators, local animal control officers, and MDNR staff. These "rabies suspect" animals are very desirable for enhanced surveillance and were submitted in an effort to increase rabies awareness in those that might receive calls for assistance from the public. Brainstem and tooth samples were collected from each animal and hair samples were collected from raccoons for DNA analysis. All animals tested negative for the rabies virus. In 2006, no animals were trapped by WS specifically for enhanced rabies surveillance.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the dFA test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

During 2006, WS tested the 94 animals mentioned above using the dRIT. Ten percent of the negative samples were sent to the Centers for Disease Control and Prevention (CDC) for confirmation. The CDC (using the dFA test) had 100% agreement with the WS dRIT test results for negative samples. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Michigan. The MDNR cooperated in the implementation of the dRIT by providing the use of their Wildlife Disease Biosafety Level 2 (BSL-2) Laboratory.

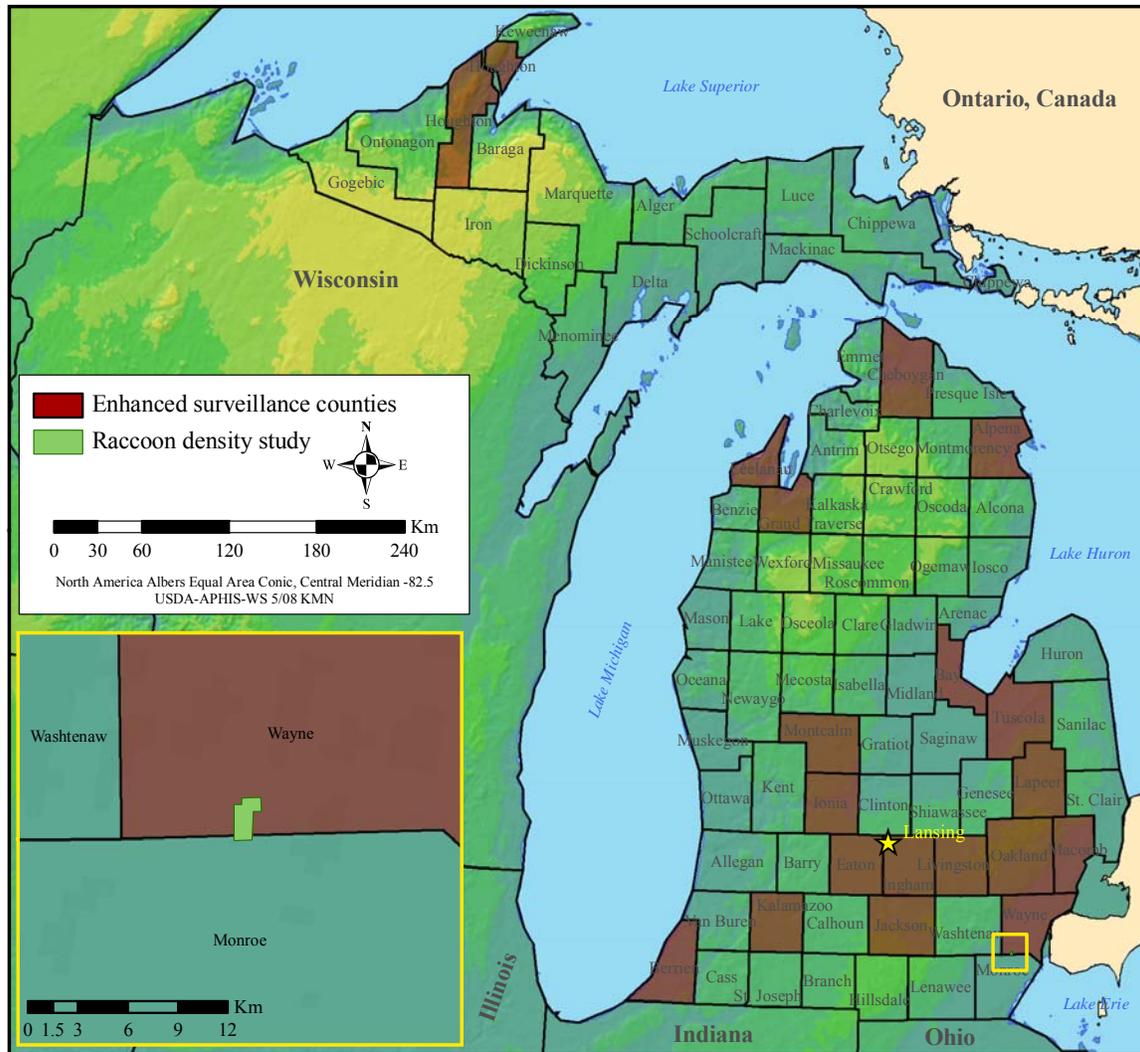


Figure 1. Wildlife Services cooperative rabies management program activities in Michigan, 2006.

## Population Monitoring

In October 2006, a raccoon relative density study was conducted at a natural area in Wayne County, Michigan with cooperation from the MDNR and MDCH (Figure 1 inset). The purpose of the study was to better understand the raccoon population of southeast Michigan, which is considered to be at the highest risk for a raccoon rabies introduction. During the previous year, a density study was conducted at a Metropark in Oakland County, Michigan, yielding a relatively high index to raccoon density of 51.7 raccoons/km<sup>2</sup>. The 2006 study site was selected to represent a more rural landscape, which is also typical habitat in southeast Michigan. Trapping and sample collection procedures were conducted following the NRMP standard protocol for raccoon density studies (50 cage traps set for 10 consecutive nights over a target study area of 3 km<sup>2</sup>). A total of 55 unique raccoons were captured and processed on a 3.14 km<sup>2</sup> study area over 500 trap nights, yielding an index to raccoon density of 17.5 raccoons/km<sup>2</sup>. In addition, 14 recaptured raccoons were trapped. All but 1 raccoon was processed and released; this animal was euthanized because it exhibited abnormal behavior. It tested negative for rabies using the dRIT. One striped skunk was also captured, processed and released. Processing procedures included chemical immobilization, ear tagging, and biological sample collection (blood, tooth, and hair/DNA).

All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations. This project also provided an opportunity for WS to work along with biologists from both the MDNR and MDCH in rabies surveillance trapping

and sampling, which adds to Michigan’s overall preparedness for a potential response to the introduction of raccoon rabies.

### Other Rabies Management Program Activities

The Michigan Rabies Working Group held a 1-day Michigan Rabies Conference in April 2006. The conference was attended by public health professionals (medical directors, nurses, epidemiologists, and environmental health staff), health care providers, veterinarians, animal control officers, nuisance wildlife control operators, and wildlife biologists. The agenda included presentations by national and state-level rabies experts, which provided information on rabies surveillance, contingency planning, control, and prevention. The primary focus was on the emergence of raccoon rabies and the roles of the target audience in surveillance and potential response.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 20 opossums (*Didelphis virginiana*), 2 Eastern cottontails (*Sylvilagus floridanus*), and 1 domestic/feral cat (*Felis catus*).

### Rabies Laboratory Cooperation

Wildlife Services’ rabies management program in Michigan cooperates with the MDCH Bureau of Laboratories (BOL) and the CDC.

*Michigan Department of Community Health Bureau of Laboratories.*--The BOL tests cross-sections of animal cerebellums and brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure) and cross-sections of the brainstems for enhanced surveillance (specimens not involved in an exposure and usually submitted by WS or the MDNR). Specimens are tested at the BOL using the dFA test. The BOL received 2,874 samples for rabies virus testing in 2006, representing a 13.2% increase from the number of samples submitted in 2005. The BOL did not test 107 of these samples because they were either unsatisfactory or were from non-rabies vector species (i.e., small rodents and lagomorphs). An additional 18 samples yielded inconclusive rabies results. Of the 2,749 samples that yielded results, 49 (1.8%) tested positive for the rabies virus (Table 1).

Raccoons, skunks, and foxes are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 93.7% of the animals tested for rabies in Michigan in 2006 (that yielded results) are reported by WS as “other.” For more information on current and historical rabies cases in Michigan please visit the Michigan Emerging Disease Issues website at: <http://www.michigan.gov/emergingdiseases/> and click on the “rabies” link on the left side.

Table 1. Animals submitted and tested for rabies by the Michigan Department of Community Health Bureau of Laboratories (BOL) and the Michigan Wildlife Services (WS) Program in 2006.

Species	Public Health Surveillance (BOL)		Enhanced Surveillance (WS)	
	Submissions	Rabies Positive	Submissions	Rabies Positive
Raccoons	130	0	76	0
Skunks	24	3 (12.5%)	4	0
Foxes	18	1 (5.6%)	14	0
Other <sup>a</sup>	2577	45 (1.7%)	0	0
Total	2749	49 (1.8%)	94	0

<sup>a</sup> Other animals included: bats, domestic cats, and sheep.

*Centers for Disease Control and Prevention.*--The CDC tests animal brainstems for rabies as part of enhanced surveillance dRIT confirmation. The CDC also analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA). In 2006, Michigan WS submitted 50 blood serum samples for rabies VNA analysis to the CDC. As there is no ORV program and consequently no ORV surveillance trapping, these 50 samples came from the raccoon relative density study conducted in October 2006. For more information

about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## **RABIES MANAGEMENT PROGRAM 2005 – EVALUATION**

### **Serology, Tetracycline Biomarker, and Age Results**

Animal blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well. Also, tetracycline can be picked up from other items in the environment other than FMP baits.

There is no ORV baiting program in Michigan, therefore the specimens collected for enhanced surveillance are considered “ORV naïve” (not previously treated with ORV). These samples represent a negative control for the NRMP and can be used to estimate “baseline” tetracycline and rabies VNA response levels. In 2005, 197 tooth samples were submitted for tetracycline screening of which 2 (1.0%) had tetracycline present. Serum samples were submitted for 173 animals of which 4 (2.3%) demonstrated a positive rabies VNA response ( $\geq 0.05$  IU).

### **SUMMARY**

During 2006, WS completed its second year of participation in cooperative rabies management efforts in Michigan. Wildlife Services began an educational campaign of local health department staff, human and animal healthcare providers, animal and wildlife control officers, local law enforcement officers, and other personnel from state and federal agencies. The education of these critical stakeholders will increase the efficiency of sample collection throughout the state and thereby enhance surveillance for raccoon rabies in Michigan. Although the number of samples collected was relatively low in 2006, the program gained valuable cooperation and started receiving non-exposure “rabies suspect” animals from a variety of sources. In 2007, these outreach efforts will continue and hopefully will expand the enhanced surveillance system.

# WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM MISSISSIPPI 2006

## BACKGROUND

The raccoon (*Procyon lotor*) variant of the rabies virus has not been documented in Mississippi. In 1964, rabies was first detected in bats (*Chiroptera* spp.) within Mississippi. The last indigenous case of rabies in a dog (*Canis familiaris*) occurred in 1961. In 1965, two puppies imported from another state were confirmed rabid before they came into contact with other animals. Since World War II, 14 cases of human rabies have occurred in the state, the last being in 2005. Most of these cases are suspected to have been canine rabies (Dr. Brigid Elchos, Mississippi Department of Health [MDH], pers. comm.). These cases were located throughout the state. The most recent case was in September 2005; a child who died and had elevated rabies titer levels. A bat was reportedly in his bedroom in April 2005. Unfortunately, the child was cremated so no post-mortem exam to reveal more information about the cause of death could be performed.

Extensive rabies vaccination programs for dogs have been conducted over the last several decades. As the number of dogs vaccinated against rabies increased, the number of positive animals confirmed by the MDH Laboratory decreased. Currently, only the bat variant of rabies is considered enzootic within Mississippi (Riecken 1984). The nearest confirmed case of raccoon rabies to Mississippi has been in Clarke County, Alabama (Figure 1). As a result of the proximity of Clarke County cases, Wildlife Services (WS) in Mississippi began an enhanced rabies surveillance program in 2003 to detect raccoon rabies, should it enter the state.

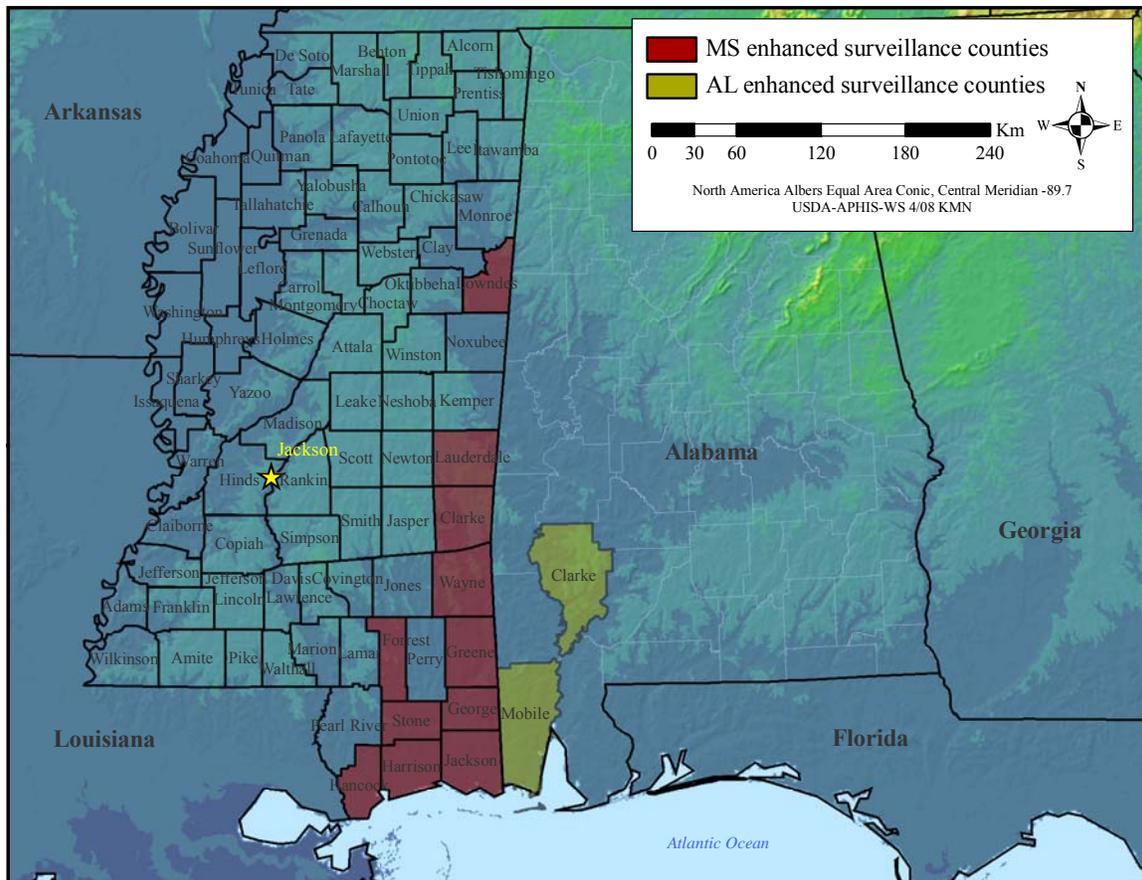


Figure 1. Wildlife Services cooperative rabies management program activities in Mississippi, 2006.

## RABIES MANAGEMENT PROGRAM 2006

### Bait Distribution

There is currently no oral rabies vaccine (ORV) bait distribution program in Mississippi.

### Enhanced Surveillance

In 2006, WS continued enhanced surveillance of rabies by collecting road killed, suspect rabid, or nuisance raccoons, striped skunks (*Mephitis mephitis*), red foxes (*Vulpes vulpes*), gray foxes (*Urocyon cinereoargenteus*), coyotes (*Canis latrans*), and bobcats (*Lynx rufus*) in Clarke, Forrest, George, Greene, Hancock, Harrison, Jackson, Lauderdale, Lowndes, Stone, and Wayne Counties of Mississippi (Figure 1). A Mississippi WS technician also conducted enhanced surveillance activities in Clarke and Mobile Counties of Alabama in 2006.

Wildlife Services cooperated with wildlife law enforcement officers, city animal control officers, and various county officials to collect 60 animal brainstem samples from Mississippi and 46 samples from Alabama in 2006. All 106 samples from Mississippi and Alabama were tested by a WS technician using the direct rapid immunohistochemistry test (dRIT) in Gulfport, MS. All samples tested negative for the rabies virus.

Table 1. Enhanced surveillance animals collected for rabies testing by Mississippi Wildlife Services in Mississippi and Alabama counties (no rabies positives confirmed), 2006.

County	Raccoon	Striped skunk	Red fox	Gray fox	Coyote	Bobcat	Total
<b>Mississippi</b>							
Clarke	2			1			3
Forrest				1			1
George	4	2					6
Greene	2	1					3
Hancock	2		1				3
Harrison	7		3				10
Jackson	16		1				17
Lauderdale	7						7
Lowndes	1				1		2
Stone	1						1
Wayne	4	1			1	1	7
Total	46	4	5	2	2	1	60
<b>Alabama</b>							
Clarke			1	1			2
Mobile	44						44
Total	44		1	1			46

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

One Mississippi WS personnel attended dRIT training in April 2005 at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia. The test was implemented in 2006.

### Rabies Laboratory Cooperation

The WS cooperative rabies management program in Mississippi currently collaborates with the MDH, Public Health Laboratory (PHL). The PHL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by veterinarians) and when possible,

via enhanced surveillance (specimens not involved in an exposure, usually submitted by WS). On average, the PHL tests over 400 animals for rabies each year (primarily domestic cats [*Felis catus*] and dogs). Numerous bats are also submitted each year. The PHL confirmed 9 cases of rabies in Mississippi in 2006; they were all bats.

For the past 3 years, WS has assisted the PHL by purchasing various rabies testing supplies (e.g., pipettes, gloves, shipping boxes, etc.). Wildlife Services implemented the dRIT in 2006 to relieve some of the rabies testing burden on the PHL, but still anticipates similar numbers of submissions to the lab. For more information about rabies in Mississippi please visit: <http://www.msdh.state.ms.us> and search on “rabies.”

## **SUMMARY**

During 2006, WS completed its fourth year of participation in cooperative rabies management efforts in Mississippi. The focus of activities for 2006 continued to be enhanced rabies surveillance in the southeastern part of the state and across the border in southwestern Alabama. In 2007, efforts will continue to focus on enhancing surveillance in these areas to detect the raccoon variant of the rabies virus, should it spread from nearby Alabama.

## **LITERATURE CITED**

Riecken, W.E. Jr. 1984. Review of Rabies in Mississippi. Mississippi Morbidity Report (June). Mississippi Department of Health. Vol 2,11:1-3.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM NEW HAMPSHIRE 2006

### BACKGROUND

The first case of raccoon (*Procyon lotor*) rabies confirmed in New Hampshire was a bit of an anomaly. In March 1992, a raccoon entered a local police officer's house in Rumney and began fighting with the family Doberman (Figure 1). The officer was forced to shoot the raccoon under the kitchen table. The raccoon was brought to a local veterinarian's office and was found to be wearing 2 flea collars, likely to be a "pet" of unknown origin (retired State Veterinarian Dr. Clifford McGinnis, New Hampshire Department of Agriculture, Markets and Food [NHDA], personal communication in 2002). The raccoon was confirmed rabid on 6 April with the raccoon variant of rabies. Rumney is approximately 128 km (80 mi) north of the New Hampshire-Massachusetts state border and officials believed the family caring for the raccoon translocated it from a rabies-infected area in southern New England. Fortunately, no additional cases were detected in the Rumney area, but the raccoon variant of the rabies virus did enter New Hampshire in the fall of 1992, as an extension of the epizootic in southern New England. Once in New Hampshire, raccoon rabies continued its northward spread at a rate of about 40 km (25 mi) a year. Raccoon rabies has been confirmed in all 10 counties, with the northernmost case occurring in Lancaster (in 1999) approximately 60 km (37.5 mi) south of the United States-Canada border (Figure 1).

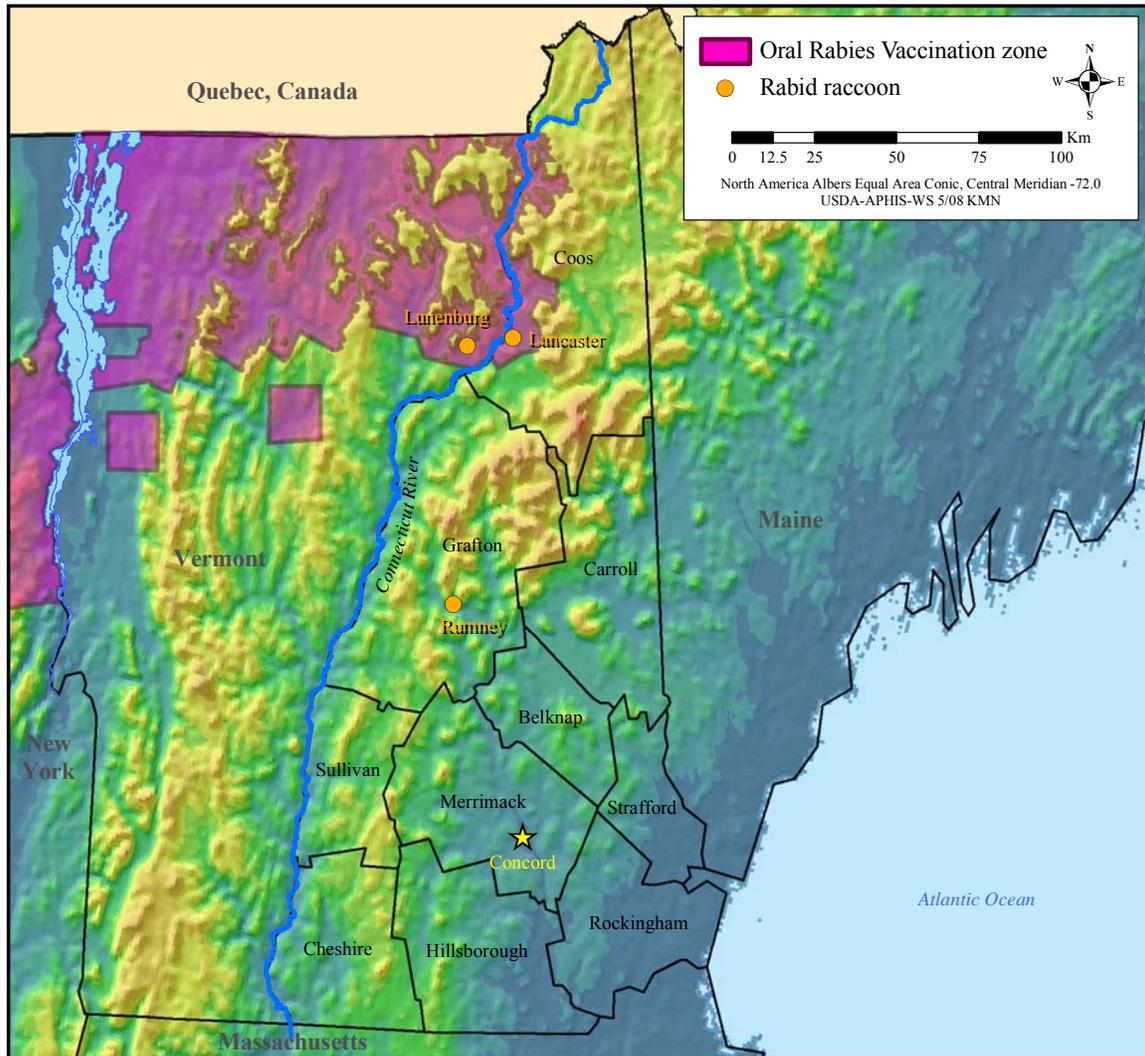


Figure 1. Wildlife Services cooperative rabies management program activities in New Hampshire, 2005.

Just months after the Lancaster, New Hampshire case was documented, a rabid raccoon was confirmed in Lunenburg, Vermont (19 October 1999) directly across the Connecticut River (state border) from Lancaster (Figure 1). This prompted concern over the spread of rabies through the Connecticut River Valley in northern New Hampshire and Vermont into Canada. In July 2001, Wildlife Services (WS) conducted a raccoon density study in northern New Hampshire to better understand the population there and the potential movement of rabies. In August 2001, an oral rabies vaccination (ORV) program was initiated in New Hampshire with the goal to prevent the northward spread of raccoon rabies.

The New Hampshire ORV program has been a cooperative effort between WS, Cornell University (CU), the NHDAG, the New Hampshire Department of Health and Human Services (NHDHHS), and the New Hampshire Department of Fish and Game. Wildlife Services has been the major source of federal funds for project implementation. Wildlife Services has also provided federal wildlife management leadership by continuing to play an active role in: program planning and coordination; organizing ground support for aerial bait distribution; working in and navigating aircraft to distribute baits; coordinating the hand distribution of baits in areas too populated to bait by air; and providing surveillance and follow-up field work by collecting blood and tooth samples from live-trapped and suspect-rabid animals within the New Hampshire ORV bait zone to test program efficacy.

## **ORV PROGRAM 2006**

### **Bait Distribution**

For the sixth consecutive year in 2006, WS participated in ORV bait distribution efforts along the upper Connecticut River Valley in northern New Hampshire, coinciding with Vermont aerial bait distribution activities; 29,473 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 529.7 km<sup>2</sup> (204.5 mi<sup>2</sup>) of Coos County (Figure 1). During 2 flights on 22 August, 28,924 CS baits were distributed by Twin Otter fixed-wing aircraft over northern New Hampshire, while the villages of Colebrook and Lancaster were hand baited with 549 total FMP baits on 23 August. Pilots and mechanics for aerial baiting were provided by the Ontario Ministry of Natural Resources, while WS provided navigators, flight crew, and ground support. Since its program inception in 2001, WS has distributed 148,092 ORV baits in New Hampshire.

### **Enhanced Surveillance**

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Currently, New Hampshire WS is not planning training or implementation of the dRIT because the NHDHHS is meeting enhanced surveillance testing needs.

### **Post-ORV Monitoring**

From 17-20 October 2006, Vermont WS conducted post-ORV trapping activities in Coos County, New Hampshire. Over 88 trap nights, 11 raccoons were trapped and blood serum and tooth samples were collected from most of them to measure rabies virus neutralizing antibody (VNA) response in the ORV zone. All raccoons were immobilized, processed and released. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

### **Non-target Captures**

No non-target species were captured in New Hampshire in 2006.

## Rabies Laboratory Cooperation

Wildlife Services' ORV program in New Hampshire cooperates with the NHDHHS, Public Health Laboratory (PHL) and the New York State Department of Health's Rabies Laboratory at the Wadsworth Center (WC).

*New Hampshire Department of Health and Human Services, Public Health Laboratory.*--The PHL routinely tests animal brainstems for rabies via public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by Conservation Officers, veterinarians, and the public). When possible, the PHL tests specimens not involved in an exposure to enhance surveillance in towns which have not yet had a confirmed case of rabies. The PHL tested 580 brainstem samples for the rabies virus in 2006 (Table 1). This represents an 18.1% increase from the number of samples tested in 2005. The 2006 samples were submitted from all 10 counties throughout the state and 48 tested positive for rabies, up significantly from the 11 positives in 2005.

Raccoons, skunks (*Mephitidae* spp.), and foxes (*Canidae* spp.) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 81.7% of the animals tested for rabies in New Hampshire in 2006 are reported by WS as "other." For more information about rabies in New Hampshire please visit:

<http://www.dhhs.state.nh.us/DHHS/CDCS/rabies.htm>

Table 1. Animals tested for rabies by the New Hampshire Department of Health and Human Services, Public Health Laboratory in New Hampshire, 2006 (rabies positives statewide in parentheses).

	Statewide	Within and adjacent <sup>a</sup> to New Hampshire ORV zone
Raccoons	38 (19)	0
Skunks	49 (17)	0
Foxes	19 (6)	0
Other <sup>b</sup>	474 (6)	7 (1.5%)
Total	580 (48)	7 (1.2%)

<sup>a</sup> Samples were from areas inside or  $\leq 6$  km (10 mi) outside the ORV zone.

<sup>b</sup> Other animals included: bats (4 rabid), domestic pets (2 rabid cats), and woodchucks.

*New York State Department of Health's Rabies Laboratory at the Wadsworth Center.*--The WC analyzed 11 serum samples from New Hampshire for rabies VNA submitted by Vermont WS in 2006. This was slightly more than the number of samples submitted by WS in 2005 (n=7). The Vermont/New Hampshire ORV program anticipates similar numbers of submissions to the WC in 2007. For more information about the Rabies Laboratory at the WC please visit: <http://www.wadsworth.org/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake. Fishmeal polymer baits contain a chemical biomarker (tetracycline) that stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker. Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005 and 2006, during the evaluation phases of the New Hampshire cooperative rabies management program, WS live-trapped 7 and 11 unique raccoons, respectively, within the ORV bait zone. Blood and tooth

samples were collected from most of these animals and serum samples were sent to the WC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA).

Two of 7 (28.6%) serum samples collected 9 weeks post-2005 ORV demonstrated a positive rabies VNA response (serum titer  $\geq 0.05$  IU/ml). No tooth samples showed a presence of tetracycline biomarker, but CS baits were distributed in 2005. The age structure of these 7 raccoons was: 4 at age <1 y.o.; 2 at age 1 y.o.; and 1 at age 8 y.o.

In 2006, 5 of 11 (45.5%) serum samples collected 8 weeks post-ORV demonstrated a positive rabies VNA response. No tooth samples showed a presence of tetracycline biomarker, but CS baits were distributed in 2006. Ten teeth were aged: 8 at age <1 y.o.; 1 at age 3 y.o.; and 1 at age 5 y.o.

## **SUMMARY**

During 2006, WS completed its sixth year of participation in cooperative rabies management efforts in New Hampshire. The focus of activities this year was ORV bait distribution and follow-up post-ORV monitoring (trapping). The New Hampshire ORV program is an important part of a larger northeastern cooperative effort, which in 2006 included: New York; Vermont; Maine; and Ontario, Quebec, and New Brunswick, Canada. The Northeastern cooperative effort is tied to national planning efforts to contain and explore strategies to eliminate the raccoon variant of the rabies virus.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM NEW JERSEY 2006

### BACKGROUND

In the early part of the twentieth century New Jersey had a significant problem with rabies in dogs (*Canis familiaris*); in 1939, 675 dogs and 4 humans died of rabies in the state. In 1942, a rabies management program involving mass vaccination of dogs and collection of stray animals was initiated. As a result of these efforts New Jersey experienced its last case of canine rabies in 1956. In 1960, the first case of bat (*Chiroptera* spp.) rabies was confirmed in New Jersey. The raccoon (*Procyon lotor*) rabies epizootic spread to New Jersey through Warren and Hunterdon Counties in 1989. In 1991, New Jersey led the nation in animal rabies cases per capita. The scale of the outbreak stimulated public support for a safety and efficacy trial of an oral rabies vaccine (ORV) in wild raccoons by the State of New Jersey and Thomas Jefferson University, Philadelphia, Pennsylvania. Between 1989 and 2005, 4,938 cases of terrestrial rabies were confirmed. Of these, 3,681 (74.5%) were raccoons.

From 1992-1994, the New Jersey Division of Fish and Wildlife worked cooperatively with the Cape May County Department of Health (CMDH) and the New Jersey Department of Health and Senior Services (NJDHSS) to conduct an experimental ORV program on the rabies-free peninsula of Cape May County (Figure 1). Since 1995, the CMDH has worked independently to distribute ORV baits in Cape May County. In 2004, Wildlife Services (WS) became involved in the ORV program to assist county and state cooperators with project evaluation.

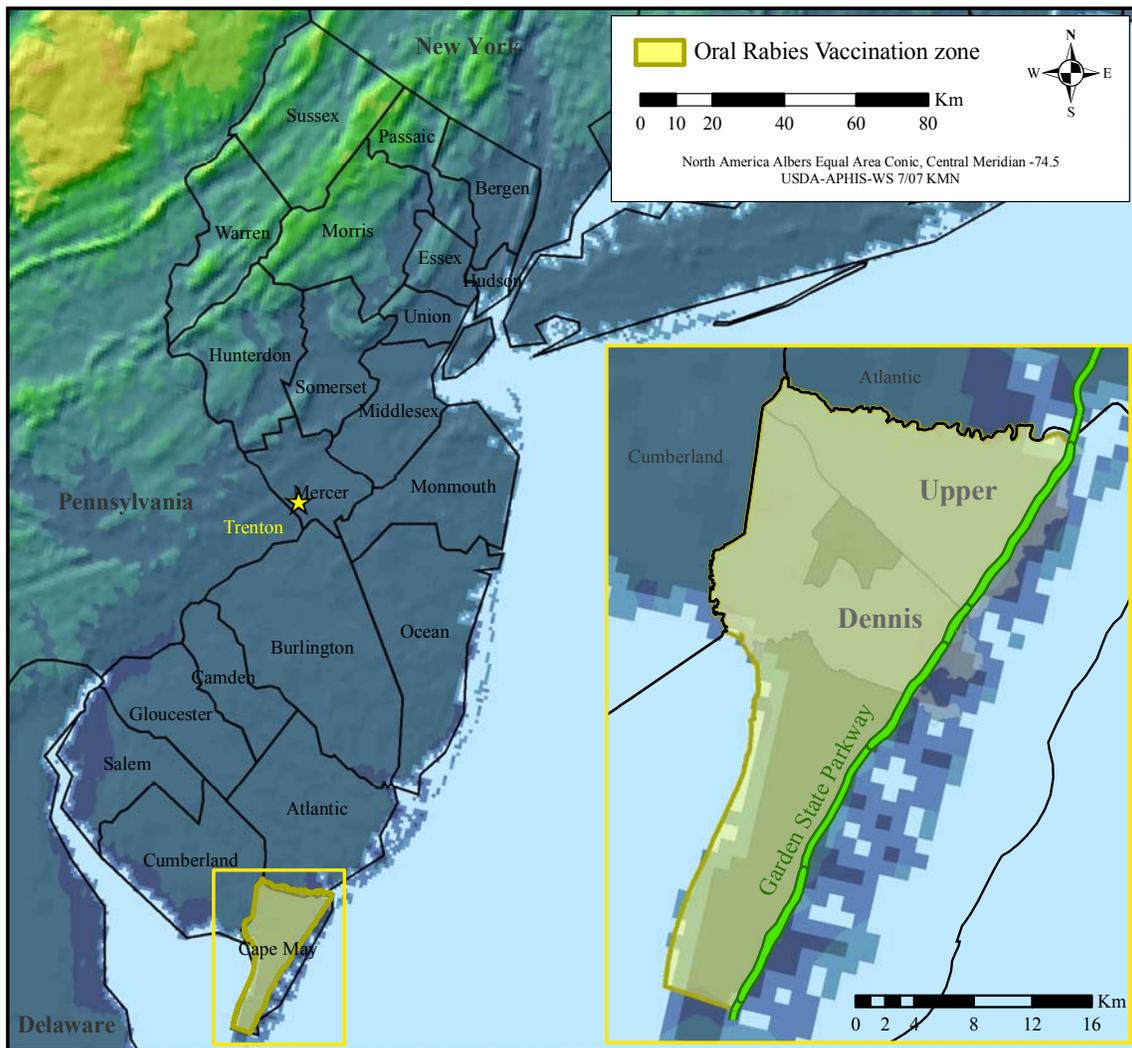


Figure 1. Wildlife Services cooperative rabies management program activities in New Jersey, 2006.

## **ORV PROGRAM 2006**

### **Bait Distribution**

In 2006, the ORV bait zone in Cape May County encompassed 556 km<sup>2</sup> (215 mi<sup>2</sup>) west of the Garden State Parkway (Figure 1). From 26 September-6 October, 45,600 ORV baits containing Raboral V-RG® vaccine (Merial Limited, Athens Georgia, USA) were distributed: 42,600 fishmeal coated sachet (CS) baits by helicopter provided by the New Jersey Mosquito Commission, and 3,000 fishmeal polymer (FMP) baits by hand. Although baits are distributed at a rate of approximately 82 baits/km<sup>2</sup>, emphasis is placed on Upper and Dennis Townships to prevent the spread of rabies from neighboring Atlantic and Cumberland Counties into Cape May County. Since 1991, approximately 348,000 baits have been distributed in Cape May County.

### **Enhanced Surveillance**

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Currently, New Jersey WS is not planning training or implementation of the dRIT.

### **Post-ORV Monitoring**

No post-ORV monitoring activities were conducted in New Jersey during 2006.

### **Rabies Laboratory Cooperation**

New Jersey WS cooperates with the NJDHSS, Public Health and Environmental Laboratory (PHEL) and the New York State Department of Health's Rabies Laboratory at the Wadsworth Center (WC).

*New Jersey Department of Health and Senior Services, Public Health and Environmental Laboratory.*--The PHEL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by County Health Officers, veterinarians, and the public). The PHEL tested 3,329 animals for the rabies virus in 2006 (Table 1), representing a 13% increase from the number of samples tested statewide in 2005. Of the animals tested in 2006, 264 were confirmed positive and of those 139 (52.7%) were raccoons. Two cows (*Bos taurus*) from Morris and Warren Counties tested positive. There were no positive cases reported from Cape May County in 2006. In addition, the PHEL tested (and confirmed rabid) 1 cat (*Felis catus*) from New York City; it is not included in these data.

Raccoons, skunks (*Mephitidae* spp.), foxes (*Canidae* spp.), and coyotes (*Canis latrans*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 81.9% of the animals tested for rabies in New Jersey in 2006 are reported by WS as "other." For more information on 2006 rabies cases in New Jersey please visit:

<http://www.state.nj.us/health/cd/documents/rabcases06.pdf>

Table 1. Animals tested for rabies by the New Jersey Department of Health and Senior Services, Public Health and Environmental Laboratory in New Jersey, 2006.

	<b>Number Tested</b>	<b>Rabies Positive</b>
Raccoons	440	139 (31.6%)
Skunks	128	45 (35.2%)
Foxes	32	8 (25.0%)
Coyotes	1	0
Other <sup>a</sup>	2,728	72 (2.6%)
<b>Total</b>	<b>3,329</b>	<b>264 (7.9%)</b>

<sup>a</sup> Other animals included: bats, bear, beaver, cats, cows, ferrets, opossums, otter, rabbits, sheep, and woodchucks.

*New York State Department of Health's Rabies Laboratory at the Wadsworth Center.*--The WC analyzes animal serum samples for rabies virus neutralizing antibodies (VNA) to evaluate the efficacy of ORV. WS did not submit any samples to the WC in 2006.

### **ORV PROGRAM 2005 and 2006 – EVALUATION**

New Jersey WS did not conduct post-ORV trapping activities in 2005 or 2006.

### **SUMMARY**

The Cape May County ORV program, although small in scale, maintains its important status as the longest continuously running ORV program in the United States. The CMDH maintains the ORV program due to its proven long term success and continued public support.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM NEW YORK 2006

### BACKGROUND

Rabies remains one of the most important zoonoses in the United States. In the past decade, wildlife rabies has reached historically high levels with a subsequent increase in human rabies post-exposure prophylaxis. An outbreak that began in the late 1970s, in the mid-Atlantic states, has been attributed to the translocation of infected raccoons (*Procyon lotor*) from Florida for use by various hunting clubs. This event is thought to have marked the beginning of a raccoon rabies outbreak that aggressively spread throughout the mid-Atlantic and northeastern United States, reaching New York in 1990. The raccoon variant of the rabies virus quickly spread and now is present throughout most of the state. In 1994, the New York State Department of Health (NYSDOH) began experimenting with the use of oral rabies vaccination (ORV) in an enzootic area of the Capital Region and was able to demonstrate a decrease in the number of rabid raccoons. This research led to the use of ORV as a rabies control technique in New York State, where 4 distinct ORV programs exist today.

*Champlain Valley.*--In 1995, after raccoon rabies made a sudden leap of 70 km from southern Essex County in the Adirondacks to mid-Clinton County, the NYSDOH initiated a point-source control plan involving the use of ORV baits. This was followed by the establishment of a Champlain Valley (CV) ORV zone to prevent further northward spread of raccoon rabies up the Lake Champlain valley into Quebec, Canada. In 1998, the NYSDOH switched from a biannual spring and fall baiting program to an annual program in August. The CV ORV zone was moved south into the enzootic area of Essex County (Figure 1) following the successful elimination of raccoon rabies in Clinton County (last case of raccoon variant rabies was in September 1997). Wildlife Services (WS) began cooperating in the CV ORV program in 1999 by providing financial and operational support.

*St. Lawrence Region.*--In 1997, raccoon rabies was documented in St. Lawrence County and the following year an epizootic was identified in the county with 138 confirmed rabies cases in terrestrial wildlife (116 raccoons, 22 striped skunks [*Mephitis mephitis*]). In 1998, a St. Lawrence Region (SLR) ORV program was implemented by Cornell University (CU), the Ontario Ministry of Natural Resources (OMNR), and WS to prevent the northward spread of raccoon rabies in New York and ultimately into Ontario, Canada. Wildlife Services has provided financial and operational support to the SLR ORV program since 1998. In July 1999, the first case of raccoon variant rabies was confirmed in Canada, just northwest of Prescott, Ontario. Since that time, the OMNR has implemented rabies control activities that in 2006 included ORV and trap-vaccinate-release (TVR) efforts (Figure 1). Also since that time, the NYSDOH has documented a marked decline in terrestrial wildlife rabies cases in St. Lawrence County. In 2000, there were only 14 cases of rabies associated with the raccoon variant (7 raccoons, 6 skunks, 1 woodchuck [*Marmota monax*]) in St. Lawrence County. Over the next 5 years, 6 cases of raccoon rabies (3 raccoons, 3 skunks) were confirmed in the county, with the last cases coming in June 2004 when 2 skunks were confirmed from Heuvelton within the existing ORV zone (Figure 1). Rabies continues to persist in Jefferson County, within the ORV zone, as there were 21 documented cases of terrestrial rabies (15 raccoons and 6 skunks) in 2006.

*Long Island.*--In August 2004, the NYSDOH confirmed raccoon rabies for the first time in Nassau County (Figure 1). By year's end, 10 rabid raccoons had been confirmed in the county. In response to this outbreak an emergency rabies surveillance and control program was initiated by the NYSDOH Zoonoses Program, WS, and the Nassau County Department of Health. High raccoon densities in conjunction with an urban environment make implementing the Long Island (LI) ORV program challenging. Bait distribution and surveillance trapping efforts are also hindered by high human populations. Rabies continued to spread across the northern portion of Nassau County during 2006. On March 16, 2006 Suffolk County became the 61<sup>st</sup> of 62 counties in New York State to confirm raccoon rabies and by year's end a total of 5 cases were documented in Suffolk County.

*Western New York.*--The OMNR has maintained a TVR zone on the Niagara peninsula of Ontario since 1994 (Figure 1). In 1995, 138 rabid raccoons were confirmed in Niagara County, New York. In response, and in an effort to prevent the westward spread of raccoon rabies into Ontario, the New York State Department of Agriculture and Markets (NYSDAM), Niagara County, and the OMNR funded an ORV program. In 1996, baiting began in Chautauqua County to initially prevent the spread of raccoon rabies into Erie County, Pennsylvania. The NYSDAM

and Chautauqua County funded this program and CU coordinated bait distribution activities. In the following years, the rabies epizootic continued in Erie County, New York (border county between Niagara and Chautauqua). In 2002, the NYSDAM, the OMNR, and WS began baiting Buffalo, New York (Erie County) using helicopters and bait stations. This collective Western New York (WNY) ORV zone has been tied to larger, national efforts to create an immune barrier from Lake Erie to the Gulf of Mexico.

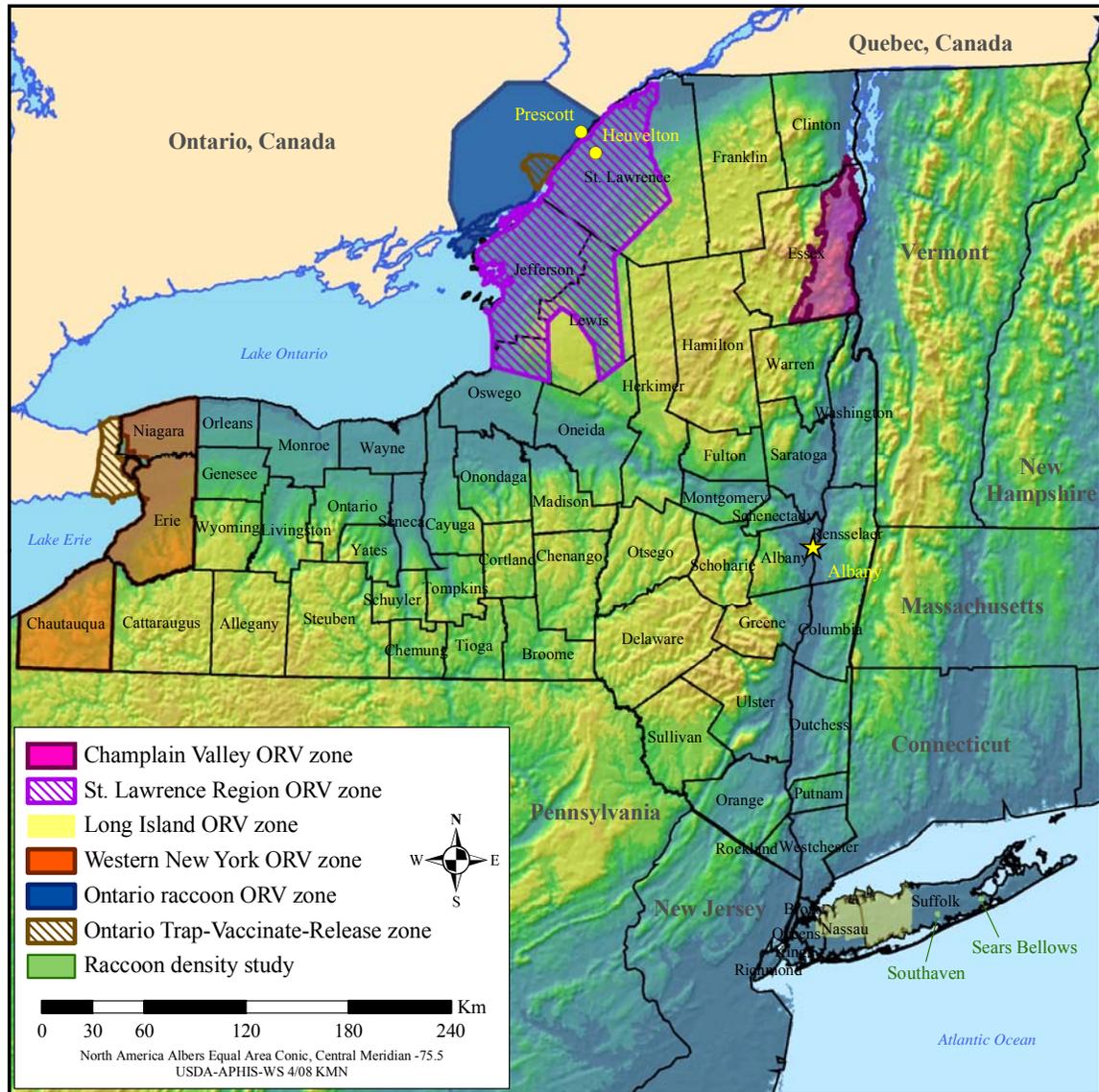


Figure 1. Wildlife Services cooperative rabies management program activities in New York, 2006.

## ORV PROGRAM 2006

### Bait Distribution

For the ninth consecutive year, WS participated in bait distribution efforts in New York; 1,579,706 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 17,908 km<sup>2</sup> (6,914 mi<sup>2</sup>) in 2006 (Figure 1). Wildlife Services has cooperated to distribute 10,522,517 ORV baits in New York since our involvement began in 1998.

*Champlain Valley.*--From 14-25 August, WS cooperated with the NYSDOH to distribute 133,000 fishmeal polymer (FMP) baits over 1909.5 km<sup>2</sup> (737.3 mi<sup>2</sup>) in Essex County. All baits were distributed aerially via helicopters provided by the New York State Police Aviation Unit (NYSPAU).

*St. Lawrence Region.*--From 25-26 August, WS cooperated with CU to distribute 474,556 ORV baits over 9616.9 km<sup>2</sup> (3713.2 mi<sup>2</sup>) in Jefferson, Lewis, Oneida, Oswego, and St. Lawrence Counties; 459,540 of those baits were coated sachets (CS) distributed by fixed wing aircraft provided by the OMNR. Ground bait distribution included 15,016 FMP baits.

*Long Island.*--From 24 September to 1 October, WS cooperated with the NYSDOH to distribute 344,949 FMP baits over 422.5 km<sup>2</sup> (163.1 mi<sup>2</sup>) in Nassau, Queens and Suffolk counties. Most baits (164,861) were distributed via ground operations by WS, state, county, and local government agencies, and community volunteers, while 137,159 baits were distributed via helicopters provided by the NYSPAU. Bait stations in Nassau and Suffolk counties distributed 32,147 and 10,782 FMP baits respectively, in areas that precluded aerial or hand distribution.

*Western New York.*--From 29-30 August, WS cooperated with CU to distribute 627,201 ORV baits over 5,958.7 km<sup>2</sup> (2300.7 mi<sup>2</sup>) in Cattaraugus, Chautauqua, Erie, Niagara, and Wyoming Counties; 522,703 of those baits were CS distributed by fixed wing aircraft provided by the OMNR, while 95,760 FMP baits were distributed via NYSPAU helicopters. Ground bait distribution included 8,738 FMP baits.

### **Enhanced Surveillance**

*St. Lawrence Region Road Kill Survey.*--The last rabid raccoon in St. Lawrence County was confirmed in March 2002. In 2006, for the second consecutive year, WS repeated an extensive road kill survey to enhance rabies surveillance and better define the prevalence and distribution of raccoon rabies in the St. Lawrence Region in preparation of shifting the ORV zone. The survey was conducted from June-August and included major roadways throughout the bait zone in Franklin, Jefferson, and St. Lawrence Counties. Over the course of a 10-week survey 24,013 km (14,921 mi) of roads were driven, resulting in 70 specimens (59 raccoons, 8 skunks, 2 gray foxes [*Urocyon cinereoargenteus*], and 1 red fox [*Vulpes vulpes*]) submitted to the NYSDOH's Rabies Laboratory at the Wadsworth Center (WC). All samples tested negative for rabies.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Currently, WS is not planning training or implementation of the dRIT because the WC is meeting enhanced surveillance testing needs.

### **Coordinated TVR**

*Western New York.*--In an effort to immunize a greater percentage of the raccoon population in the highly urbanized areas of Buffalo and Niagara Falls, and to complement ongoing TVR efforts in Ontario, Canada, Coordinated TVR was conducted in Western New York from 19-29 September. The effort focused on 2 international bridge crossings, targeting raccoons likely to use the bridges as travel routes into Canada. Following the OMNR protocol, 100 cage traps were set for 12 consecutive nights at each 2.5 km<sup>2</sup> study site. In Erie County, at the Peace Bridge, 10 raccoons and 12 skunks were captured and hand vaccinated with Imrab3® rabies vaccine (Merial Limited). In Niagara County, adjacent to the Rainbow Bridge, 50 raccoons were vaccinated.

### **Population Monitoring**

In 2006, WS replicated 2 raccoon density studies in Suffolk County (Figure 1) from 2005. The National Rabies Management Program standard protocol (50 traps set on a target study area of 3km<sup>2</sup> for 10 consecutive nights) was used and both studies coincided with post-ORV trapping in October. The studies will help determine

ORV bait densities for Suffolk County, should raccoon rabies spread and reach the population there. Both study areas were within forested county parks with campgrounds near a suburban interface. Sears Bellows County Park (eastern Suffolk County) is a 693-acre park within the Long Island pine barrens, while Southaven County Park (central Suffolk County) is a 1,323-acre pine-oak forested park. The index to raccoon density varied from approximately 8 raccoons/km<sup>2</sup> to over 22 raccoons/ km<sup>2</sup>, but was very similar to indices from 2005 (Table 1). Blood samples were collected and sent to the WC for rabies virus neutralizing antibody (VNA) testing. All raccoons were immobilized, processed and released. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

Table 1. Index to raccoon densities in Suffolk County (Long Island), New York, 2005 and 2006.

	Sears Bellows County Park		Southaven County Park	
	2005	2006	2005	2006
Trap nights	500	500	500	500
Unique raccoons	24	27	75	67
Recaptured raccoons	10	16	32	27
Total raccoons	34	43	107	94
Trap success <sup>a</sup>	4.8%	5.4 %	15.0%	13.4%
Non-target captures	39	13	24	9
Area (km <sup>2</sup> )	3.39	3.39	2.94	2.94
Raccoon density index <sup>b</sup>	7.1	7.9	25.5	22.8

<sup>a</sup> Trap success = (unique raccoons ÷ trap nights) x 100.

<sup>b</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

## Post-ORV Monitoring

*Champlain Valley.*--Wildlife Services did not conduct post-ORV monitoring activities in the Champlain Valley during 2006.

*St. Lawrence Region.*--Post-ORV sampling for the SLR ORV zone was initiated on 3 October 2006. Cage traps were used to capture 117 unique raccoons in the SLR zone. All raccoons were immobilized, processed and released.

*Long Island.*--Wildlife Services assisted the NYSDOH with post ORV evaluation trapping in October 2006 and collected blood serum and tooth samples as well as other biological data from 373 raccoons in Nassau and Suffolk Counties. All raccoons were immobilized, processed and released. All raccoons captured in the LI ORV zone in 2006 were hand vaccinated with Imrab3® rabies vaccine.

*Western New York.*--Post-ORV sampling for the WNY ORV zone was initiated on 3 October 2006. Cage traps were used to capture 82 unique raccoons. With the exception of 1 rabies suspect raccoon, all were immobilized, processed, and released. The raccoon that was euthanized tested positive for rabies.

## St. Lawrence River Study

The inability to aerially distribute baits at prescribed densities along river shorelines, in conjunction with raccoon fidelity for developed shoreline habitat, may result in a significantly under-vaccinated subset of the local raccoon population. These under-vaccinated areas may represent corridors through which raccoon rabies could spread, thereby compromising larger ORV efforts; raccoons have been documented crossing major rivers (e.g., New York WS has captured raccoons tagged in Canada by the OMNR).

In 2001, WS began a multi-year study to assess aerial ORV baiting efficacy along the St. Lawrence River shoreline (Figure 2). Flight lines have traditionally been flown perpendicular to the shoreline. To avoid a significant loss of baits in the river and striking clustered shoreline houses and residents, ORV baiting machines are turned off as aircraft approach the shoreline. Steep topography, numerous small islands, a highly developed shoreline, and a state highway paralleling the river typify the study area, posing challenges for aerial bait distribution to targeted raccoon habitat (Figure 2 inset).

To address these concerns, rabies VNA in raccoons trapped prior to annual ORV were compared to post-

ORV antibody levels in 2001 and 2002; FMP baits were used at a standard aerial baiting density of 75 baits/km<sup>2</sup> (Table 2). Serology samples collected prior to annual ORV were from areas that had been baited in previous years. In 2003, the river study continued to be baited from aircraft but was supplemented by ground baiting using FMP baits at 75 baits/km<sup>2</sup>. These data were compared to aerial baiting only from previous years (Table 2). In 2004, the study mimicked 2003, but the ground baiting density was increased to 150 baits/km<sup>2</sup> and a second study area was established along the river to compare CS baits to FMP baits. The new CS study area was divided into 2 subsections. Both sections were aerially baited but supplemented with ground baiting; 1 section at 75 baits/km<sup>2</sup>, the other at 150 baits/km<sup>2</sup> (Table 2).

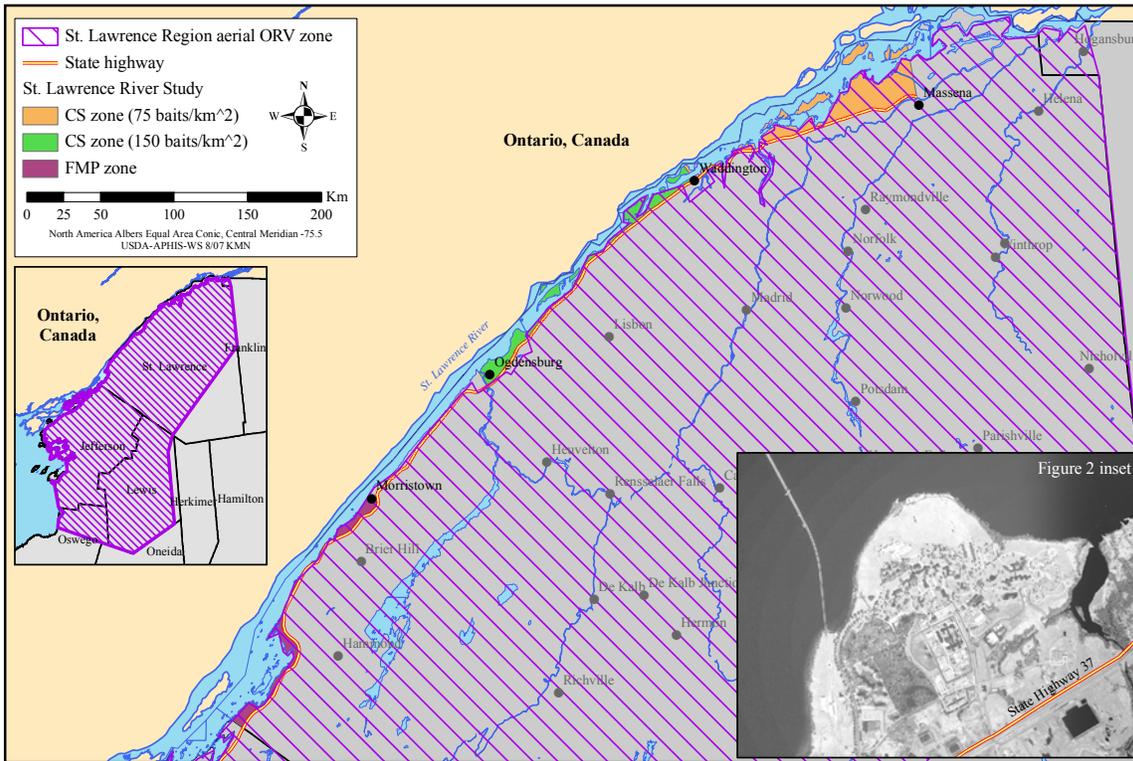


Figure 2. St. Lawrence River study area showing different oral rabies vaccine (ORV) bait types and bait densities, 2001-2005; and typical river shoreline habitat with clustered residential housing (inset).

Table 2. Results of raccoon biological samples collected by Wildlife Services during the St. Lawrence River Study in New York, 2001-2005.

	2001 Pre <sup>a</sup>	2001 Post	2002 Pre	2002 Post	2003 Post	2004 Post	2004 Pre	2004 Post	2004 Post	2005 Post
ORV bait type <sup>b</sup>	FMP	FMP	FMP	FMP	FMP	FMP	CS	CS	CS	CS
Bait density/km <sup>2</sup>	75	75	75	75	75	150	75	75	150	75
Bait distribution method	aerial	aerial	aerial	aerial	ground <sup>c</sup>	ground <sup>c</sup>	aerial	ground <sup>c</sup>	ground <sup>c</sup>	aerial
Weeks post-ORV	48	4-8	44	4-8	4-8	4-8	32	4-8	4-8	4-8
<b>Serology</b>										
Testable blood samples	76	32	58	73	81	80	113	51	48	45
Positive rabies antibody response (≥0.05 IU)	11 (14.5%)	7 (21.9%)	17 (29.3%)	19 (26.0%)	40 (49.4%)	25 (31.3%)	29 (25.7%)	11 (21.6%)	9 (18.8%)	12 (26.7%)
<b>Tetracycline biomarker</b>										
Testable tooth samples	69	33	50	69	81	77	n/a <sup>d</sup>	n/a <sup>d</sup>	n/a <sup>d</sup>	20 <sup>d</sup>
Presence of tetracycline biomarker	21 (30.4%)	10 (30.3%)	19 (38.0%)	23 (33.3%)	42 (51.9%)	53 (68.8%)				13 (65.0%)

<sup>a</sup> Pre means samples were collected prior to that year's bait distribution, but samples were collected in an area that had been previously baited.

<sup>b</sup> ORV=oral rabies vaccination; FMP=fishmeal polymer; CS=fishmeal coated sachet.

<sup>c</sup> Ground baiting was conducted in addition to normal aerial baiting at 75 baits/km<sup>2</sup>.

<sup>d</sup> Teeth not tested because coated sachet baits do not contain a tetracycline biomarker.

<sup>e</sup> Only teeth >1 year old were aged and analyzed for biomarker in 2005.

Information collected during the River Study was used to: identify the percentage of raccoons in the study areas with detectable levels of rabies antibodies, identify the number of animals that consumed baits, and assess the impact of ground baiting to augment aerial bait distribution in shoreline habitat. Aerial flight lines are now flown parallel to the river. The results of this study may have broader implications to national ORV programs, as increased human activity is associated with shorelines.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 126 opossums (*Didelphis virginiana*), 33 gray squirrels (*Sciurus carolinensis*), 29 Norway rats (*Rattus norvegicus*), 15 domestic/feral cats (*Felis catus*), 9 Eastern cottontails (*Sylvilagus floridanus*), 2 purple finches (*Carpodacus purpureus*), 2 red squirrels (*Tamiasciurus hudsonicus*), and 2 woodchucks (*Marmota monax*).

### Rabies Laboratory Cooperation

New York WS cooperates with the NYSDOH's Rabies Laboratory at the WC in support of the ORV programs throughout New York. The WC cooperates in rabies surveillance efforts by testing brainstems for the rabies virus. They also collaborate in ORV evaluation efforts by testing wildlife serum for rabies VNA.

*Rabies Virus Testing.*--The WC tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure, usually submitted by Animal Control Officers, Conservation Officers, county health departments, veterinarians, and the public) and enhanced surveillance (specimens not involved in an exposure and usually submitted by WS). The WC received 8,928 animals for rabies testing in 2006, representing a 7.3% increase from the number of samples received statewide in 2005. In 2006, animals were submitted to the WC from 57 of 62 counties throughout the state, including 14 of 15 ORV counties (all but Queens County). The New York City Department of Health and Mental Hygiene (NYCDH) examines rabies specimens from the 5 New York City counties: Bronx, Kings, New York, Queens, and Richmond. The WC confirmed 612 cases of rabies statewide in 2006; of these, 13.5% were raccoons from counties treated with ORV (Table 3). The NYCDH confirmed 43 cases of rabies (out of 821 specimens received). In addition, 1 raccoon from Cattaraugus County was diagnosed in Pennsylvania and 1 cat from Staten Island (Richmond County) was diagnosed in New Jersey. None of these data are included in Table 3. In total, there were 657 confirmed cases of rabies from New York State in 2006.

Raccoons, skunks, and foxes are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 79.6% of the animals submitted to the WC for rabies testing in New York in 2006 are reported by WS as "other." For a detailed listing of current and historical rabies data from New York State please visit:

<http://www.wadsworth.org/rabies/index.htm>

Table 3. Rabies positives/animals submitted for rabies testing to the New York State Department of Health's Rabies Laboratory at the Wadsworth Center in New York, 2006.

	Statewide <sup>a</sup>	Champlain Valley <sup>b</sup> ORV counties	St. Lawrence Region <sup>b</sup> ORV counties	Long Island <sup>b</sup> ORV counties	Western New York <sup>b</sup> ORV counties
Raccoons	320/1349	1/4	22/164	23/485	37/135
Skunks	95/303	3/6	13/46	0/0	9/24
Foxes	31/172	0/0	7/22	0/2	2/8
Other <sup>c</sup>	166/7104	0/53	19/793	3/254	11/1029
Total	612/8928	4/63	6/1025	26/741	59/1196

<sup>a</sup> A small number of submissions (but no positives) were from outside of New York State.

<sup>b</sup> Champlain Valley: Essex County; St. Lawrence Region: Franklin, Jefferson, Lewis, Oneida, Oswego, and St. Lawrence Counties; Long Island: Nassau and Suffolk Counties (Queens County is not represented in this table because the New York City Department of Health and Mental Hygiene tests rabies specimens from Queens County); Western New York: Cattaraugus, Chautauqua, Erie, Niagara, and Wyoming Counties.

<sup>c</sup> Other animals included: bats, beavers, cats, cows, coyotes, deer, dogs, horses, mules, lagomorphs, rodents, woodchucks, and other domestic and wild animals not listed.

*Rabies Virus Neutralizing Antibody Testing.*--The WC also analyzed 293 blood serum samples for rabies VNA submitted by New York WS in 2006. This represented a slight increase over the 252 samples submitted in 2005. New York WS anticipates approximately 200-300 serum sample submissions to this laboratory in 2007.

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year’s monitoring to provide time for laboratory analyses. At the time of this report however, the current year’s (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

As the lead agency on the CV and LI ORV zones, the NYSDOH reports those data. Wildlife Services conducted live-trapping activities to evaluate the success of the SLR ORV zone in 2005 and the SLR and WNY zones in 2006. During the evaluation phases, WS live-trapped 101 raccoons in the 2005 SLR zone and 117 and 82 raccoons in the SLR and WNY zones in 2006 (Table 4). Blood and tooth samples were collected from most of these animals and serum samples were sent to the WC, while tooth samples were sent to Matson’s Laboratory LLC (Milltown, Montana, USA). The tooth samples collected in 2006 were not analyzed for presence of tetracycline biomarker because FMP baits have not been aerially distributed in this area since 2003.

Table 4. Serology results of raccoon blood samples collected by Wildlife Services during the evaluation phase of the St. Lawrence Region (SLR) and Western New York (WNY) oral rabies vaccination (ORV) program in New York, 2005-2006<sup>a</sup>.

	SLR 2005	SLR 2006	WNY 2006
Bait density/km <sup>2</sup>	75	75	75
Bait distribution method	aerial	aerial	aerial
Weeks post-ORV	4-8	4-8	4-8
Unique raccoons	101	117	82
		<b>Serology</b>	
Testable blood samples	101	117	81
Positive rabies antibody response ( $\geq 0.05$ IU)	28 (27.7%)	21 (17.9%)	22 (27.1%)

<sup>a</sup> Tooth samples were not analyzed for tetracycline biomarker presence because coated sachet baits do not contain tetracycline and fishmeal polymer baits (which do) have not been aerially distributed in the SLR ORV zone since 2003.

*Age Results.*--In 2005 and 2006, 161 and 202 raccoon teeth, respectively, were aged using premolars of live-captured animals and canines of animals found dead or euthanized (Figure 3). These samples were collected from raccoons in the SLR and WNY ORV zones, as well as the Suffolk County density studies. The population was dominated by young of the year juveniles (<1 y.o.) and 1 year old raccoons. One raccoon was 14 years old.

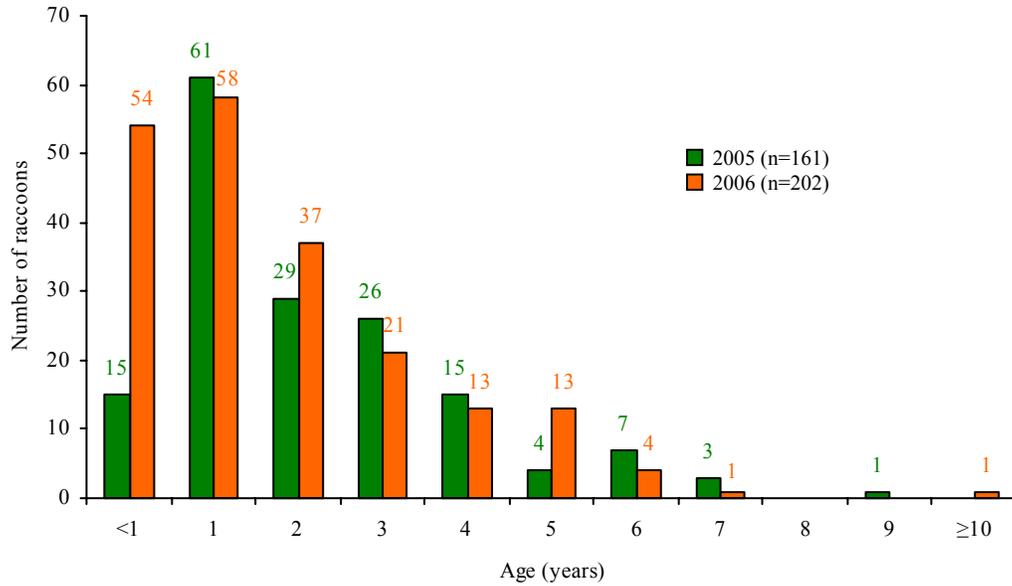


Figure 3. Age results of 161 and 202 raccoon tooth samples collected in the St. Lawrence Region and Western New York oral rabies vaccination zones, and during density studies on Long Island, New York, 2005 and 2006.

## SUMMARY

In 2006, New York WS conducted its ninth year of rabies control activities in the SLR ORV zone. Wildlife Services also provided financial support for the second time in the WNY ORV zone and for the third year in the LI ORV zone. Over 1.5 million baits were distributed across the 4 ORV zones to stop the spread of raccoon rabies. While the CV, SLR, and WNY zones are part of a larger northeastern rabies control effort that included Vermont, New Hampshire, Maine and Ontario, Quebec, and New Brunswick, Canada in 2006, the LI ORV program remains critical to limiting raccoon rabies on Long Island.

In 2007, WS will continue to enhance rabies surveillance in the SLR ORV zone, perform raccoon density studies on Long Island, and conduct post-ORV monitoring and evaluation in all 4 zones. Future ORV baiting strategies in New York will continue to be tied to national and international planning efforts to contain the disease and explore strategies to eliminate the raccoon variant of the rabies virus from North America.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM NORTH CAROLINA 2006

### BACKGROUND

Nettles et al. (1979) reported rabies positive raccoons (*Procyon lotor*) being translocated into North Carolina from Florida by raccoon hunting clubs. It was not until 1991, however, that significant numbers of rabid raccoons from the wild were being reported. The first counties experiencing the increase occurred in the northeastern portion of the state as the mid-Atlantic epizootic crossed into North Carolina. By 1992 it was evident that a second distinct epizootic front crossed into North Carolina in the southeastern portion of the state, with several cases showing up in Brunswick and Bladen Counties. In 1993, the epizootic had clearly moved into the southern piedmont and coastal plains. During 1995, a third distinct epizootic outbreak occurred when the disease spread into the northwestern corner of the state, from southwestern Virginia. North Carolina has thus become the meeting point for 3 waves of the raccoon variant of the rabies virus. Presently, 97 of 100 counties have reported cases of raccoon rabies with two additional counties being added in 2006 (Mitchell and Macon). The remaining 3 counties not reporting cases are in the western portion of the state (Figure 1). They are all in rural areas and do not have active animal control agencies, likely contributing to the fact that they have not reported any cases of raccoon rabies. In 2005, rabies positive cases were reported in Tennessee just west of the North Carolina border along the Nolichucky River basin (Figure 1). These cases, and the increasing number of positives from North Carolina border counties (especially Yancey County), confirmed that the epizootic was breaching the Appalachian Ridge (AR) oral rabies vaccination (ORV) zone along the major river valleys. In an attempt to control this breach, Wildlife Services (WS) decided to extend the AR zone into North Carolina in 2005, along both the Nolichucky and French Broad River basins. In 2006, the ORV zone was widened slightly into Jackson and Swain Counties (Figure 1).

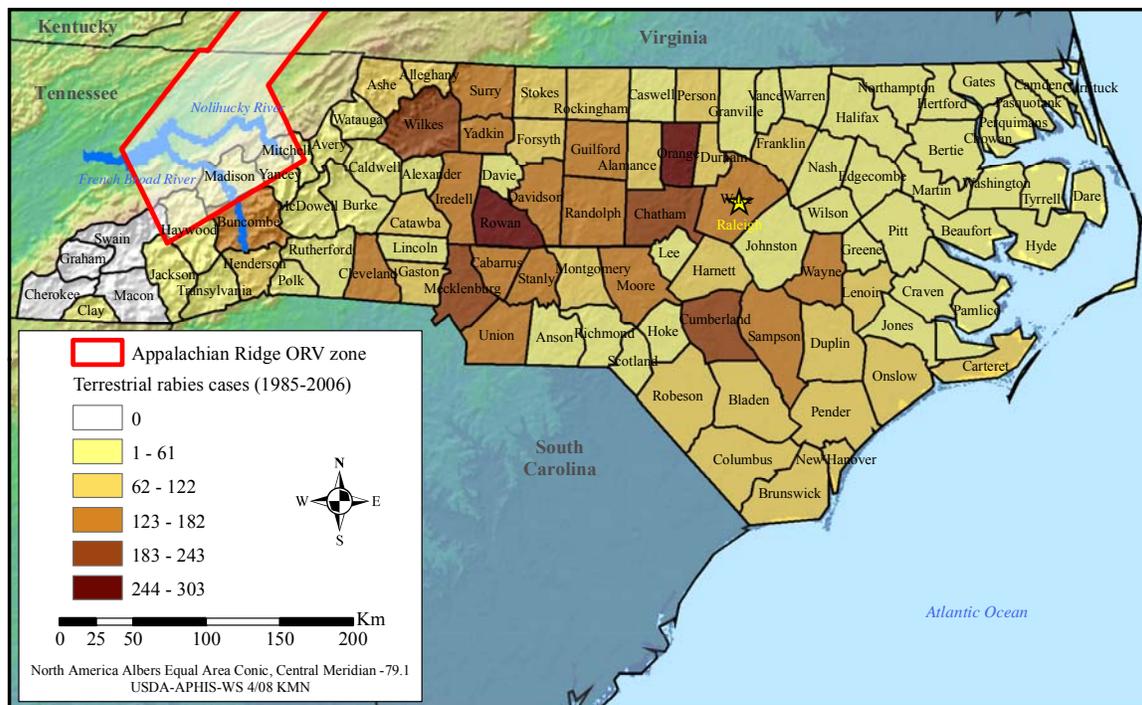


Figure 1. Wildlife Services cooperative rabies management program activities in North Carolina, 2006.

### ORV PROGRAM 2006

#### Bait Distribution

The 2006 season marked the second year WS participated in bait distribution efforts in North Carolina; 184,932 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 2,888.5 km<sup>2</sup> (1115.3 mi<sup>2</sup>). The ORV zone included portions of Buncombe, Haywood, Jackson, Madison, Mitchell,

Swain and Yancey Counties. The ORV zone was expanded in 2006 to include portions of Jackson and Swain Counties and a larger portion of Mitchell County. During 7-16 August bait distribution efforts, 182,772 fishmeal coated sachets (CS) were distributed by fixed-wing aircraft (provided by Dynamic Aviation Group Inc. [Bridgewater, Virginia, USA]), while 2,160 fishmeal polymer (FMP) baits were distributed by hand (in urban areas of Madison, Mitchell, and Yancey Counties).

### **Enhanced Surveillance**

North Carolina WS initiated an enhanced rabies surveillance program in 2004 to better document the extent of rabies cases near the western front of the disease (along the North Carolina-Tennessee border). All of the Tennessee border counties are along the AR, thought to pose a potential natural barrier to the westward spread of raccoon rabies. Historically, only 4 of 10 border counties had active animal control agencies and many used regional health departments rather than county level departments. The lack of animal control personnel, coupled with the rural nature of the area, contributed to sporadic rabies surveillance efforts as compared to the rest of the state.

An increase in rabid animals in Yancey County in 2005 (16 animals compared to 1 case of rabies in 2004) prompted the county to add a full time animal control person to their staff. In addition, WS made a half-time position (filled in 2004 to enhance rabies surveillance) a full-time position in 2005. This Wildlife Specialist worked with local authorities and to collect 105 specimens in 2006: 89 raccoons, 14 gray foxes (*Urocyon cinereoargenteus*), 1 striped skunk (*Mephitis mephitis*), and 1 coyote (*Canis latrans*). Cooperators were able to freeze carcasses of road killed and suspect rabid animals, while completing a data sheet containing basic biological information about the animal.

*Direct Rapid Immunohistochemistry Test (dRIT)*-- The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Wildlife Services personnel prepared the carcasses (head/brainstem removal, etc.) and tested all 105 samples using the dRIT in our lab in Raleigh, confirming 25 rabid animals: 23 (25.0% raccoons, 1 (7.1%) gray fox, and 1 (100%) striped skunk. All positive samples were confirmed as the raccoon variant of the rabies virus by the Centers for Disease Control and Prevention (CDC). Ten percent of the negatives were submitted for confirmation. The CDC (using the dFA test) had 100% agreement with the WS dRIT results for positive and negative samples. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in North Carolina.

### **Post-ORV Monitoring**

From 5-22 September 2006, WS conducted post-ORV trapping activities in Buncombe, Haywood, Madison, Mitchell, and Yancey Counties. Blood serum samples from 121 raccoons were collected to measure the presence of rabies virus neutralizing antibody (VNA) response in this ORV zone. Tooth samples from 104 of those raccoons were also collected and submitted for age analysis. All raccoons but 3 (118) were immobilized, processed and released. Three raccoons were euthanized because they were showing neurological symptoms consistent with rabies. Wildlife Services tested them using the dRIT and 1 was positive for rabies. The CDC confirmed it as raccoon variant. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

### **Non-target Captures**

Non-target animals captured and released by WS in 2006 included: 152 opossums (*Didelphis virginiana*), 24 domestic cats (*Felis catus*), 4 domestic dogs (*Canis familiaris*), 4 Eastern box turtles (*Terrapene carolina*), 4 woodchucks (*Marmota monax*), 3 Eastern woodrats (*Neotoma floridana*), 2 striped skunks, and 1 Eastern cottontail (*Sylvilagus floridanus*).

## Rabies Laboratory Cooperation

Wildlife Services' ORV program in North Carolina cooperates with the North Carolina Department of Health and Human Services, State Health Laboratory (SHL), and the CDC.

*North Carolina Department of Health and Human Services, State Health Laboratory.*--The SHL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure to the rabies virus). The SHL received 4,257 samples for rabies virus testing in 2006 and confirmed 521 positives (Table 2). In recent years, the SHL has conducted rabies virus typing on all non-raccoon terrestrial wildlife samples; they have all been confirmed as raccoon variant.

Raccoons, foxes, skunks, coyotes, and bobcats (*Lynx rufus*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 78.2 % of the animals tested for rabies by the SHL in 2006 are reported by WS as "other." For a full listing of rabid animals in North Carolina by county and species from 1990 to the present please visit:

<http://www.epi.state.nc.us/epi/rabies/state.html>

Table 2. Animals submitted and tested for rabies by the North Carolina Department of Health and Human Services, State Health Laboratory in North Carolina, 2006.

	Submissions	Rabies Positive
Raccoons	658	298 (45.3%)
Foxes	139	50 (36.0%)
Skunks	115	91 (79.1%)
Coyotes	9	2 (22.2%)
Bobcats	9	5 (55.6%)
Other <sup>a</sup>	3,327	75 (2.3%)
Total	4,257	521 (12.2%)

<sup>a</sup> Other animals included: alpaca, bat, bear, beaver, buffalo, cat, coatimundi, cow, deer, degu, dog, donkey, equine, ferret, gerbil, goat, guinea pig, hamster, kangaroo, human, llama, leopard, mink, mouse, muskrat, nutria, ocelot, opossum, otter, pig, prairie dog, rabbit, rat, rodent, sheep, squirrel, vole, weasel, wolf, wolf-dog hybrid, and woodchuck (groundhog).

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies VNA. North Carolina WS submitted 121 blood serum samples for rabies VNA analysis to the CDC in 2006 and 163 in 2005. Results of samples submitted in 2005 and 2006 are shown in the following section. The North Carolina ORV program anticipates similar numbers of serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In July 2005, WS live-trapped 30 raccoons during ORV naïve monitoring prior to the first ORV bait distribution effort in North Carolina; 1 (3.3%) of these raccoons showed presence of rabies VNA ( $\geq 0.05$  IU).

During September 2005, 4-5 weeks post-ORV bait distribution, WS live-trapped 133 raccoons with 9 (6.8%) showing presence of rabies VNA. Tooth samples were collected from 104 raccoons and sent to Matson's Laboratory LLC (Milltown, Montana, USA) for age analysis. In addition, 37 raccoons were aged in the fall as young of the year without tooth extraction.

During September 2006, 4-5 weeks post-ORV, WS live-trapped 121 raccoons and 16 (13.2%) showed presence of rabies VNA. Tooth samples were collected from 104 raccoons and aged at Matson's, while 5 were aged as <1 y.o. without tooth extraction. The populations in both years were dominated by young ( $\leq 1$  y.o.) raccoons (Figure 2). In both years, teeth were not tested for tetracycline because CS baits were distributed.

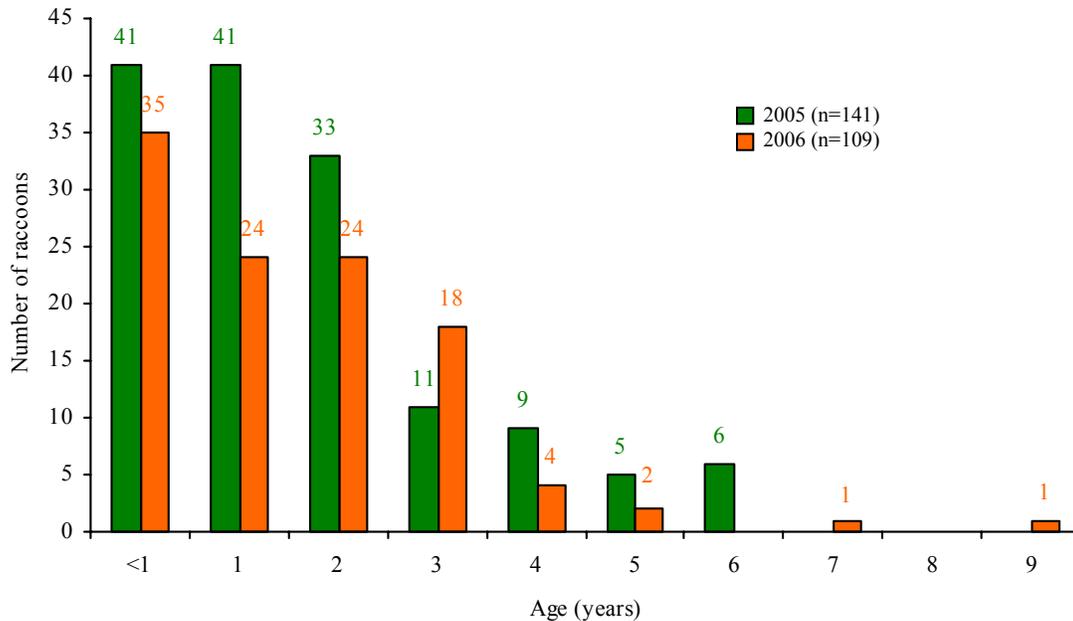


Figure 2. Age class distribution of 141 and 109 raccoons collected by Wildlife Services during the cooperative rabies management program in North Carolina, 2005 and 2006.

## SUMMARY

During 2006, WS completed its third year of cooperative participation in rabies management in North Carolina, and its second year conducting ORV bait distribution activities. Other activities in 2006 included enhanced surveillance of raccoon rabies and post-ORV monitoring and evaluation in western North Carolina along the Tennessee border. Future ORV baiting strategies in North Carolina will continue to be directed towards halting the spread of raccoon rabies into the western U.S. The North Carolina ORV zone will continue to be tied to national and international planning efforts to contain the disease and explore strategies to eliminate the raccoon variant of the rabies virus from North America.

## LITERATURE CITED

Nettles, V. F., J. H. Shaddock, R. K. Sikes, and C. R. Reyes. 1979. Rabies in translocated raccoons. *Am. J. Public Health* 69:601-602.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM OHIO 2006

### BACKGROUND

The raccoon (*Procyon lotor*) variant of the rabies virus was first documented in Ohio in 1996 (Mahoning County). Rabies cases continued to escalate and in April 1997 an epizootic of raccoon rabies was identified in northeastern Ohio, with 62 positive cases by year's end. Due to this epizootic and a peak in public interest, an oral rabies vaccination (ORV) program was initiated in Ohio in an attempt to prevent the further westward spread of raccoon rabies. The original ORV bait zone included Trumbull, Mahoning, and Columbiana Counties, and encompassed 1,780 km<sup>2</sup> (688 mi<sup>2</sup>). With increased surveillance, raccoon rabies cases were confirmed outside the ORV zone. Subsequently, in the fall 1999, the ORV zone more than tripled in size to include Ashtabula, Trumbull, Mahoning, Columbiana, Carroll, and Jefferson Counties, encompassing 6,497 km<sup>2</sup> (2,509 mi<sup>2</sup>). By 2004, the ORV zone had nearly doubled again to 11,845 km<sup>2</sup> (4,573 mi<sup>2</sup>) and included Ashtabula, Trumbull, Mahoning, Columbiana, Carroll, Jefferson, Harrison, Belmont, Monroe, and Washington Counties. Despite this long-standing ORV zone, in July 2004, a rabid raccoon (with raccoon variant) was confirmed 10.6 km (6.6 mi) west of the zone in Lake County, marking the western-most case of raccoon rabies in Ohio. This case triggered a contingency action response (that included enhanced rabies surveillance via local raccoon population reduction, trap-vaccinate-release, and ORV bait distribution) over 2,471 km<sup>2</sup> (954 mi<sup>2</sup>) of Cuyahoga, Geauga, Lake, Portage, and Summit Counties. In 2006, Ohio maintained both the contingency action (CA) and the historic Appalachian Ridge (AR) ORV zones (Figure 1), which are part of a larger zone that stretches from Lake Erie to the Appalachian Mountains of Tennessee and North Carolina.

The Ohio Department of Health (ODH) is the lead agency for Ohio's ORV program. Wildlife Services (WS) is an active cooperator, now providing a major source of cooperative funding and federal wildlife management leadership. Additional cooperators include the Ohio Department of Agriculture (ODA), the Ohio Department of Natural Resources (ODNR) Division of Wildlife, the Centers for Disease Control and Prevention (CDC), Ohio State University, the Ohio Department of Transportation, and local/county health departments.

### ORV PROGRAM 2006

#### Bait Distribution

Wildlife Services participated in bait distribution efforts in eastern Ohio for the tenth consecutive year in the AR ORV zone and for the third year in the CA ORV zone; 1,110,046 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 12,165 km<sup>2</sup> (4,697 mi<sup>2</sup>) in 2006 (Figure 1). Target bait density for the Ohio ORV zones is 75/km<sup>2</sup>. Since its program inception in 1997, WS has cooperated to distribute 9,404,700 ORV baits in Ohio.

*Contingency Action.*--In April 2006, the CA ORV zone in Cuyahoga, Geauga, Lake, Portage, and Summit Counties was baited for the third year (initially baited in September 2004). From 19-20 April, 243,232 ORV baits were distributed over 2,984 km<sup>2</sup> (1,152 mi<sup>2</sup>): 137,932 fishmeal coated sachet (CS) baits via fixed-wing aircraft; 91,980 fishmeal polymer (FMP) baits via ground operations (hand baiting); and 13,320 FMP baits via helicopter. Fixed-wing aircraft and flight crew were provided by the Ontario Ministry of Natural Resources (OMNR), while helicopters and flight crew were provided by the ODNR. Ground and hand baiting support was provided by WS, the ODH, the ODNR, and county/local health departments. The CA ORV zone was baited again in September 2006 as part of the larger AR ORV zone (see *Appalachian Ridge* below).

*Appalachian Ridge.*--In 2006, the Ohio portion of the AR ORV zone was expanded to include all of Belmont County and covered 12,165 km<sup>2</sup> (4,697 mi<sup>2</sup>) in 16 counties (Figure 1). From 5-21 September, 866,814 ORV baits were distributed: 642,894 CS baits via fixed-wing aircraft; 166,680 FMP baits via hand baiting; and 57,240 FMP baits via helicopter. Fixed-wing aircraft and flight crew were provided by the OMNR, while helicopters and flight crew were provided by the ODNR. Ground and hand baiting support was provided by WS, the ODH, the ODNR, the ODA, the Ohio National Guard, and county/local health departments.

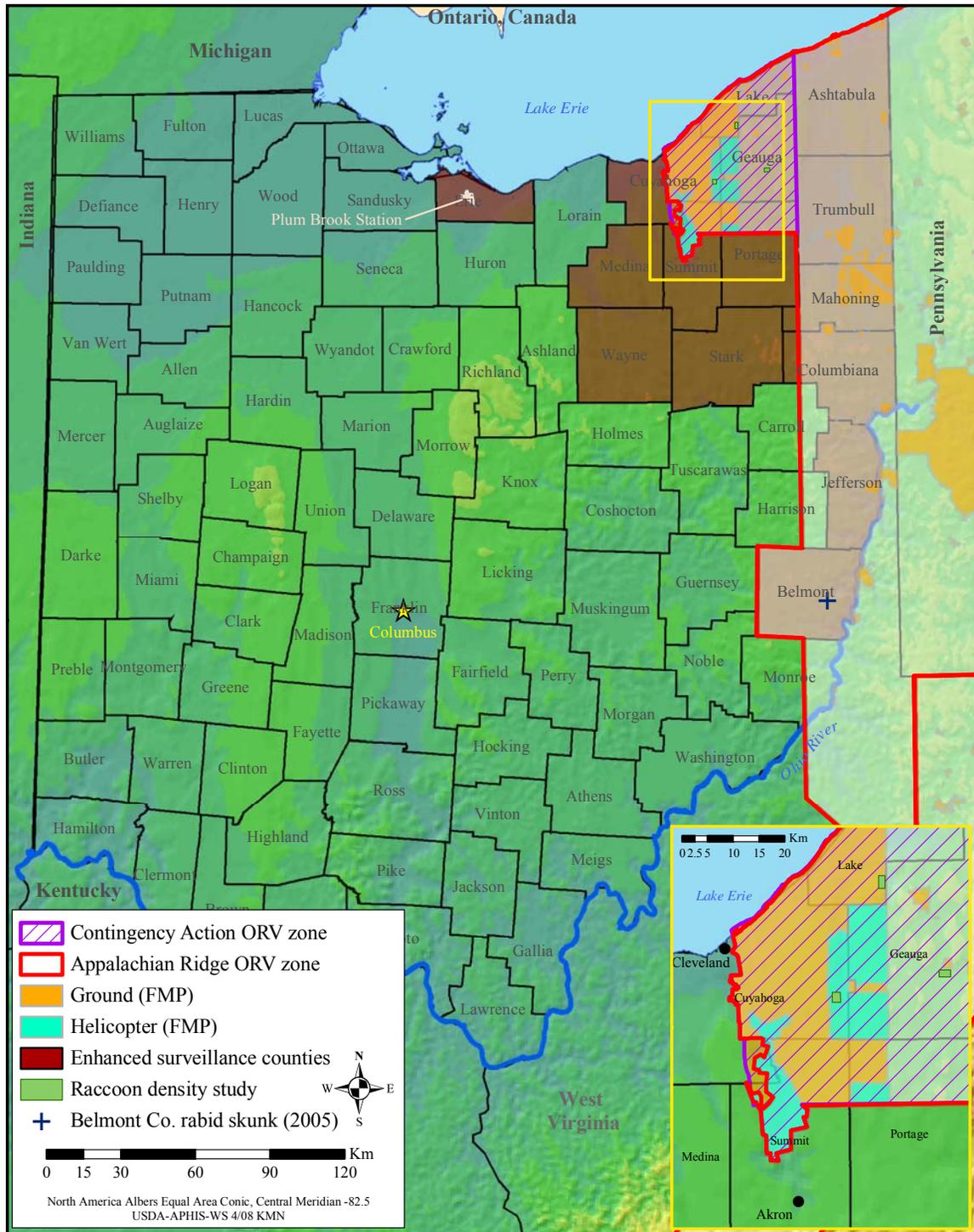


Figure 1. Wildlife Services cooperative rabies management program activities in Ohio, 2006 (coated sachet baits are distributed via fixed-wing aircraft in both zones unless otherwise noted [FMP=fishmeal polymer]).

### Enhanced Surveillance

In 2006, WS continued to enhance surveillance of raccoon rabies by collecting, submitting, and testing suspect rabid animals from counties in or near the Ohio ORV zones (Figure 1). Wildlife Services collected animals by the following methods: 1) humanely euthanizing (according to the American Veterinary Medical Association’s

Panel on Euthanasia recommendations) raccoons that had puncture wounds/bite marks, exhibited disorientation, or showed signs of illness during trapping activities; 2) conducting road kill surveillance and collecting mammals in suitable testing condition; 3) obtaining wildlife reported by Ohio residents as displaying rabies-like symptoms; and 4) obtaining wildlife from nuisance wildlife control officers. Wildlife Services also continued to work with state and local health departments to increase the number of enhanced surveillance samples for rabies testing.

As a result of enhanced surveillance efforts throughout Ohio, WS cooperated to collect and submit 609 animals for rabies testing: 563 raccoons, 21 striped skunks (*Mephitis mephitis*), 9 domestic/feral cats (*Felis catus*), 1 coyote (*Canis latrans*), 6 red foxes (*Vulpes vulpes*), 4 woodchucks (*Marmota monax*), 1 fox squirrel (*Sciurus niger*), 1 mink (*Mustela vison*), and 3 opossums (*Didelphis virginiana*). Of those samples, 481 (79.0%) came from counties in or adjacent to the 2004 CA ORV zone.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

During 2006, Ohio WS personnel continued to utilize the dRIT by testing all 609 samples that were collected during enhanced surveillance activities and confirming 4 positive animals (Table 1). All positives and 10% of all negatives were sent to the CDC for confirmation and variant typing. The CDC had 100% agreement with WS dRIT results and confirmed the 4 positives as raccoon rabies variant. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Ohio.

Table 1. Animals tested for rabies by Wildlife Services using the Direct Rapid Immunohistochemistry Test (dRIT) in or adjacent to ORV counties in Ohio, 2006 (raccoon rabies variant positives in parentheses).

County	Raccoon	Skunk	Red fox	Coyote	Feral cat	Other <sup>a</sup>	Total
Ashtabula	2						2
Belmont	86						86
Columbiana	5		1				6
Cuyahoga	140 (1)	10	1	1	6	3	161 (1)
Erie <sup>b</sup>	6						6
Geauga	101						101
Jefferson	1						1
Lake	83 (3)	8	2		2	4	99 (3)
Mahoning	26		1				27
Medina <sup>b</sup>	29	1	1				31
Portage	51	1			1		53
Stark <sup>b</sup>						1	1
Summit	28						28
Trumbull	4	1				1	6
Wayne <sup>b</sup>	1						1
Total	563 (4)	21	6	1	9	9	609 (4)

<sup>a</sup> Other animals included: 1 fox squirrel, 1 opossum, and 1 woodchuck in Cuyahoga Co.; 2 opossums and 2 woodchucks in Lake Co.; 1 woodchuck in Stark Co.; and 1 mink in Trumbull Co.

<sup>b</sup> ORV not applied in this county.

## Population Monitoring

In 2006, WS conducted 3 raccoon density studies using the National Rabies Management Program (NRMP) protocols; 1 in Cuyahoga, 1 in Geauga and 1 in Lake Counties. The Lake and Cuyahoga studies provided an index to raccoon densities in urban/suburban environments, representative of habitat found throughout the CA ORV zone. During 2 studies, the NRMP standard protocol for indexing raccoon density was used (50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights), and the NRMP high density protocol (traps set for 15

consecutive nights) was used during the study in Cuyahoga County. The Cuyahoga study was replicated from 2005 and the index to density was similarly high (34.3 raccoons/km<sup>2</sup> compared to 41.6/km<sup>2</sup> in 2005). During the 3 studies, 259 unique raccoons were captured and blood and tooth samples were collected from most of them (Table 2). One raccoon was found dead in a field during the Lake County study; 143 raccoons were immobilized, processed and released; and 116 raccoons were immobilized, processed, euthanized and tested to enhance rabies surveillance. One raccoon from the Cuyahoga County study tested positive for rabies (Table 1). All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

Table 2. Index to raccoon densities in Geauga, Lake, and Cuyahoga Counties in Ohio, 2006.

	<b>Geauga</b>	<b>Lake</b>	<b>Cuyahoga 2006</b>
Time of study	17-27 Jul	17-27 Jul	31 Jul- 15 Aug
Weeks post-ORV	12-13	12-13	14-15
Macrohabitat	Agricultural	Urban/suburban	Urban/suburban
Target trap nights	500	500	750
Trap type	cage trap	cage trap	cage trap
Unique raccoons	69	82	108
Recaptured raccoons	18	26	0
Non-target captures	15	20	14
Area (km <sup>2</sup> )	3.11	3.17	3.15
Raccoon density index <sup>a</sup>	22.2	25.9	34.3

<sup>a</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

### **ORV Naïve Monitoring**

Wildlife Services refers to areas that have never been treated with ORV as “ORV naïve.” On 21 August 2006, WS initiated a raccoon live trapping effort in an ORV naïve area of Belmont County. Trapping was conducted 2 weeks prior to the 2006 ORV distribution in this area. Samples were collected from 24 raccoons over 150 trap nights. All raccoons were immobilized, processed, euthanized and tested to enhance rabies surveillance; they all tested negative (Table 1).

### **Post-ORV Monitoring**

Post-ORV sampling for the spring baiting of the CA ORV zone was initiated on 22 May 2006. Results from this trapping effort will help determine the efficacy of CS versus FMP baits (FMPs were distributed via hand baiting and helicopter operations in urban areas). Cage traps were used to capture 134 unique raccoons over 774 trap nights. Eighty four raccoons were immobilized, processed and released, while 50 raccoons were euthanized and tested to enhance rabies surveillance; they all tested negative (Table 1).

Post-ORV sampling for Ohio's fall baiting of the AR ORV zone (which included a repeat of the CA ORV zone) was initiated on 2 October 2006. Trapping efforts were concentrated in Belmont County (AR ORV zone) as well as Portage and Geauga Counties which are part of the CA ORV zone. Cage traps were used to capture 91 unique raccoons over 817 trap nights: 2 raccoons were immobilized, processed and released, while 89 raccoons were euthanized and tested to enhance rabies surveillance.

### **Other Rabies Management Program Activities**

*Belmont County Enhanced Surveillance Trapping.*--On 10 November 2005, the first case of raccoon variant rabies was confirmed in Belmont County, Ohio (in a striped skunk) (Figure 1). This was also the first ever animal confirmed with raccoon rabies west of the Ohio River, which had presumably served as natural barrier to the spread of raccoon rabies. In response, WS began trapping within a 1.6 km (1 mi) radius of the skunk to help identify additional cases. Wildlife Services also worked with the state and local health departments to enhance rabies surveillance within the county. In 2006, WS continued with enhanced surveillance trapping within the same area. Trapping efforts initiated on 24 April 2006 yielded 20 raccoons over 286 trap nights. Nineteen raccoons were

ethanized and 1 was found dead, all were tested for rabies. No additional cases of rabies were identified in the area through increased surveillance efforts in 2006.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 55 opossums, 7 woodchucks, 2 Eastern cottontails (*Sylvilagus floridanus*), 2 red squirrels (*Tamiasciurus hudsonicus*), 1 Eastern chipmunk (*Tamias striatus*) and 1 Eastern fox squirrel (*Sciurus niger*).

Non-target animals that were captured and euthanized by WS in 2005 included: 5 opossums, and 2 woodchucks. These animals were all euthanized at the request of property owners or to enhance rabies surveillance.

### Rabies Laboratory Cooperation

As part of the ORV program in Ohio, WS cooperates with the ODH Laboratory (ODHL) and the CDC. There were 4,210 animals tested for rabies in Ohio in 2006 (Table 3), and 59 confirmed cases of rabies: 48 bats (*Chiroptera* spp.), 10 raccoons, and 1 opossum. All raccoons had raccoon rabies variant. The opossum could not be variant typed by the CDC. Total samples tested by the ODHL, the CDC, and WS decreased 37.8% in 2006, as compared to 2005 (n=6,767). Raccoons and skunks from northeast and eastern Ohio accounted for the largest testing decreases, 70.5% and 77.3% respectively. The decrease in samples was a result of eliminating nuisance samples being collected by local health departments at WS pickup sites. In 2007, cooperators anticipate an equal or greater number of animals will be tested for rabies given that raccoon rabies cases are still occurring within a 3 county area in northeastern Ohio.

Raccoons, skunks, foxes, and coyotes are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. This explains why 62.5% of enhanced surveillance samples were from raccoons, skunks, foxes, and coyotes. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 89.2% of public health surveillance samples are reported by WS as “other” (not raccoons, skunks, foxes, and coyotes).

*Ohio Department of Health Laboratory.*--The ODHL primarily tests animals for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure), although they have historically tested, and continue to test, hundreds of enhanced surveillance specimens annually (animals not involved in an exposure and usually submitted by WS). The ODH Zoonotic Disease Program (ZDP) relies on local health departments to collect and submit animals for rabies testing. Historically, most jurisdictions have been unable to implement enhanced surveillance due to a lack of resources (staff and funding), thereby depending on public health surveillance to monitor rabies in Ohio. Over the last 2 years, WS has provided refrigerators to local health departments (to store animals prior to submission/testing), while ODH has provided reimbursement contracts with 14 northeast and eastern counties. These contracts provide local health departments with \$60 per animal in compensation for collection, decapitation (usually by a veterinarian), shipment preparation, and staff costs related to animal submission. The ZDP also pays for shipping all animal heads to the ODHL for rabies testing (\$35,000 per year) and provides the Laboratory \$189,000 per year to support rabies testing. This assistance has led to an increase in rabies testing in Ohio since 2003 (when 3,223 samples were tested in the state). For more detailed information on rabies in Ohio over the last decade please visit: <http://www.odh.ohio.gov/odhPrograms/idc/zoodis/rabies/rab1.aspx>

Table 3. Animals tested for rabies via public health and enhanced surveillance systems from within ORV counties in Ohio and statewide, 2006 (rabies testing conducted by the Ohio Department of Health Laboratory, the Centers for Disease Control and Prevention, and Wildlife Services).

Species	Public health surveillance		Enhanced surveillance			Statewide total <sup>a</sup>
	Statewide	Within ORV counties	Statewide	Within ORV counties	Unknown surveillance type	
Raccoons	272	119 (43.8%)	788	686 (87.1%)	4	1,064
Skunks	26	15 (57.7%)	33	27 (81.8%)	1	60
Foxes	8	2 (25.0%)	9	6 (66.7%)	0	17
Coyotes	3	2 (66.7%)	1	1 (100.0%)	0	4
Other <sup>b</sup>	2,559	582 (22.7%)	498	129 (25.9%)	8	3,065
Total	2,868	720 (25.1%)	1,329	849 (63.9%)	13	4,210

<sup>a</sup> Includes unknown surveillance type.

<sup>b</sup> Other animals included: bats, cats, chipmunks, cows, deer, dogs, ferrets, goats, horses, llamas/alpacas, mice, mink, muskrats, opossums, rabbits, rats, rodents (unspecified), sheep, squirrels, weasels, woodchucks (groundhogs), wolf-dog hybrids, and wild animals (unspecified).

*Centers for Disease Control and Prevention.*--The CDC tests animal brainstems for rabies as part of enhanced surveillance. WS sends all positives, indeterminates and 10% of the negatives found using the dRIT to the CDC for confirmation and variant typing. The CDC also analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA).

In 2006, the CDC tested and confirmed 4 wildlife brainstem samples positive for raccoon variant rabies, found by Ohio WS using dRIT. The CDC also tested and confirmed 10% of the dRIT negatives (56) tested by Ohio WS in 2006. In addition, Ohio WS submitted 508 blood serum samples for rabies VNA analysis to the CDC in 2006. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## **ORV PROGRAM 2005 and 2006 – EVALUATION**

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### **Serology, Tetracycline Biomarker, and Age Results**

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, WS live-trapped 1,012 raccoons; blood and tooth samples were collected from most of these animals. Serum samples were sent to the CDC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA). Seventy raccoons were trapped in March and April 2005 from ORV naïve areas in Portage County (CA zone) to gather background serology on raccoons prior to CS distribution (Table 4). Baits were then distributed at 75 baits/km<sup>2</sup> and 4-8 weeks later, raccoons were trapped again from these areas to evaluate the efficacy of the bait distribution. The positive rabies antibody response declined from 7.1% to 5.0% following the April bait distribution (Table 4). On 1 June, WS began 2 density studies in Cuyahoga and Summit Counties (CA zone) to evaluate the antibody response in areas that were helicopter and hand baited using FMP baits at 75 baits/km<sup>2</sup>. The positive antibody response in the Summit study (helicopter baited) was 8.9% compared to that of the Cuyahoga study (hand baited) at 17.9% (Table 4). On 1 August, WS began 2 additional density studies in Cuyahoga County. These studies were conducted in similar habitat types and were also baited by helicopter. These studies combined produced a positive antibody response of 11.6% (Table 4). In mid-July, WS trapped 122 raccoons on the Plum Brook Research Station in Erie County. Rabies antibody response of those animals was compared to that of 2 ORV naïve density studies in Huron County, which were approximately 24 km (15 mi) from the research station. Rabies antibody response was 6.7% in Erie County compared to 1.1% in the Huron County studies (Table 4.) On 21 September, baits were distributed over the CA and AR ORV zone at 75 baits/km<sup>2</sup>. Four to 8 weeks post-ORV, raccoons trapped in the CA and Belmont County area of the AR ORV zones showed a 14.4% presence of rabies VNA (Table 4). In addition, tooth samples were collected from 887 raccoons that were collected during non-trapping enhanced surveillance activities in FMP helicopter/hand baited areas; 72 (8.1%) showed presence of tetracycline biomarker.

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during cooperative rabies management program activities in Ohio, 2005.

	CA <sup>a</sup> ORV naïve <sup>b</sup>	CA post- ORV	Summit <sup>c</sup> post- ORV	Cuyahoga 1 <sup>c</sup> post-ORV	Cuyahoga 2 & 3 <sup>c</sup> post-ORV	Erie (Plum Brook) <sup>d</sup>	Huron 1 & 2 <sup>c</sup> naïve <sup>b</sup>	CA & AR <sup>a</sup> post- ORV
Sample collection timeframe	21 Mar.- 22 Apr.	24 May- 24 Jun.	1-11 June	1-16 June	1-16 Aug.	12 Jul.- 5 Aug.	25 Jul.- 4 Aug.	17 Oct.- 17 Nov.
Weeks post-ORV	n/a	4-8	5-6	5-7	14-16	47-49	n/a	4-8
ORV bait type/ distribution method	n/a	CS/fixe- wing	FMP/helo	FMP/hand	FMP/helo	n/a	n/a	CS/fixe- wing
Unique raccoons	70	159	79	106	285	122	95	90
<b>Serology</b>								
Testable blood samples	70	159	79	106	284	120	95	90
Positive rabies antibody response (≥0.05 IU)	5 (7.1%)	8 (5.0%)	7 (8.9%)	19 (17.9%)	33 (11.6%)	8 (6.7%)	1 (1.1%)	13 (14.4%)
<b>Tetracycline biomarker</b>								
Testable tooth samples	62	139	79	101	270	106	90	62
Presence of tetracycline biomarker	0	1 (0.7%)	19 (24.1%)	27 (26.7%)	74 (27.4%)	15 (14.2%)	0	3 (3.8%)

<sup>a</sup> CA=Contingency Action; AR=Appalachian Ridge; ORV=oral rabies vaccination; CS=coated sachet; FMP=fishmeal polymer.

<sup>b</sup> Samples were collected in an ORV naïve area (never before treated with ORV).

<sup>c</sup> Density study conducted.

<sup>d</sup> Experimental ORV baiting was conducted on Plum Brook Station by an Ohio State University graduate student in 2003 and 2004; no operational ORV has been conducted there.

In 2006, WS live-trapped 528 raccoons; blood and tooth samples were collected from most of these animals. Serum samples were sent to the CDC, while tooth samples were sent to Matson's Lab. Tooth results were not tested for the presence of tetracycline because most animals captured were collected in areas where CS baits were distributed (CS baits do not contain a biomarker). From 19-20 April, baits were distributed over the CA zone at 75 baits/km<sup>2</sup> (Figure 1). Four to 8 weeks post-ORV, raccoons trapped from this zone demonstrated a 14.5% positive rabies antibody response (Table 5). In July/August, WS conducted 1 density study each in Geauga, Lake, and Cuyahoga Counties to evaluate the antibody response in areas that were baited by different distribution methods and bait type at 75 baits/km<sup>2</sup> (Figure 1). The Geauga and Lake County studies had comparable rabies VNA responses (67.2% and 66.7%, respectively), while the Cuyahoga study was 25.0%. This study was a replicate of a 2005 study and antibody response was similar to last year (27.4%). On 21 August, trapping began in an ORV naïve area of Belmont County to gather background serology data prior to the distribution of CS baits in that area. Four to 5 weeks after bait distribution, raccoons were trapped again from the area and the positive antibody response increased from 8.3% to 21.6%. During that 5 September baiting, the CA ORV zone was again baited at 75 baits/km<sup>2</sup> (as part of the larger AR zone). The CA zone was baited twice to achieve above the targeted bait density. Four to 8 weeks post-ORV, raccoons were sampled and showed a 42.6% presence of rabies VNA.

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during cooperative rabies management program activities in Ohio, 2006.

	CA <sup>a</sup> post-ORV spring	Gauga <sup>b</sup> post-ORV	Lake <sup>b</sup> post-ORV	Cuyahoga <sup>b</sup> post-ORV	Belmont ORV naïve <sup>c</sup>	Belmont post-ORV	CA post-ORV fall
Sample collection timeframe	24 May- 24 Jun.	17 Jul.- 27 Aug.	17-27 July	31 Jul.- 15 Aug.	21-25 Aug.	2-13 Oct.	16 Oct.- 3 Nov.
Weeks post-ORV	4-8	12-13	12-13	14-15	n/a	4-5	4-8
ORV bait type/ distribution method	CS/fixe- wing	CS/fixe- wing	FMP/hand	FMP/helo	n/a	CS/fixe- wing	CS/fixe- wing
Unique raccoons	154	69	82	108	24	37	54
<b>Serology</b>							
Testable blood samples	124	64	66	108	24	37	54
Positive rabies antibody response (≥0.05 IU)	18 (14.5%)	43 (67.2%)	44 (66.7%)	27 (25.0%)	2 (8.3%)	8 (21.6%)	23 (42.6%)

<sup>a</sup> CA=Contingency Action; AR=Appalachian Ridge; ORV=oral rabies vaccination; CS=coated sachet; FMP=fishmeal polymer.

<sup>b</sup> Density study conducted.

<sup>c</sup> Samples were collected in an ORV naïve area (never before treated with ORV).

*Age Results.*--In 2005 and 2006, 1,771 and 341 raccoon teeth, respectively, were aged using premolars of live-captured, found dead, or euthanized animals collected during trapping and enhanced surveillance activities (Figure 2). Age results are typical, with the populations dominated by young of the year juveniles (<1 y.o.) and 1 y.o. animals. Six raccoons in 2005 were aged at >10 y.o.: 3 at 11 y.o., 1 at 12 y.o., 1 at 13 y.o., and 1 at 14 y.o., while 3 raccoons in 2006 were aged at >10 y.o.: 1 at 11 y.o.; 1 at 12 y.o.; and 1 at 13 y.o.

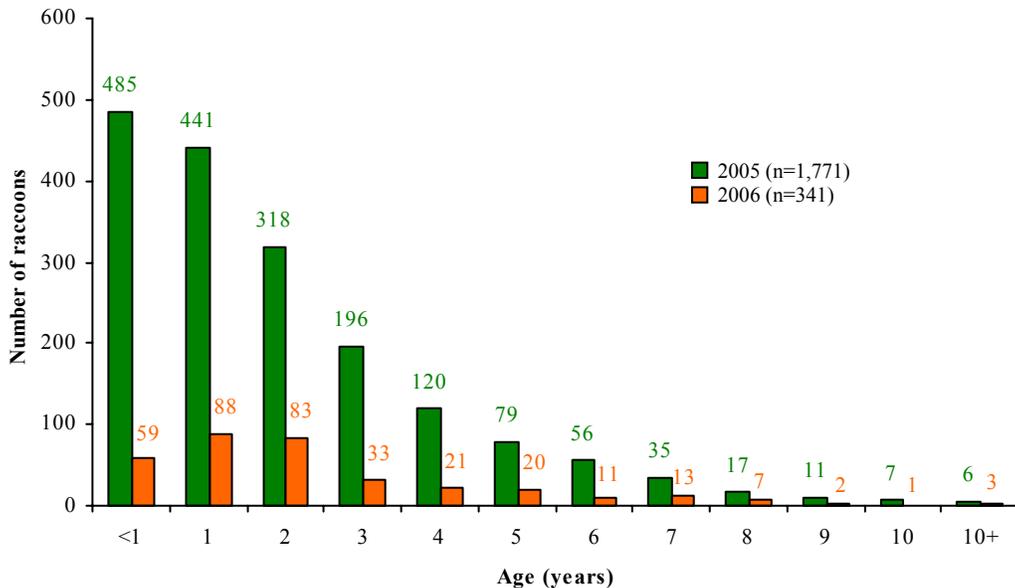


Figure 2. Age class distribution of 1,771 and 341 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in Ohio, 2005 and 2006.

## SUMMARY

In 2006, WS completed its tenth year of cooperative participation in the Ohio ORV program. Over 1.1 million baits were distributed over the AR and CA ORV zones. The focus in 2006 was on enhancing rabies surveillance, particularly in the CA zone, and continuing the use of dRIT to reduce the burden on public health laboratories and improve turnaround time of test results during a rabies outbreak. Wildlife Services cooperated to collect and submit 609 animals for rabies testing, and tested all of those animals using the dRIT. Three raccoon density studies were conducted in (2 in urban/suburban environments near Cleveland and 1 in an agricultural environment indicative of eastern Ohio). Trapping was conducted in an ORV naïve area up to 4 weeks prior to bait distribution and then trapped again post-ORV to determine the effectiveness of CS baits. Post-ORV trapping activities focused on counties within the CA zone and yielded 467 raccoons (including density study captures). An additional 20 raccoons were trapped in Belmont County during enhanced surveillance activities in continued response to the first case of raccoon variant rabies in the county (confirmed in 2005).

In 2007, WS will continue to: enhance rabies surveillance in the AR and CA zones focusing on Belmont and nearby counties and areas near major urban population centers (Cleveland and other Cuyahoga County cities); participate in ORV bait distribution; execute a trap-vaccinate-release program in the CA zone; and conduct post-ORV monitoring and evaluation in both zones. The Ohio ORV zones continue to be an integral part of a larger Appalachian effort that in 2006 included Maryland, North Carolina, Ohio, Pennsylvania, Tennessee, Virginia, and West Virginia. Future ORV baiting strategies in Ohio will continue to be tied to national planning efforts to prevent the westward spread of raccoon rabies and explore strategies to eliminate this variant from the U.S.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM PENNSYLVANIA 2006

### BACKGROUND

Raccoon (*Procyon lotor*) rabies was first reported in Pennsylvania in 1982. The first documented cases occurred in Bedford, Fulton, and Franklin Counties. Twelve years later raccoon rabies had become enzootic throughout the Commonwealth's 67 counties. Since 1995, >350 animals have been confirmed positive for rabies annually. The first oral rabies vaccine (ORV) baits were distributed in Pennsylvania during the fall of 2001; 138,602 baits were hand distributed across 1,875 km<sup>2</sup> within 2 counties in the northwest corner of the state. This baiting effort was tied to the Appalachian Ridge (AR) ORV zone, with the goal of strengthening the existing ORV zone in eastern Ohio and expanding it eastward to reduce the area where raccoon rabies occurs. In 2002 and 2003, Pennsylvania expanded its baiting program to cover 25,189 km<sup>2</sup> in 18 western counties bordering Maryland, Ohio, and West Virginia. The program became an integral part of national efforts to create an ORV zone from Lake Erie to the Gulf of Mexico to prevent the westward spread of raccoon rabies. In 2004, Wildlife Services (WS) distributed baits across a similar (although slightly smaller) area of western Pennsylvania and also in previously ORV naïve areas of Cambria, Indiana, Somerset, and Westmoreland Counties (as a spring bait efficacy study coupled with a raccoon density study). The following year, the spring bait study (SBS) area was treated again, a live rabies challenge study was conducted (within the SBS area), and Pennsylvania's portion of the AR zone was baited. In 2006, the SBS was not baited, but the same part of the AR zone was. Density studies were again conducted in Allegheny County and enhanced rabies surveillance increased significantly. The Pennsylvania Department of Agriculture (PDA) provided the state leadership for the baiting effort, while WS provided wildlife management leadership and contributed significant funding. This cooperative initiative should create a vaccinated area of sufficient scope and allow for the exploration of methods to eliminate raccoon rabies from Pennsylvania.

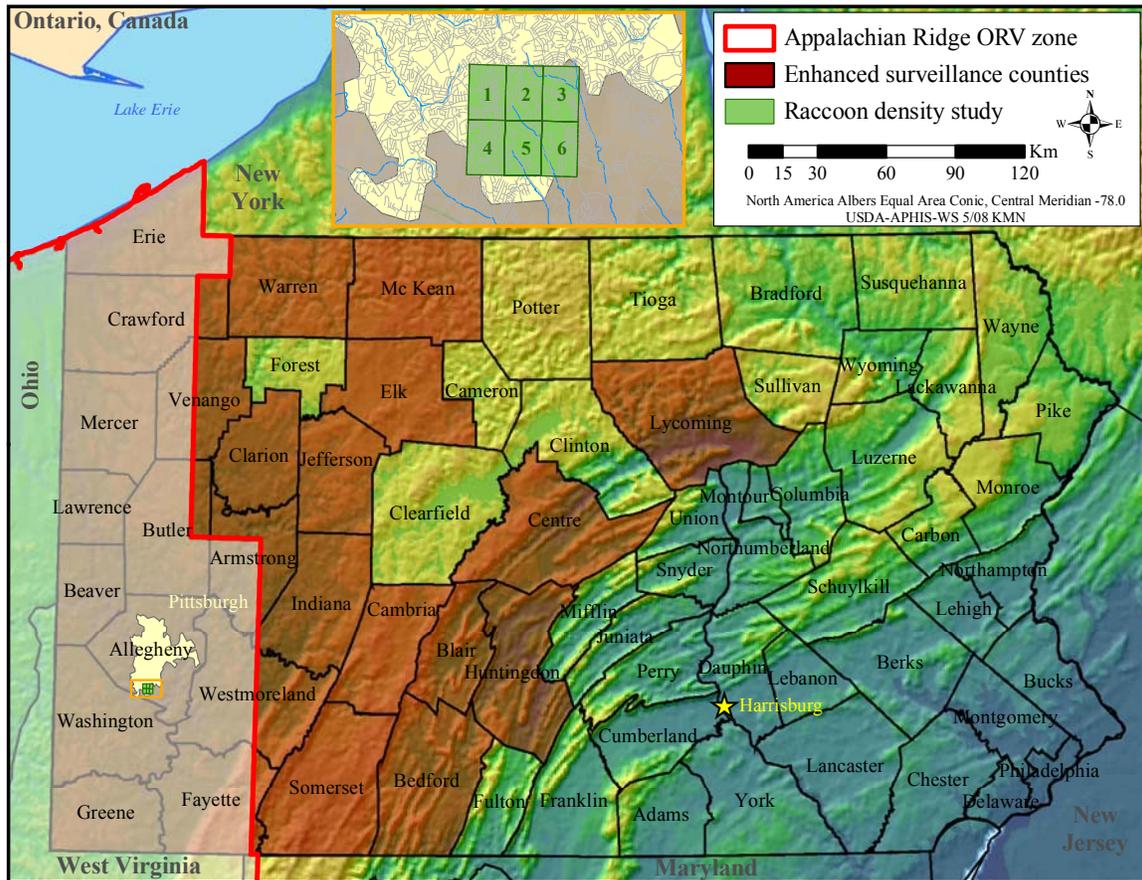


Figure 1. Cooperative rabies management program activities in Pennsylvania, 2006.

## ORV PROGRAM 2006

### Bait Distribution

For the sixth consecutive year in 2006, WS participated in bait distribution efforts throughout western Pennsylvania; 1,359,812 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 15 counties in the western part of the state (Figure 1). The AR ORV zone encompassed 20,580 km<sup>2</sup> (7,946 mi<sup>2</sup>). From 8-26 August, 387,142 fishmeal polymer (FMP) baits were distributed from the ground in areas too populated to bait by air. Fixed-wing aircraft were used to distribute 606,769 fishmeal-coated sachet (CS) and 365,901 FMP baits from 5-21 September.

During ground (hand) bait operations, assistance was provided to WS by: the PDA, the Pennsylvania Department of Health (PDH), the Erie County Department of Health, and the Allegheny County Health Department (ACHD). During aerial baiting, aircraft and pilots were provided by the Ontario Ministry of Natural Resources, while WS personnel served as navigators and flight crew in the planes. Ground support for the flights was offered by: the Ohio Department of Health, the Ohio National Guard, the Pennsylvania Game Commission, the PDA, the PDH, and WS employees from Pennsylvania, Maryland, Ohio, and West Virginia.

Since its program inception in 2001, WS has distributed 7,937,300 ORV baits in Pennsylvania.

### Enhanced Surveillance

In 2006, WS conducted enhanced surveillance of raccoon rabies by collecting, submitting, and testing suspect rabid animals from counties in or near the Pennsylvania ORV zone. Wildlife Services collected animals by the following methods: 1) humanely euthanizing (according to the American Veterinary Medical Association's Panel on Euthanasia recommendations) raccoons that had puncture wounds/bite marks, exhibited disorientation, or showed signs of illness during trapping activities; 2) conducting road kill surveys and collecting mammals in suitable testing condition; 3) obtaining wildlife reported by Pennsylvania residents as displaying rabies-like symptoms; and 4) obtaining wildlife from nuisance wildlife control officers. Wildlife Services also continued to work with state and local health departments to increase the number of enhanced surveillance samples for rabies testing.

As a result of enhanced surveillance efforts in western and central Pennsylvania, WS cooperated to collect and submit 2,712 animals for rabies testing (Table 1). Of those samples, 76 animals (2.8%) tested positive for the raccoon variant of the rabies virus.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Since 2005, Pennsylvania WS personnel have been conducting dRIT for all non-human and non-domestic exposure animals they collected. In 2006, WS tested 2,442 enhanced surveillance samples (90.0%) using the dRIT, with 47 positives. All positives and 10% of all negatives were sent to the CDC for confirmation and strain typing. The CDC (using the dFA test) had 100% agreement with the WS dRIT results for positive and negative samples and typed all positives (non bats) as raccoon rabies variant and all bat positives as bat variant. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Pennsylvania.

Table 1. Animals submitted for rabies testing by Wildlife Services (rabies positives in parentheses) in or adjacent to ORV counties in Pennsylvania, 2006.

County	Raccoon	Skunk	Coyote	Gray fox	Red fox	Bobcat	Other <sup>a</sup>	Total
Allegheny	715 (6)	124	5	1	1		8	854 (6)
Armstrong	23 (2)	1			1		2	27 (2)
Beaver	59	5	1	2	2		1	70
Bedford <sup>b</sup>	1							1
Blair <sup>b</sup>	1							1
Butler	45 (3)	2					6	53 (3)
Cambria <sup>b</sup>	36 (5)	12					5	53 (5)
Centre <sup>b</sup>			1					1
Clarion <sup>b</sup>	8							8
Crawford	55	4		1	4		3	67
Elk <sup>b</sup>	1							1
Erie	270 (4)	215			3		229	717 (4)
Fayette	53 (1)	5		3	1		3	65 (1)
Greene	34 (1)						4	38 (1)
Huntingdon <sup>b</sup>	3	3						6
Indiana	44 (7)	22 (1)		2	1		4 (1) <sup>c</sup>	73 (9)
Jefferson <sup>b</sup>	3						2	5
Lawrence	76 (2)						4	80 (2)
Lycoming <sup>b</sup>					2			2
McKean <sup>b</sup>	2							2
Mercer	53	5					5 (1) <sup>c</sup>	63
Somerset <sup>b</sup>	34 (10)	6 (2)			1 (1)		1	41 (13)
Venango	13	1					4(2) <sup>c</sup>	18 (2)
Warren	6 (1)				1		6 (1) <sup>c</sup>	13 (2)
Washington	71	2	5	1	1		12	92
Westmoreland	261 (23)	31		8 (2)	6		55	361 (25)
Total	1,867 (65)	438 (3)	12	18 (2)	24 (1)	0	354 (5)	2712 (76)

<sup>a</sup> Other animals included: bat, black bear, cat, chipmunk, dog, fisher, mink, opossum, rabbit, squirrel, weasel, white-tailed deer, and woodchuck.

<sup>b</sup> ORV not applied in this county.

<sup>c</sup> Rabid fisher in Indiana County; rabid bats in Mercer, Venango, and Warren Counties.

## Population Monitoring

In 2006, WS conducted 6 raccoon density studies in Allegheny County using the National Rabies Management Program standard protocol of 50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights (Figure 1). The studies were conducted in wooded metro parks (with playground areas, picnic pavilions, bike trails, and ball fields) on the periphery of Pittsburgh. All 6 study areas had been treated with ORV (hand baited area) since 2002. Blood and tooth samples were collected from most of the 606 unique raccoons captured during the 6 studies (Table 2). These raccoons were also hand vaccinated prior to release to bolster the immunity of the population. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines.

Table 2. Index to raccoon densities in Allegheny County, Pennsylvania, 2006.

	SP06-1	SP06-2	SP06-3	SP06-4	SP06-5	SP06-6
Time of study	11-21 Jul.	10-20 Jul.	10-20 Jul.	11-21 Jul.	10-20 Jul.	11-21 Jul.
Weeks post-ORV	38-40	38-40	38-40	38-40	38-40	38-40
Macrohabitat	Urban/suburban	Urban/suburban	Urban/suburban	Urban/suburban	Urban/suburban	Urban/suburban
Target trap nights	500	500	500	500	500	500
Unique raccoons	61	98	123	32	135	157
Recaptured raccoons	26	46	29	4	19	46
Area (km <sup>2</sup> )	3.00	3.00	3.00	3.00	3.00	3.00
Raccoon density index <sup>a</sup>	20.3	32.7	41.0	10.7	45.0	52.3

<sup>a</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

### Post-ORV Monitoring

Post-ORV sampling for Pennsylvania was initiated on 12 April and continued until 10 November 2006 in areas that were baited in previous years or approximately 5 weeks prior. Cage traps and various types of lures were used to capture 470 unique raccoons from Allegheny, Armstrong, Beaver, Butler, Crawford, Erie, Fayette, Greene, Lawrence, Mercer, Somerset, Washington and Westmoreland Counties. Most raccoons (403) were immobilized, processed and released. The remaining 67 animals were either euthanized to enhance rabies surveillance or taken as part of other WS control activities in the ORV zone.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 202 opossums (*Didelphis virginiana*), 88 woodchucks (*Marmota monax*), 38 domestic/feral cats (*Felis catus*), 24 Eastern cottontails (*Sylvilagus floridanus*), 15 striped skunks (*Mephitis mephitis*), 5 muskrat (*Ondatra zibethicus*), 5 Norway rats (*Rattus norvegicus*), 4 red squirrels (*Tamiasciurus hudsonicus*), 2 porcupines (*Erethizon dorsatum*), 1 chipmunk (*Tamias striatus*), 1 fox squirrel (*Sciurus niger*), 1 gray squirrel (*Sciurus carolinensis*), 1 mink (*Mustela vison*), 1 American robin (*Turdus migratorius*), 1 common grackle (*Quiscalus quiscula*), and 1 English house sparrow (*Passer domesticus*).

### Rabies Laboratory Cooperation

Wildlife Services' ORV program in Pennsylvania cooperates with the PDH Bureau of Laboratories (PDHBL), the PDA Veterinary Laboratory (PDAVL), the ACHD, and the CDC. Wildlife Services has had an efficient and cooperative relationship with all 4 laboratories since 2001, and they remain critical to the surveillance and monitoring phases of the ORV program in Pennsylvania.

*Pennsylvania Department of Health Bureau of Laboratories.*--The PDHBL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed human exposure, usually submitted by Wildlife Conservation Officers and the public). The PDHBL tested 3,805 animals for the rabies virus in 2006, representing a 6.5% increase from the number of samples tested in 2005 (Table 3). Animals were submitted from all 67 counties throughout the state, including the ORV counties of: Allegheny, Armstrong, Beaver, Butler, Crawford, Erie, Fayette, Greene, Lawrence, Mercer, Venango, Washington, and Westmoreland. Of the animals tested statewide by the PDHBL, 14.4% came from a county treated with ORV.

Raccoons, skunks, and foxes are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 91.4% of the animals tested for rabies by the PDHBL in 2006 are reported by WS as "other." For more general information on rabies from the PDH please visit: <http://www.dsf.health.state.pa.us/health/cwp/view.asp?A=171&Q=230513>

Table 3. Animals tested for rabies by the Pennsylvania Department of Health Bureau of Laboratories via the public health surveillance system in Pennsylvania, 2006.

	Statewide	Within ORV counties
Raccoons	247	109 (44.1%)
Skunks	44	10 (22.7%)
Foxes	35	5 (14.3%)
Other <sup>a</sup>	3,479	502 (14.4%)
Total	3,805	626 (16.5%)

<sup>a</sup> Other animals included: bats, cats, dogs, woodchucks, and other unspecified animals.

*Pennsylvania Department of Agriculture Veterinary Laboratory.*--The PDAVL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed domestic animal exposure, usually submitted by Wildlife Conservation Officers, veterinarians, and the public). The PDAVL tested 1,462 animals for the rabies virus in 2006, representing a 22% decrease from the number of samples tested in 2005 (Table 4). Animals were submitted from all 67 counties throughout the state, including 13 ORV counties. Of the animals tested statewide by the PDAVL, 14.0% came from a county treated with ORV. For more general information on rabies from the PDA please visit:

<http://www.agriculture.state.pa.us/agriculture/cwp/view.asp?q=127956>

Table 4. Animals tested for rabies by the Pennsylvania Department of Agriculture Veterinary Laboratory via the public health surveillance systems in Pennsylvania, 2006.

	Statewide	Within ORV counties
Raccoons	404	98 (24.3%)
Skunks	106	8 (7.5%)
Foxes	75	6 (8.0%)
Other <sup>a</sup>	877	92 (10.5%)
Total	1,462	204 (14.0%)

<sup>a</sup> Other animals included: bats, cats, dogs, woodchucks, and other unspecified animals.

*Allegheny County Health Department.*--The ACHD tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed human or domestic animal exposure) within Allegheny County (and sometimes nearby counties). The ACHD tested 451 animals for the rabies virus in 2006: 61 raccoons, 3 skunks, 3 foxes, and 384 other animals (bats [*Chiroptera* spp.], domestic/feral cats, domestic/feral dogs, woodchucks, and other unspecified animals). This was a 9% increase from the number of samples tested in 2005. Animals were submitted to the ACHD from 9 counties in western Pennsylvania: Allegheny, Armstrong, Beaver, Butler, Indiana, Lawrence, Mercer, Washington, and Westmoreland. For more general information on rabies from the ACHD please visit: <http://www.achd.net/factsheet/rabies.htm>

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies VNA. Pennsylvania WS submitted 619 blood serum samples for rabies VNA analysis to the CDC in 2006. This is approximately the same number of samples that was submitted in 2005. The Pennsylvania ORV program anticipates similar numbers of serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology, and prevention and control) on a national level please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## ORV PROGRAM 2005 – EVALUATION

At the time of this report, the current year's (2006) evaluation data were not available.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits

do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, WS live-trapped 773 unique raccoons; 769 were captured within the ORV zone. Blood and tooth samples were collected from most of these animals and serum samples were sent to the CDC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA). Of the 373 samples collected during the optimal evaluation period of 4-12 weeks post-ORV, most (315) were collected during evaluation of the SBS. Only 8 (2.5%) samples had rabies antibodies, contributing to the low overall serology of 3.7% (Table 5).

Table 5. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Pennsylvania, 2005.

	Spring bait study (pre-2005 ORV) <sup>a</sup>	Spring bait study (post-2005 ORV)	AR <sup>b</sup> zone density studies (pre-2005 ORV) <sup>a</sup>	Fall AR <sup>b</sup> (post-2005 ORV)
Sample collection timeframe	12-26 April	1 Jun.-15 Jul.	20 Jul.-12 Aug.	12 Oct.-10 Nov.
Weeks post-ORV	50-52	5-12	48-52	4-10
ORV bait type	FMP	FMP/CS	FMP	FMP
Unique raccoons	41	349	299	80
<b>Serology</b>				
Testable blood samples	41	315	263	58
Positive rabies antibody response (≥0.05 IU)	0	8 (2.5%)	12 (4.6%)	5 (8.6%)
<b>Tetracycline biomarker</b>				
Testable tooth samples	41	160 <sup>c</sup>	202	46
Presence of tetracycline biomarker	4 (9.8%)	54 (33.8%)	48 (23.8%)	13 (28.3%)

<sup>a</sup> Samples collected prior to 2005 ORV bait distribution, but from areas that had been baited in 2004.

<sup>b</sup> AR=Appalachian Ridge; FMP=fishmeal polymer.

<sup>c</sup> To minimize stress, teeth were not taken from animals being processed and held for the live rabies challenge study which accounts for the small percentage of testable tooth samples.

**Age Results.**--In 2005, 907 raccoon teeth were aged using premolars of live-captured animals and canines of animals found dead or euthanized (Figure 2). Age results are typical, with the populations dominated by raccoons ≤1 y.o. Three raccoons were aged at >10 y.o.: 2 at 11 y.o. and 1 at 12 y.o.

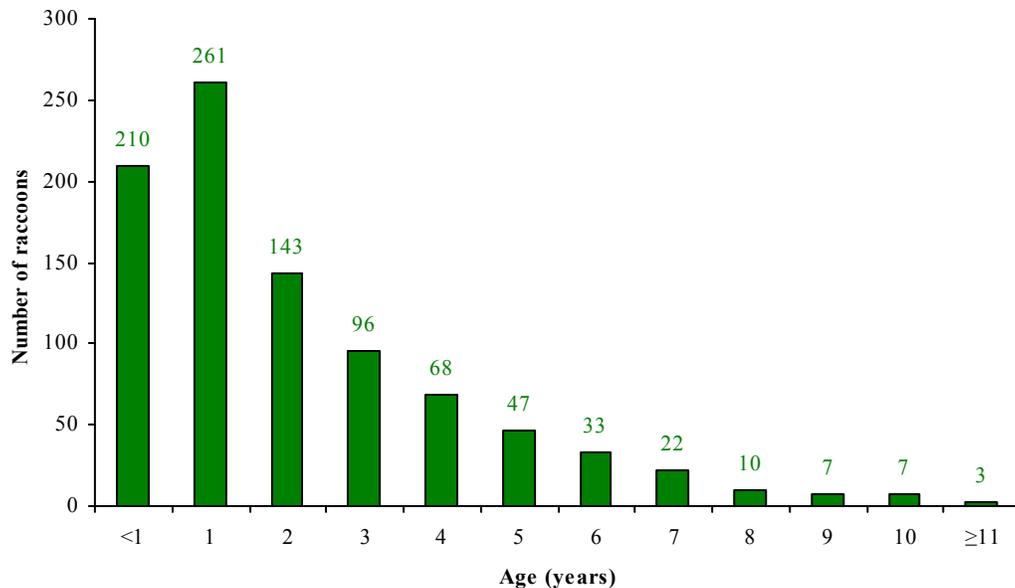


Figure 2. Age class distribution of 907 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in Pennsylvania, 2005.

## **SUMMARY**

During 2006, WS completed its sixth year of cooperative participation in the Pennsylvania cooperative rabies management program. Activities included distributing 1,359,812 ORV baits by boat, hand, and from aircraft, across 15 counties encompassing 20,580 km<sup>2</sup>. Six contiguous raccoon density studies were conducted in urban/suburban areas on the edge of Pittsburgh (Allegheny County) with densities ranging from 10.7 to 52.3 raccoons/km<sup>2</sup>. Efforts in 2006 also focused on enhanced rabies surveillance, with 2,712 samples collected from 26 counties (up from 719 animals in 2005). Nearly 8 million baits have been distributed in Pennsylvania since the ORV program began in 2001. Pennsylvania's baiting effort is part of a larger AR ORV zone, which in 2006 included Maryland, North Carolina, Ohio, Pennsylvania, Tennessee, Virginia, and West Virginia.

In 2007, WS will continue to work cooperatively to distribute ORV baits in the western part of the state. Even greater emphasis will be placed on enhanced rabies surveillance and urban/suburban raccoon density studies (i.e., Pittsburgh) will be replicated. The Pennsylvania ORV program is integral to national planning efforts to contain raccoon rabies and explore strategies to eliminate this unique variant of the rabies virus.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM TENNESSEE 2006

### BACKGROUND

In 2002, an oral rabies vaccination (ORV) program was initiated in Tennessee as part of Wildlife Services (WS) National Rabies Management Program (NRMP) to stop the westward spread of the raccoon (*Procyon lotor*) variant of rabies. Raccoon rabies had not been found in Tennessee at that point, but was reported across the border in nearby North Carolina. In an effort to stay ahead of the disease front, WS extended the Appalachian Ridge (AR) ORV zone (which began at Lake Erie) into northeastern Tennessee. In June 2003, the rabies front, which had stalled in North Carolina, crossed into northeastern Tennessee and 4 cases of raccoon rabies were confirmed in Carter County, while 1 case was confirmed in Johnson County. There were no cases found in this area during 2004 despite increased surveillance. In 2005, 6 cases were confirmed in wildlife in the area, including positives in Washington and Unicoi Counties where raccoon rabies had not been previously documented. Four cases were confirmed in northeastern Tennessee in 2006 (1 in Johnson and 3 in Unicoi Counties).

In November 2003, WS established the Georgia-Alabama-Tennessee (GAT) ORV zone where the Georgia and Alabama borders meet southern Tennessee. At the time, raccoon rabies was in northwestern Georgia and moving westward. The Alabama-Coosa River system to the south and the Appalachian Mountains to the north were serving as potential natural barriers to the westward spread of raccoon rabies. The GAT zone was established to help fill a gap between these potential barriers and to prevent the spread of raccoon rabies into the Tennessee Valley and subsequently the interior of the United States. In January 2004, raccoon rabies entered southeastern Tennessee from Georgia and reached the GAT ORV zone. In response to the first positive case of raccoon rabies inside the GAT zone, WS began baiting the city of Chattanooga and surrounding areas of Hamilton County in the spring, while baiting these areas again in the fall as part of the larger GAT ORV effort. During 2004, 14 cases of raccoon rabies were documented in wildlife in Hamilton County. During 2005, only 1 animal (a raccoon) was confirmed with raccoon rabies in Hamilton County and the virus was not detected in any surrounding counties. Although no cases of raccoon rabies were documented in Hamilton County in 2006, 1 case was confirmed in adjacent Bradley County in a gray fox (*Urocyon cinereoargenteus*).

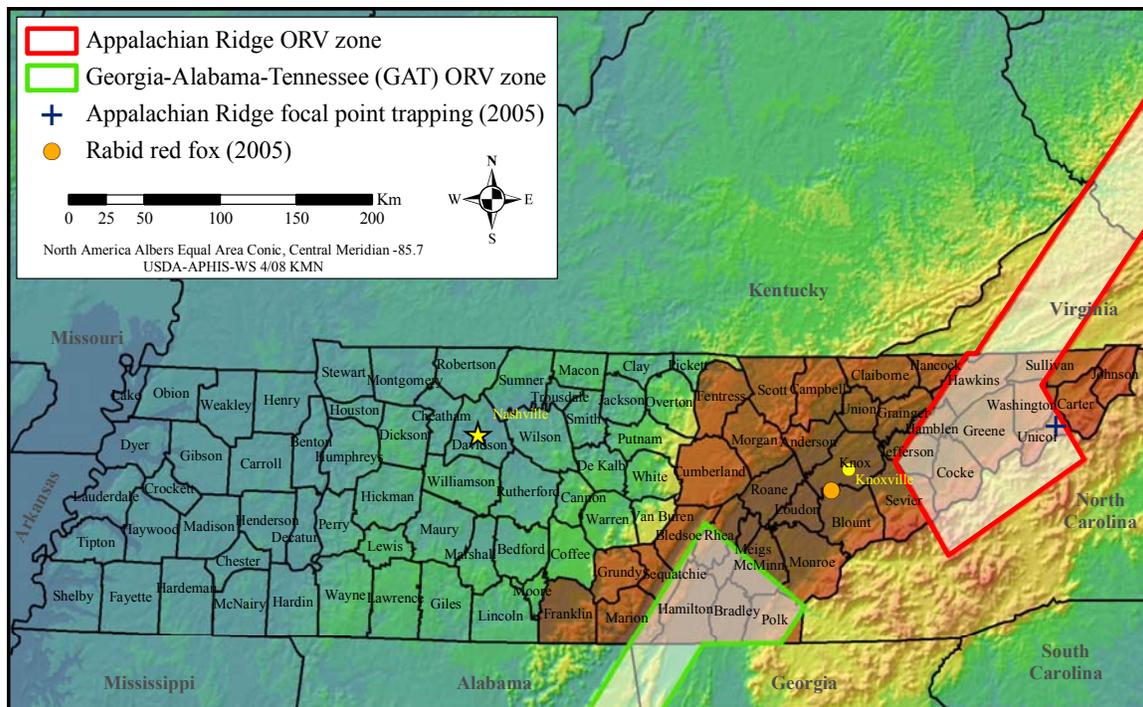


Figure 1. Wildlife Services cooperative rabies management program activities in Tennessee, 2006.

Despite the first positive case of raccoon variant rabies emerging in Knox County in 2005 (Figure 1), there were no additional cases in the area in 2006. However, in response to the 2005 case, the enhanced surveillance area

was expanded to include Anderson, Campbell, Claiborne, Cumberland, Morgan, Roane, and Union Counties in 2006, and enhanced surveillance efforts were greatly increased in Knox County.

In Tennessee, the AR and GAT ORV programs are being conducted by WS, in cooperation with the Tennessee Department of Health (TDH), the Tennessee Department of Agriculture, the Tennessee Wildlife Resources Agency (TWRA), the Chattanooga/Hamilton County Department of Health, and the Centers for Disease Control and Prevention (CDC). Numerous individual landowners and many federal, state, and local agencies provided WS access to private and government owned properties for ORV program trapping and monitoring. Many other agencies and numerous animal control departments, nuisance wildlife control operators, and wildlife rehabilitators assisted WS by collecting suspicious acting and road killed animals for rabies testing in the surveillance zone.

## **ORV PROGRAM 2006**

### **Bait Distribution**

For the fifth consecutive year, WS participated in bait distribution efforts in northeastern Tennessee (AR ORV zone) and for the fourth year in southeastern Tennessee (GAT ORV zone); 770,618 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 12,257 km<sup>2</sup> (4,733 mi<sup>2</sup>) (Figure 1). Since its program inception in 2002, WS has distributed 2,249,376 ORV baits in Tennessee. Aircraft and pilots for both Tennessee ORV programs in 2006 were provided by Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA), while WS provided ground support and served as navigators and flight crew in the planes.

*Appalachian Ridge.*--In 2006, the Tennessee portion of the AR ORV zone was expanded to include 7,018 km<sup>2</sup> (2,710 mi<sup>2</sup>) in 12 counties. From 4-8 August, 46,440 fishmeal polymer (FMP) baits were distributed by hand, while 340,300 fishmeal-coated sachet baits (CS) were distributed by fixed-wing aircraft from 7-16 August.

*Georgia-Alabama-Tennessee.*--In October 2006, the Tennessee portion of the GAT ORV zone was expanded to include 5,239 km<sup>2</sup> (2,023 mi<sup>2</sup>) in 10 counties. From 14-16 October, 150,465 FMP baits were distributed by hand, while 233,413 CS baits were distributed by fixed-wing aircraft from 16-23 October.

### **Enhanced Surveillance**

In 2006, WS continued enhanced surveillance for the raccoon variant of the rabies virus in Tennessee. Wildlife Services personnel continued to recruit state and local agencies and work cooperatively with local nuisance control operators to collect suspect animals for testing. An emphasis was placed on raccoons, striped skunks (*Mephitis mephitis*), gray foxes, red foxes (*Vulpes vulpes*), and coyotes (*Canis latrans*) that exhibited strange behavior, were found dead in unusual places, or were fresh road killed animals. The enhanced surveillance area included 37 counties, creating a surveillance corridor along the borders with Virginia, North Carolina, Georgia and Alabama (Figure 1). During these efforts, 1,573 animals were collected throughout the surveillance zone, including 650 animals from Knox County (the site of the rabid red fox in 2005). All 1,573 animals (1,149 raccoons, 317 striped skunks, 46 gray foxes, 29 red foxes, 27 coyotes, 2 opossums [*Didelphis virginiana*], 2 woodchucks [*Marmota monax*], and 1 bobcat [*Lynx rufus*]) were tested by WS using the direct rapid immunohistochemistry test (dRIT); they all tested negative for rabies. Ten percent of the negatives were sent to the CDC for confirmation. Using the direct fluorescent antibody (dFA) test, the CDC had 100% agreement with the WS dRIT results. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Tennessee.

### **Post-ORV Monitoring**

*Appalachian Ridge.*--Post-ORV sampling for Tennessee's AR ORV zone was initiated on 11 September 2006. Cage traps were used to capture 133 unique raccoons from Greene, Hamblen, Hawkins, Sullivan and Unicoi Counties; all raccoons were immobilized, processed and released.

*Georgia-Alabama-Tennessee.*--Post-ORV sampling for Tennessee's GAT ORV zone was initiated on 13 November 2006. Cage traps were used to capture 134 unique raccoons from Hamilton County; all raccoons were immobilized, processed and released.

## Non-target Captures

Non-target animals captured and released by WS in 2006 included: 125 opossums, 11 feral cats (*Felis catus*), 6 striped skunks, 2 Eastern cottontails (*Sylvilagus floridanus*), 2 snapping turtles (*Chelydra serpentina*), 1 fox squirrel (*Sciurus niger*), and 1 gray squirrel (*Sciurus carolinensis*). In addition, 1 fox squirrel was captured and died in the trap.

## Rabies Laboratory Cooperation

The ORV program in Tennessee cooperates with the TDH laboratories in Jackson, Nashville, and Knoxville and the CDC.

*Tennessee Department of Health Laboratories.*--Each of the TDH labs is responsible for testing animal brainstems for rabies within their designated region. Overall, the TDH confirmed 131 cases of rabies in Tennessee in 2006. Positive samples are sent to the Nashville Branch Laboratory for rabies variant typing. The Knoxville Branch Laboratory (KBL) is responsible for the East Tennessee region which includes 26 of the 37 designated enhanced surveillance counties. The KBL tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure) and enhanced surveillance (specimens not involved in an exposure) at the request of WS.

The KBL tested 968 brainstem samples for the rabies virus in 2006 (Table 1). This represents a 6.6% decrease from the number of samples tested in 2005. The 2006 samples were submitted from 40 counties in the East Tennessee region and from 31 counties within the designated enhanced rabies surveillance zone.

Raccoons, skunks, foxes, and coyotes are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 83.0% of the animals tested for rabies in the East Tennessee region in 2006 are reported by WS as “other.” For general information on rabies in Tennessee please visit: <http://www.state.tn.us/health/>

Table 1. Animal tested for rabies by the Tennessee Department of Health, Knoxville Branch Laboratory in the East Tennessee region, 2006.

	East Tennessee region	Within Wildlife Services' enhanced rabies surveillance zone
Raccoons	120	118 (98.3%)
Skunks	31	27 (87.1%)
Foxes	13	13 (100%)
Coyotes	1	1 (100%)
Other <sup>a</sup>	803	758 (94.4%)
Total	968	917 (94.7%)

<sup>a</sup> Other animals included: bats, bear, cats, cattle, chipmunks, deer, dog, gerbils, goats, guinea pigs, hamsters, horses, llamas, mice, moles, opossums, rabbits, rats, sheep, squirrels, and woodchucks (groundhogs).

The KBL currently handles the disposal of medical waste created during WS ORV surveillance, monitoring and evaluation projects. During 2006 the KBL also provided positive and negative rabies controls for the implementation of the dRIT by WS.

The TDH is working to create a real-time online disease tracking system for the entire state. This system would allow laboratory personnel to input test results immediately for a variety of diseases. It would allow health professionals to quickly identify disease trends and help in identifying possible breaches in the ORV zones should they occur. In addition, the KBL began rabies variant typing in 2006, further increasing the abilities of the TDH and WS to respond quickly to potential breaches in the ORV zones.

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA). Tennessee WS submitted 276 blood serum samples for rabies VNA analysis to the CDC in 2006. This represents a 44.1% decrease from the 494 samples submitted by WS in 2005. The decrease in sample numbers is attributed to a lack of density studies and focal point trapping events in 2006. In addition, there were spring and fall trapping events in the GAT zone in 2005, but only 1 post-bait trapping event for this region in 2006. The Tennessee ORV program anticipates an increase in number of

serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake. Fishmeal polymer baits contain a chemical biomarker (tetracycline) that stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker. Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005 and 2006, during the evaluation phases of the Tennessee cooperative rabies management program, WS live-trapped 494 and 276 unique raccoons, respectively, within the ORV bait zones (Tables 2 and 3). Blood and tooth samples were collected from most of these animals and serum samples were sent to the CDC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA).

Table 2. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Tennessee, 2005.

	Chattanooga evaluation	AR <sup>a</sup> focal point evaluation	AR evaluation	GAT <sup>a</sup> evaluation	Incidental take evaluation	Total
Sample collection timeframe	23 May-8 Jun.	4-5 Aug.	6-28 Sep.	28 Nov.-15 Dec.	Year-long	2005
Weeks post-ORV	4-6	n/a <sup>b</sup>	3-6	4-7	n/a <sup>b</sup>	
Bait type/distribution method	FMP <sup>a</sup> /hand	n/a	CS <sup>a</sup> /fixed-wing & FMP/hand	FMP/fixed-wing & hand	n/a	
Unique raccoons	138	6	160	190	8	494
<b>Serology</b>						
Testable blood samples	138	6	160	190	0	494
Positive rabies antibody response (≥0.05 IU)	38 (27.5%)	0	18 (11.3%)	55 (28.9%)		111 (22.5%)
<b>Tetracycline</b>						
Testable tooth samples	122	6	129	161	8	426
Presence of tetracycline biomarker	66 (54.1%)	0	42 (32.6%)	92 (57.1%)	0	200 (46.9%)

<sup>a</sup> AR=Appalachian Ridge; GAT=Georgia-Alabama-Tennessee; FMP=fishmeal polymer; CS=coated sachet.

<sup>b</sup> Samples were collected in an ORV naïve (never before treated with ORV) area.

*Age Results.*--In 2005 and 2006, 426 and 230 raccoon teeth, respectively, were aged using premolars of live-captured animals (Figure 2). These samples were collected from raccoons in the AR and GAT ORV zones (including the Chattanooga ORV zone in 2005).

Table 3. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during the cooperative rabies management program in Tennessee, 2006.

	AR <sup>a</sup> evaluation	GAT <sup>a</sup> evaluation	Incidental take evaluation	Total
Sample collection timeframe	11 Sep.-12 Oct	28 Nov.-15 Dec.	Year-long	2006
Weeks post-ORV	4-8	5-7	n/a <sup>b</sup>	
Bait type/distribution method	CS <sup>3</sup> /fixed-wing & FMP <sup>3</sup> /hand	FMP/fixed-wing & hand	n/a	
Unique raccoons	133	134	9	276
<b>Serology</b>				
Testable blood samples	133	134	9	276
Positive rabies antibody response (≥0.05 IU)	32 (24.1%)	62 (46.3%)	1 (11.1%)	95 (34.4%)
<b>Tetracycline</b>				
Testable tooth samples	112	118	0	230
Presence of tetracycline biomarker	38 (33.9%)	64 (54.2%)		102 (44.3%)

<sup>a</sup> AR=Appalachian Ridge; GAT=Georgia-Alabama-Tennessee; CS=coated sachet; FMP=fishmeal polymer.

<sup>b</sup> Samples were collected in ORV naïve (never before treated with ORV) areas.

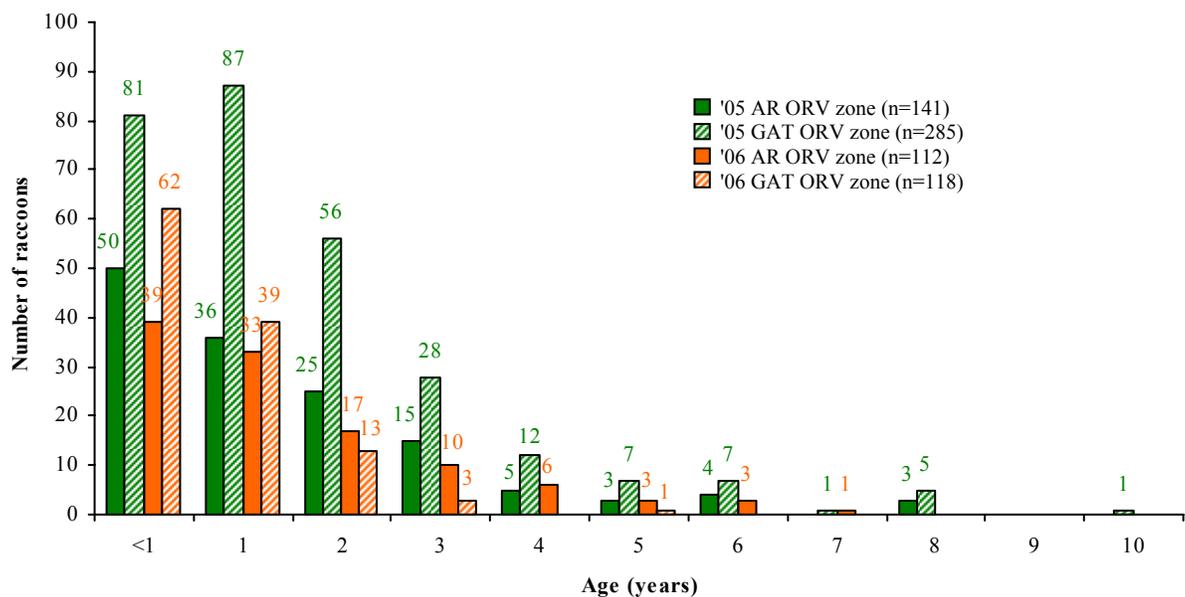


Figure 2. Age class distribution of 426 and 230 raccoon tooth samples collected by Wildlife Services during the evaluation phases of the cooperative rabies management program in Tennessee, 2005 and 2006 (AR=Appalachian Ridge; GAT=Georgia-Alabama-Tennessee).

## SUMMARY

In 2006, Tennessee WS conducted its fifth and fourth year of rabies management activities in the AR and GAT ORV zones, respectively. During bait distribution efforts, 770,618 ORV baits were distributed in eastern Tennessee. Trapping activities yielded 276 raccoons sampled for rabies VNA testing. Enhanced surveillance efforts resulted in the collection of 1,573 animals; all tested negative for rabies by WS using the dRIT.

During 2007, WS will continue to conduct intensive enhanced surveillance in Knox and surrounding counties and throughout the remainder of the 37-county surveillance zone to determine the prevalence of raccoon rabies. Should additional positives be confirmed, population reduction and/or trap-vaccinate-release efforts will likely be used in conjunction with large scale ORV baiting to contain this potential outbreak. Efforts will be made to recruit additional local law enforcement agencies, TWRA personnel, and the general public to report and/or collect suspect rabid animals. Additional density study areas and monitoring sites will be secured to look at potential differences in raccoon population trends among different habitats, elevations, and ORV zones.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM TEXAS 2006

### BACKGROUND

Two canine rabies epizootics emerged in Texas in 1988: 1 involving coyotes (*Canis latrans*) and dogs (*C. familiaris*) in south Texas and the other involving gray foxes (*Urocyon cinereoargenteus*) in west-central Texas. The south Texas epizootic has resulted in 2 human deaths and required over 3,000 people to receive post exposure rabies treatment. In 1994, the public health threat created by these 2 expanding epizootics prompted the Governor of Texas to declare rabies a state health emergency. By 1996, the 2 epizootics expanded to involve 69 Texas counties. In February 1995, an oral rabies vaccination program (ORVP) was initiated as a multiyear effort with a goal of creating zones of vaccinated coyotes and gray foxes (January 1996) along the leading edges of the epizootics, thereby halting the spread of the virus. The ORVP is a cooperative program involving: Wildlife Services (WS); the Texas Department of State Health Services (TDSHS) formerly the Texas Department of Health; the Texas Cooperative Extension-Wildlife Services; the Texas National Guard; the Centers for Disease Control and Prevention (CDC); the Department of Defense Veterinary Food Analysis and Diagnostic Laboratory (DOD-VFADL) at Ft. Sam Houston; and other local, state, and federal agencies.

### ORV PROGRAM 2006

#### Bait Distribution

For the twelfth consecutive year, WS participated in bait distribution efforts in south Texas (coyote ORV zone) and for the eleventh year in west-central Texas (gray fox ORV zone); 2,807,763 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 91,313 km<sup>2</sup> (35,257 mi<sup>2</sup>) (Figure 1). Since its program inception in 1995, WS has cooperated to distribute over 27 million ORV baits in Texas.

*Coyote.*--From 5-10 January 2006, WS participated as a member of the ORVP by helping to distribute 772,000 oral rabies vaccine (ORV) baits over 33,123 km<sup>2</sup> (12,789 mi<sup>2</sup>) of south Texas (Figure 1). This 64.4 km wide maintenance barrier is in place to prevent the re-emergence of canine rabies variant from Mexico. Fixed-wing and rotary aircraft distributed 759,760 fishmeal polymer (FMP) baits across portions of 14 counties, while city employees hand distributed 12,240 FMP baits throughout communities in Cameron, Hidalgo, and Webb Counties. Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA), provided the fixed-wing aircraft services and the Texas WS program provided the helicopter. Fishmeal polymer baits contain 150 mg of tetracycline biomarker and a 2 ml sachet of Raboral V-RG® vaccine.

*Gray fox.*--From 12-21 January 2006, WS participated in the aerial distribution of 2,035,763 ORV baits over 58,190 km<sup>2</sup> (22,468 mi<sup>2</sup>) to contain a variant of rabies unique to gray foxes in west-central Texas (Figure 1). Gray fox baits are composed of a dog food polymer (DFP) containing 150 mg of tetracycline biomarker and a 2 ml sachet of Raboral V-RG® vaccine. With the gray fox baits supplied by WS during 2006, the Texas ORVP cooperative effort was able to continue the encirclement of the gray fox rabies epizootic and apply ORV baits within Big Bend National Park and adjacent areas.

#### Enhanced Surveillance

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

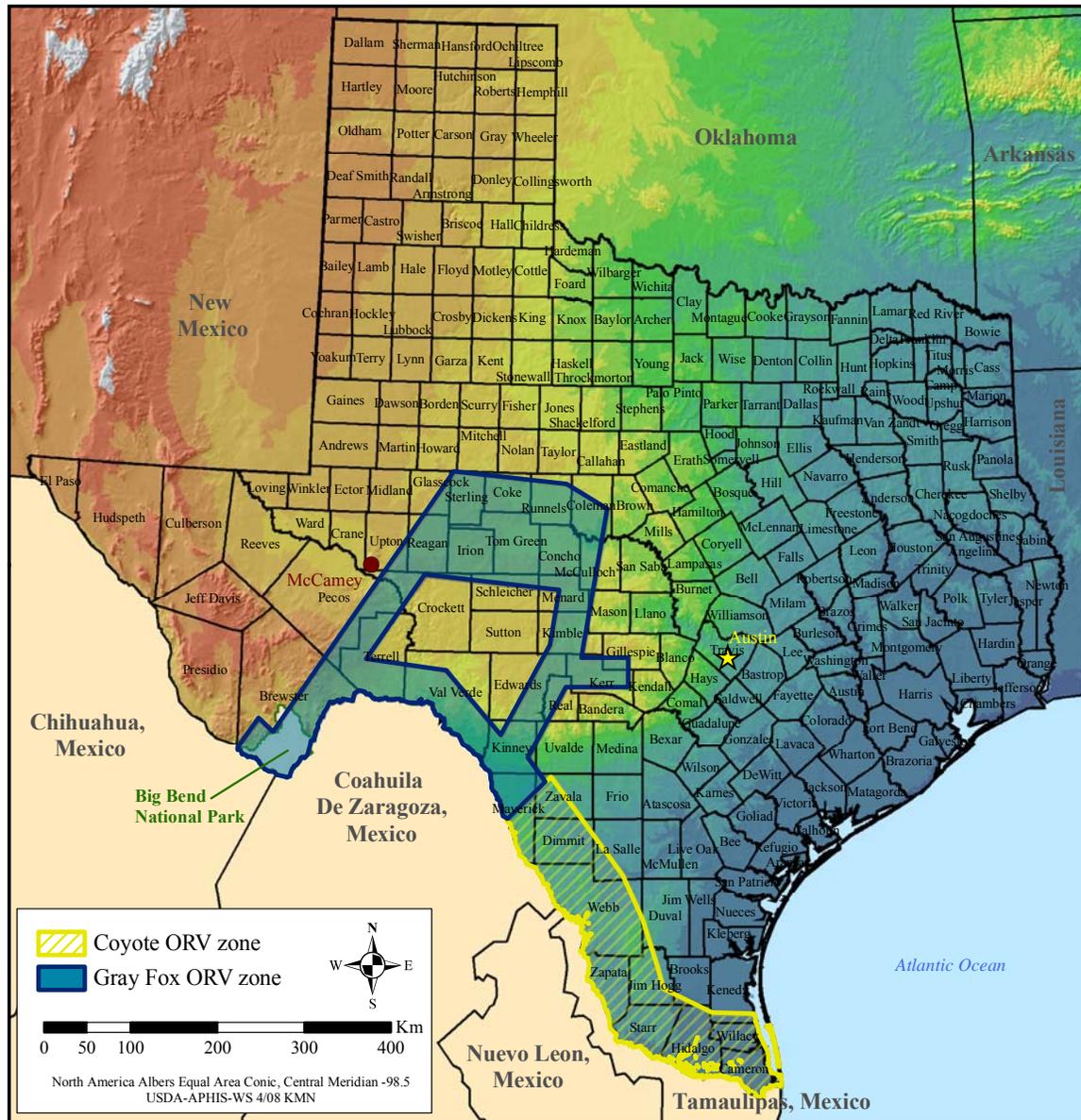


Figure 1. Wildlife Services cooperative rabies management program activities in Texas, 2006.

Texas WS personnel attended dRIT training in October 2005 at the CDC in Atlanta, Georgia and TDSHS personnel attended dRIT training in March 2006 in Laredo, Texas. During 2006, WS and TDSHS personnel collected 210 animals and tested them for rabies using the dRIT; 3 tested positive and 207 tested negative (Table 1). All positives and 10% of all negatives were sent to the CDC for confirmation and strain typing. The CDC (using the dFA test) had 100% agreement with the WS and TDSHS dRIT results. Wildlife Services and TDSHS will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Texas.

Table 1. Animals tested for rabies by Wildlife Services and the Texas Department of States Health Services using the direct rapid immunohistochemistry test (dRIT) in or adjacent to oral rabies vaccination (ORV) counties in Texas, 2006 (rabies positives in parentheses).

County	Raccoon	Coyote	Gray fox	Bobcat	Total
Cameron		2			2
Coke	8		2	3	13
Duval		1			1
Edwards			1(1)	1(1)	2(2)
Jim Hogg		21			21
McCulloch	1		1(1)		2(1)
Runnels	3	6		6	15
Starr		7			7
Sterling	17		7	14	38
Webb		40			40
Willacy		1			1
Zapata		68			68
Total	29	146	11(2)	24(1)	210(3)

### Laboratory Cooperation

The ORV program in Texas cooperates with the TDSHS Laboratory (TDSHSL), the DOD-VFADL, and Johnston Biotech (JB) (Sania, Ontario, Canada). The Texas ORVP has had an efficient and cooperative relationship with these laboratories since 1995, and they remain critical to the program's evaluation and surveillance efforts.

*Texas Department of State Health Services.*--The TDSHSL tests full cross sections of the animal's brainstems plus at least one other section to include cerebellum or hippocampus (preferably all three) for rabies through public health surveillance (specimens involved in a potential or confirmed exposure) and enhanced surveillance testing (samples not involved in an exposure). The TDSHSL tested 14,251 brainstem samples for the rabies virus in the course of routine public health surveillance in 2006 (Table 2). Of these, 888 tested positive for rabies. Samples were submitted from 240 of 254 counties throughout the state. No samples were submitted as part of enhanced rabies surveillance efforts. In addition, a 16-year-old male from the Houston area, succumbed to rabies from a variant associated with the Mexican free-tailed bat (*Tadarida brasiliensis*). For detailed information on rabies in Texas, please visit: <http://www.dshs.state.tx.us/idcu/disease/rabies/>

Table 2. Animal brainstem samples testing positive for the rabies virus by the Texas Department of State Health Services Laboratory via the public health surveillance system in Texas, 2006<sup>a</sup>.

Species	Unvaccinated interior of the gray fox epizootic <sup>c</sup>	Within gray fox ORV zone	Within coyote ORV zone	Remainder of State
Coyotes	0	0	0	1 (0.1%)
Foxes	13 (1.5%)	9 (1.3%)	0	9 (1.1%)
Raccoons	1 (0.1%)	4 (0.5%)	0	14 (1.5%)
Skunks	2 (0.2%)	18 (2.0%)	0	331 (35.9%)
Bobcats	2 (0.2%)	6 (0.8%)	0	2 (0.2%)
Bats	1 (0.1%)	1 (0.1%)	8 (0.9%)	421 (48.3%)
Other <sup>d</sup>	3 (0.4%)	4 (0.5%)	0	38 (4.2%)
Total	22	42	8	816

<sup>a</sup> Includes all rabies virus variants.

<sup>b</sup> ORV=oral rabies vaccination.

<sup>c</sup> Samples submitted from counties interior to the circular gray fox ORV zone where ORV is currently not used.

<sup>d</sup> Other includes domestic animals and livestock.

*Department of Defense Veterinary Food Analysis and Diagnostic Laboratory.*--The DOD-VFADL analyzes wildlife serum samples (submitted by the TDSHS and WS) for levels of rabies virus neutralizing antibodies (VNA). The DOD-VFADL analyzed 339 serum samples for rabies VNA in 2006, representing a 14.5% increase from the 296 samples submitted in 2005. Both the TDSHS and WS anticipate an increase in the number of submissions in 2007.

*Johnston Biotech.*--Johnston Biotech analyzes coyote and gray fox teeth submitted for age and the presence of tetracycline biomarker. In 2006, JB analyzed 339 tooth samples, representing a 14.5% increase in the number of samples submitted from 2005. The number of submissions in 2007 is expected to increase.

## ORV PROGRAM 2006 – EVALUATION

Annual evaluations of the ORVP are based on 3 criteria: 1) the detection of the tetracycline biomarker in dental tissue which can be used to demonstrate the number of baits eaten in a given year and the annual bait consumption by animals over a period of multiple years. However, inaccuracies happen because older animals do not deposit the biomarker and the presence of tetracycline does not confirm that the vaccine sachet has been punctured or consumed; 2) the number of positive rabies antibody responses from animals collected within the vaccination zone (at  $\geq 0.05$  IU); and 3) the epidemiology of rabies cases in the target area. All biological samples collected to evaluate the success of the ORVP were obtained from targeted animals. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association’s Panel on Euthanasia recommendations. The 2006 Texas ORV bait distribution occurred in January and 2006 program evaluation data (serology, tetracycline, and age results) were available at the time of this report.

### Serology, Tetracycline Biomarker, and Age Results

*Coyote.*--In 2006, TDSHS and WS collected 145 coyotes, for ORVP evaluation, from within the south Texas vaccination zone. Forty-nine of 145 coyotes tested (33.8%) within the ORV zone showed a positive rabies antibody response to the vaccine, while 113 of 145 coyotes tested (77.9%) were positive for the tetracycline biomarker included in the bait material (Figure 2). Since the initiation of the ORVP in south Texas, canine rabies cases have declined from 122, reported during the pre-program year in 1994, to 0 in 2000. During 2001 and again in 2004, a single canine rabies case involving a stray dog was confirmed in Laredo, Texas. Both isolated cases occurred within 1 mile of the U.S.-Mexico border. In response, the City of Laredo Health Department’s Animal Control Division implemented an aggressive isolation and vaccination protocol. Additional control measures included increased vaccination clinics for domestic pets throughout the city. No additional cases have been reported since.

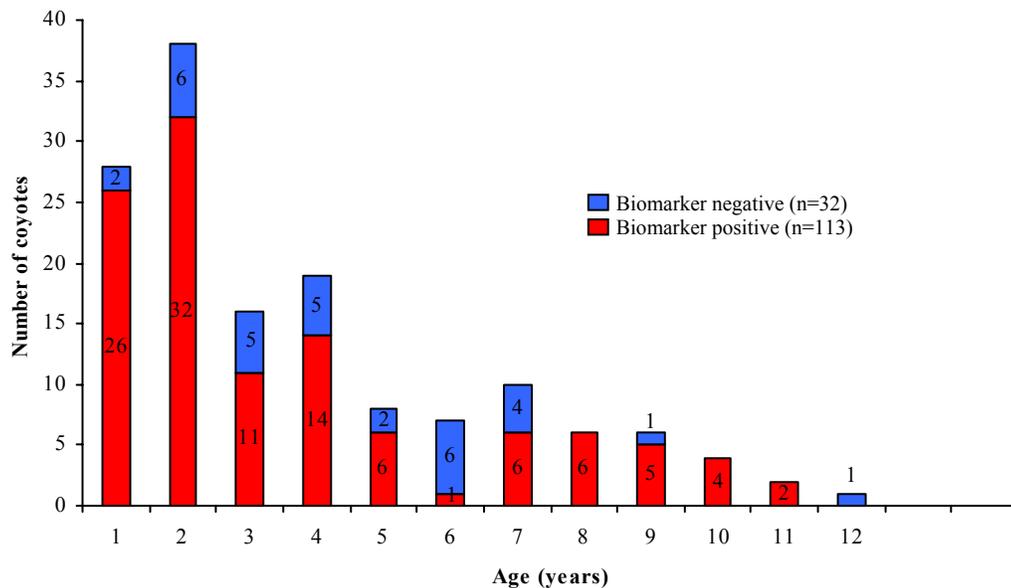


Figure 2. Age class distribution and presence/absence of tetracycline biomarker for 145 coyotes collected during post-bait oral rabies vaccination program evaluation in the coyote zone of south Texas, 2006.

*Gray fox.*--During the evaluation phase of the 2006 gray fox ORVP in west-central Texas, WS and the TDSHS collected 146 gray foxes. Blood and tooth samples were taken from all gray foxes as well as 19 coyotes, 8 bobcats (*Lynx rufus*), 12 striped skunk (*Mephitis mephitis*), and 9 raccoons (*Procyon lotor*). Overall, 88 of 194 animals (45.4%) demonstrated a positive rabies VNA response, while 78 of 194 animals (40.2%) tested showed the presence of tetracycline biomarker (Table 3). In addition, the 146 gray foxes were analyzed for age and the population was dominated by 1 and 2 year olds (Figure 3).

Table 3. Serology and tetracycline biomarker results of biological samples collected during post-bait oral rabies vaccination program evaluation in the gray fox zone of west-central Texas, 2006.

Species	Positive rabies VNA <sup>a</sup> response	Presence of tetracycline biomarker	Total
Gray fox	74 (50.7%)	74 (50.7%)	146
Coyote	4 (21.1%)	3 (5.3%)	19
Striped skunk	8 (66.7%)	2 (16.7)	12
Raccoon	1 (11.1%)	1 (11.1%)	9
Bobcat	1 (12.5%)	0	8

<sup>a</sup> VNA=virus neutralizing antibody ( $\geq 0.05$  IU).

The gray fox ORVP in west-central Texas continues to show success with a decline in the number of confirmed cases involving the gray fox variant of rabies. During the pre-program year of 1995, 244 cases were reported. In 2006, the number of cases increased to 45 from the previous low of 8 cases reported during 2005. All but 11 cases occurred within or inside the 32 km wide immune barrier. Late in the year a foci of gray fox variant cases developed near McCamey, Texas, 19 km (12 mi) outside of the vaccination barrier, and included a small portion of Upton and Pecos Counties. Passive surveillance of this area and adjacent counties was employed to monitor the area until ORV baits could be applied in January 2007.

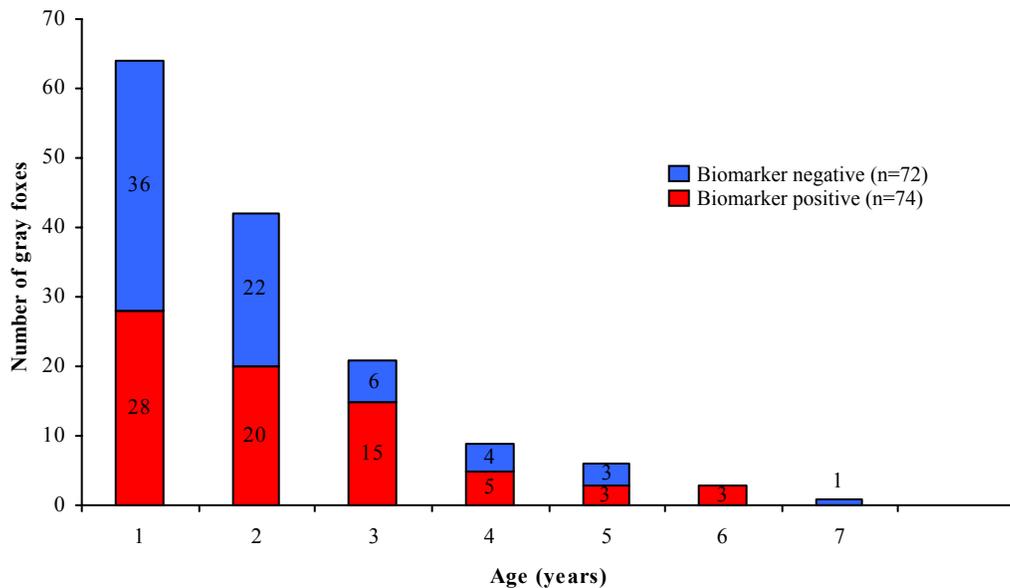


Figure 3. Age class distribution and presence/absence of tetracycline biomarker for 146 gray foxes collected during post-bait oral rabies vaccination program evaluation in the gray fox zone of west-central Texas, 2006.

## SUMMARY

Since 1995, 11.58 million ORV baits have been distributed over south Texas by the coyote ORVP. This has proved to be highly effective in the elimination of the canine variant of rabies in that area. A maintenance strategy has been developed to sustain a zone of immunized wildlife along the Texas-Mexico border with only 2 incursions into the zone at Laredo since 2001, thus preventing the re-emergence of the variant. With continued

support for the cooperative ORVP effort, similar success is sought in the gray fox epizootic in west-central Texas where 16.59 million ORV baits have been distributed since 1996.

## **WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM VERMONT 2006**

### **BACKGROUND**

In 1994, the raccoon (*Procyon lotor*) variant of the rabies virus was first confirmed in several Vermont towns on the Massachusetts border. In less than 2 years, raccoon rabies had spread north through 8 counties in Vermont and by May 1996 was just south of Burlington, Vermont's largest city, approximately 70 km (42 miles) south of the U.S.-Canada Border (Figure 1). One year later, on 5 May 1997 an intensive oral rabies vaccination (ORV) program was initiated to prevent further northward spread of raccoon rabies up the Lake Champlain valley into Quebec, Canada. The initial ORV zone encompassed 1,637 km<sup>2</sup> (632 mi<sup>2</sup>) in northwestern Vermont including all of Franklin County and portions of Chittenden and Grand Isle Counties. This area was baited the following year, as well as 556 km<sup>2</sup> near Lyndonville in response to a rabid skunk (*Mephitis mephitis*) confirmed in the town to the south. In 1999, 2 rabid raccoons were confirmed just south of Newport and the ORV zone was doubled in size to include the Lake Memphremagog basin (south to Lyndonville). In 2001, the ORV zone was expanded again along the Connecticut River Valley in Essex County in response to 2 rabid raccoons: 1 on each side of the river (state border with New Hampshire). The Vermont ORV zone has continued to expand and encompassed approximately 35% of the state in 2006.

Since its inception, the Vermont ORV program has been a cooperative effort between Wildlife Services (WS), Cornell University (CU), the Vermont Departments of Health (VDH) and Fish and Wildlife (VTFW), the Vermont Agency of Agriculture, Food, and Markets (VTAG), the Ontario Ministry of Natural Resources (OMNR), and the Province of Quebec. Wildlife Services has been the major source of federal funds for program implementation. Wildlife Services has also provided federal wildlife management leadership by continuing to play an active role in: program planning and coordination; organizing ground support for aerial bait distribution; working in and navigating aircraft to distribute baits; coordinating the hand distribution of baits in areas too populated to bait by air; enhancing rabies surveillance by collecting suspect rabid animals; and evaluating program efficacy by monitoring post-ORV rabies virus neutralizing antibody (VNA) levels and bait uptake (when appropriate) in raccoons.

### **ORV PROGRAM 2006**

#### **Bait Distribution**

For the tenth consecutive year, WS participated in bait distribution efforts in northern Vermont; 383,179 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 8,704 km<sup>2</sup> (3,361 mi<sup>2</sup>) in 2006 (Figure 1). Two new low bait density zones were added, one each in Addison and Washington Counties (each 250 km<sup>2</sup> in size). The efficacy of this low density baiting (35 baits/km<sup>2</sup>) will be compared to the standard target bait density (70 baits/km<sup>2</sup>) for the Vermont ORV zone.

From 22-23 August, 374,008 fishmeal-coated sachet (CS) baits were distributed over 9 counties: Addison, Caledonia, Chittenden, Essex, Franklin, Grand Isle, Lamoille, Orleans, and Washington. Fixed-wing aircraft were provided by the OMNR, while WS personnel served as navigators and flight crew in the planes. Ground support for aerial baiting was provided by WS, the OMNR, and CU. From 22 August-12 September, WS distributed 9,171 fishmeal polymer (FMP) baits via ground operations (hand/truck baiting) in 24 small cities and villages. Since its program inception in 1997, WS has cooperated to distribute 2,556,281 ORV baits in Vermont.

#### **Enhanced Surveillance**

In June 2006, the Province of Quebec confirmed their first-ever case of raccoon variant rabies, approximately 11 km (6.6 mi) north of the Vermont border. Prior to that, the northernmost case of raccoon variant had been confirmed in January 2001 in a skunk in Coventry, Vermont (Lake Memphremagog basin) about 16 km (9.6 mi) south of the US-Canada Border. In response to this new case in Quebec, Vermont WS increased their enhanced rabies surveillance in the northern part of the state by improving existing and establishing new relations with the Vermont Agency of Transportation, VTFW Game Wardens, State Police, US Border Patrol, Animal Control Officers, Town Health Officers, local town officials, and farmers and landowners. This surveillance network focused on road killed, strange acting, and nuisance animals not involved in an exposure that may otherwise have not been tested through the public health surveillance system. By year's end, WS had collected 139 animals

(104 raccoons, 29 skunks, 5 red foxes, 1 coyote) from 8 counties within the ORV zones; 29 tested positive for rabies (21 raccoons and 8 skunks).

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the dFA test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Vermont WS personnel attended dRIT training in May 2005 at the Centers for Disease Control and Prevention in Atlanta, Georgia. Currently, the VDH is meeting enhanced surveillance testing needs; however, WS plans on implementing the dRIT in the future.

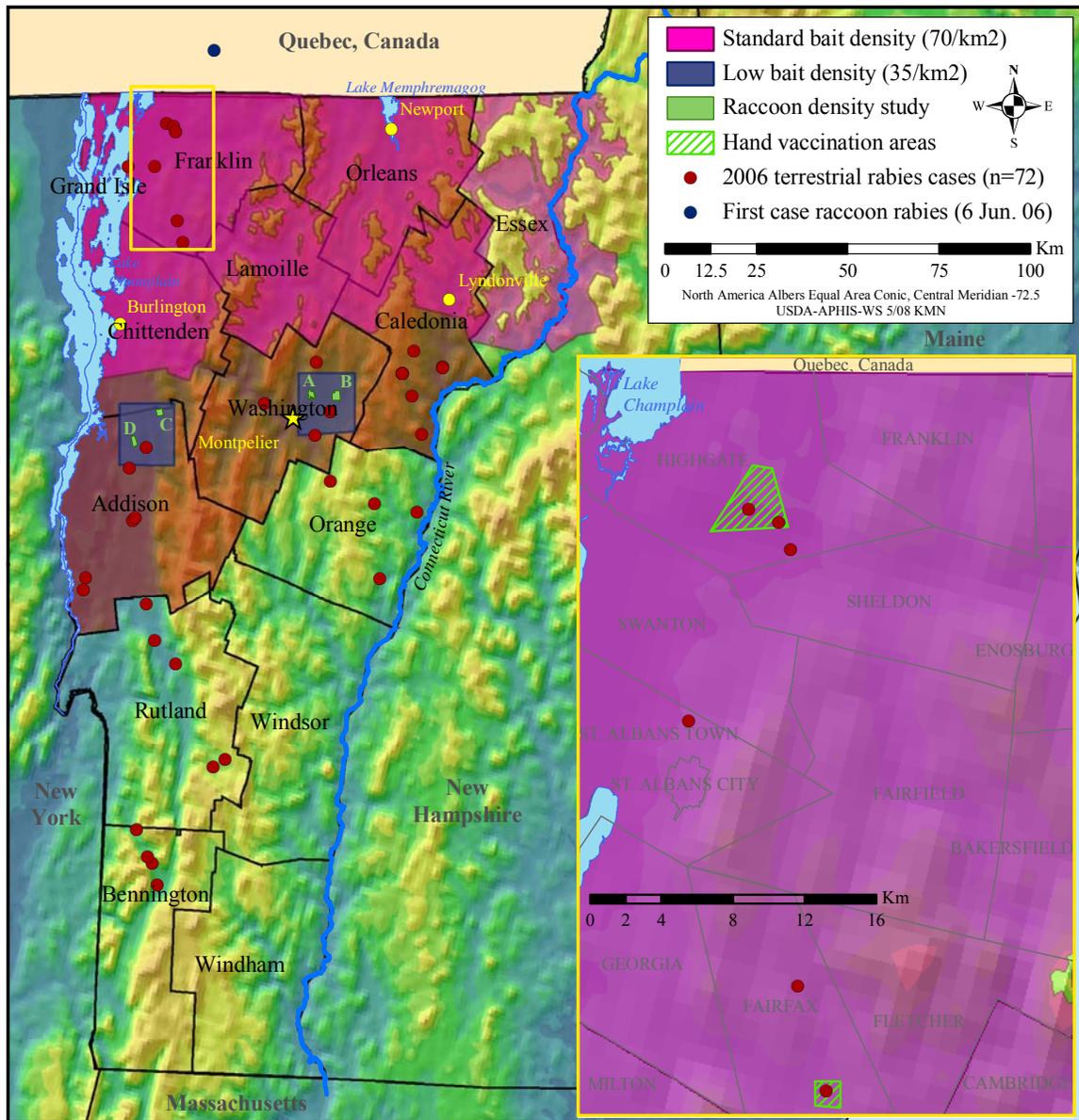


Figure 1. Wildlife Services cooperative rabies management program activities in Vermont, 2006.

## ORV Naïve Monitoring

Wildlife Services refers to areas that have never been treated with ORV as “ORV naïve.” From 8-18 August 2006, WS conducted roadside trapping in the ORV naïve areas of Addison and Washington Counties that were baited 2 weeks later at a low bait density. During this effort, 35 unique raccoons and 1 skunk were trapped, processed and released. Tooth and blood serum samples were collected for age analysis and to measure the baseline presence of rabies VNA and tetracycline biomarker in these ORV naïve areas. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines

## Population Monitoring

In 2006, WS conducted 4 raccoon density studies, all in the low bait density zones of Addison and Washington Counties (Figure 1). The National Rabies Management Program (NRMP) standard protocol of 50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights was used. These studies coincided with post-ORV trapping and were conducted in areas of representative habitat found throughout the low bait density ORV zones (primarily agricultural). During the 4 studies, 43 unique raccoons and 21 unique skunks were captured and processed. All animals were released except for a skunk which was euthanized at the landowner's request. One raccoon was processed and released, then found dead near the trap the next day. Both animals tested negative for rabies. One additional skunk was trapped and released without processing because it was a late-year juvenile, too small to have consumed a bait. Raccoon density indices ranged from 1.1-4.4 raccoons/km<sup>2</sup> (Table 1). All animals euthanized by WS in 2006 were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

Table 1. Index to raccoon densities in Addison and Washington Counties, Vermont, 2006.

	Washington-A	Washington-B	Addison-C	Addison-D
Time of study	25 Sep.-5 Oct.	25 Sep.-5 Oct.	25 Sep.-5 Oct.	25 Sep.-5 Oct.
Macrohabitat	Agriculture	Agriculture	Agriculture	Agriculture
Target trap nights	500	500	500	500
Unique raccoons	12	6	14	11
Recaptured raccoons	3	3	5	4
Non-target captures <sup>b</sup>	21	2	14	13
Area (km <sup>2</sup> )	3.34	5.26	3.16	3.33
Raccoon density index <sup>a</sup>	3.6	1.1	4.4	3.3

<sup>a</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

<sup>b</sup> Includes traps that were sprung but with no animal captured.

## Post-ORV Monitoring

Post-ORV monitoring in the Vermont ORV zone was conducted 3-9 weeks after ORV bait distribution. Sampling included raccoons and skunks from population monitoring (above) and non-density study “roadside trapping”. In addition to the 43 raccoons and 10 skunks processed during the density studies, cage traps were used to capture and process 153 unique raccoons and 2 unique skunks from Addison, Franklin, Grand Isle, Orleans, and Washington Counties. One additional skunk was trapped and released without processing because it was a late-year juvenile, too small to have consumed a bait. Twenty-three raccoons were euthanized to reduce predation on nesting grounds of State Threatened turtles near Lake Champlain, within the bait zone; they all tested negative for rabies. All remaining animals were released.

*Hand Vaccination.* --On 27 October 2006, a skunk tested positive in Fairfax (Franklin County). This was the first case of raccoon variant in the county in 6 years (26 Oct. 2000). Wildlife Services responded by trapping around the case and hand vaccinating all raccoons and skunks captured. In early November, a skunk in Highgate was confirmed rabid and trapping commenced around that case as well. Trapping continued until 1 December when winter was setting in. During that time, 20 unique raccoons and 4 unique skunks were captured. Fifteen raccoons and 1 skunk were vaccinated and released, while 5 raccoons and 3 skunks were euthanized due to abnormal behavior or lesions; 1 raccoon tested positive for rabies. The skunk that was released by WS had originally been trapped,

vaccinated, and released by Quebec officials on 8 September 2006. On 14 November it was trapped by WS in Highgate, Vermont, approximately 32 km (20 mi) from its original point of capture near Cowansville, Quebec.

### Non-target Captures

Non-target (not processed) animals captured and released by WS in 2006 included: 5 domestic/feral cats (*Felis catus*), 5 Eastern cottontails (*Sylvilagus floridanus*), 4 fishers (*Martes pennanti*), 4 opossums (*Didelphis virginiana*), 3 porcupines (*Erethizon dorsatum*), 1 gray squirrel (*Sciurus carolinensis*), and 1 snowshoe hare (*Lepus americanus*). Additionally, WS captured and euthanized 2 red foxes (*Vulpes vulpes*) to reduce predation on nesting turtles and found 1 gray squirrel dead in a trap.

### Rabies Laboratory Cooperation

Wildlife Services' ORV program in Vermont cooperates with the VDH Laboratory (VDHL) and the New York State Department of Health's Rabies Laboratory at the Wadsworth Center (WC). Wildlife Services has had an efficient and cooperative relationship with both laboratories since 1997, and they remain critical to the surveillance and monitoring phases of the ORV program in Vermont.

*Vermont Department of Health Laboratory.*--The VDHL tests animal brainstems for rabies via routine public health surveillance throughout the state (specimens involved in a potential or confirmed exposure usually submitted by Game Wardens, WS, veterinarians, and the public). The VDHL also tests animals to enhance rabies surveillance in counties within the ORV zone (specimens not involved in an exposure and usually submitted by WS). The VDHL tested 473 animals for the rabies virus in 2006 (Table 2), representing a 54.6% increase from the number of samples tested in 2005. Animals were submitted from all 14 counties throughout the state, including the ORV counties. Of the raccoons tested statewide by the VDHL, 73.7% came from a county treated with ORV. The VDHL confirmed 73 rabid animals in 2006: 42 raccoons, 24 striped skunks, 4 cattle (*Bos taurus*), 1 red fox, 1 woodchuck (*Marmota monax*), and 1 big brown bat (*Eptesicus fuscus*) from 10 counties. All terrestrial animals had the raccoon variant of rabies.

Raccoons, skunks, foxes and coyotes (*Canis latrans*) are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 40.8% of the animals tested for rabies by the VDHL in 2006 are reported by WS as "other." For a full listing of rabid animals in Vermont by town, county and species from 2002 to the present please visit:

<http://healthvermont.gov/prevent/rabies/Rabies.aspx>

Table 2. Animals tested for rabies by the Vermont Department of Health Laboratory in Vermont, 2006 (statewide rabies positives in parentheses).

	Statewide	Within ORV counties
Raccoons	186 (42)	137 (73.7%)
Skunks	69 (24)	6 (8.7%)
Foxes	22 (1)	19 (86.4%)
Coyotes	2 (0)	1(50.0%)
Bobcat	1 (0)	0
Other <sup>a</sup>	193 (6)	62 (32.1%)
Total	473 (73)	225 (47.6%)

<sup>a</sup> Other animals included: bats, cats, dogs, other domestic pets, and woodchucks.

*New York State Department of Health's Rabies Laboratory at the Wadsworth Center.*--The WC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies VNA. In 2006, Vermont WS submitted 249 blood serum samples for rabies VNA analysis to the WC. This was similar to the number of samples submitted in 2005 (n=250). The Vermont ORV program anticipates over 800 serum sample submissions to the WC in 2007 due to planned large scale trap-vaccinate-release (TVR) efforts. For more information about the Rabies Laboratory at the WC please visit: <http://www.wadsworth.org/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon and skunk blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and teeth are analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain a biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, WS live-trapped 240 raccoons and 11 skunks, 4-9 weeks after bait distribution, to evaluate ORV efficacy. Blood samples were collected from all but 1 raccoon and sera were sent to the WC; 38 raccoons (15.9%) showed presence of rabies VNA ( $\geq 0.05$  IU). No skunks demonstrated a detectable rabies antibody response. Tooth samples were collected from most raccoons (no skunks) and sent to Matson's Laboratory LLC (Milltown, Montana, USA). The raccoon tooth samples collected were analyzed for tetracycline biomarker presence (n=220), although FMP baits have not been aerially distributed in the Vermont ORV zone (on a broad scale) since 2000. In 2005, FMP baits were distributed by hand in 24 small cities and villages and likely account for the 15 raccoons (6.8%) that showed the presence of tetracycline biomarker.

In 2006, WS live-trapped 251 unique raccoons and blood and tooth samples were collected from most of these animals (Table 3). Serum samples were sent to the WC, while tooth samples were sent to Matson's Lab. The tooth samples that showed presence of tetracycline biomarker were likely from hand baited areas where FMP baits are distributed. In addition, blood samples were collected from 13 skunks and 1 red fox; there were no detectable rabies antibodies in these samples. Four skunk teeth were tested for biomarker, but none showed the presence of tetracycline.

Table 3. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during cooperative rabies management program activities in Vermont, 2006.

	Low density zone (ORV <sup>a</sup> naïve)	Low density zone (post-ORV)	Standard density zone (post-ORV)	Hand vaccination (post-ORV)
Sample collection timeframe	8-18 Aug.	26 Sep.-27 Oct.	24 Sep.-20 Oct.	2 Nov.-1 Dec.
Weeks post-ORV	n/a <sup>b</sup>	5-9	5-8	10-14
Bait density (baits/km <sup>2</sup> )	n/a	35	70	70
ORV bait type/ distribution method	n/a	CS <sup>c</sup> /fixed-wing & FMP/hand	CS/fixed-wing & FMP/hand	CS/fixed-wing & FMP/hand
Unique raccoons	35	46	150	20
<b>Serology</b>				
Testable blood samples	34	46	126	17
Positive rabies antibody response ( $\geq 0.05$ IU)	3 (8.8%)	10 (21.7%)	25 (19.8%)	4 (23.5%)
<b>Tetracycline biomarker<sup>c</sup></b>				
Testable tooth samples	23	34	124	2
Presence of tetracycline biomarker	0	0	10 (8.1%)	0

<sup>a</sup> ORV=oral rabies vaccination; CS=coated sachet; FMP=fishmeal polymer.

<sup>b</sup> Samples were collected in an ORV naïve area (never before treated with ORV).

<sup>c</sup> In addition, 20 raccoon tooth samples were collected throughout 2006 during enhanced surveillance activities; none showed presence of tetracycline.

*Age Results.*--In 2005 and 2006, 221 and 207 raccoon teeth were aged using premolars of live-captured animals collected throughout the field seasons (Figure 2). Age results were typical in both years, with the populations dominated by animals  $\leq 1$  y.o. In addition, teeth from 6 skunks (4 at  $<1$  y.o.; 1 at 1 y.o.; 1 at 4 y.o.) and 1 red fox ( $<1$  y.o.) were aged.

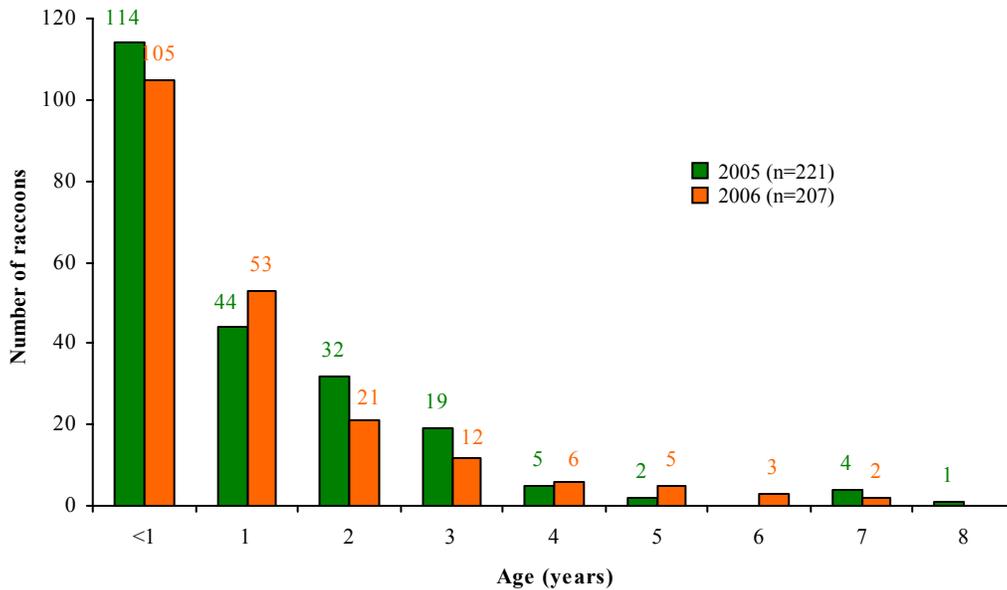


Figure 2. Age class distribution of 221 and 207 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in Vermont, 2005 and 2006.

## SUMMARY

The summer of 2006 marked the tenth year of WS cooperative participation in the Vermont ORV Program. Over those 10 years, the distribution of 2.5 million ORV baits helped prevent raccoon rabies from reaching Quebec, Canada. In June 2006 however, the raccoon variant was confirmed approximately 11 km (6.6 mi) north of the Vermont border in Dunham, Quebec. In response, Vermont WS established a network of cooperators to assist with enhancing rabies surveillance in the ORV bait zones. By year's end, WS had collected 139 animals (up from 7 samples in 2005) and 29 were confirmed rabid. Two low bait density (35/km<sup>2</sup>) ORV zones were established in the middle of the state and 4 density studies were conducted there (ranging from 1.1-4.4 raccoons/km<sup>2</sup>). Baits were distributed in northern Vermont at a standard rate of 70/km<sup>2</sup> and will be compared to the low bait zones. The efficacy of baiting at a lower density may prove more cost-effective in areas with low raccoon densities. Trapping post-ORV yielded 179 animals including 16 animals that were hand vaccinated in an effort to quell rabies outbreaks heading into the winter season.

In 2007, WS will implement a large-scale TVR effort in northwestern Vermont to bolster the percentage of vaccinated animals and suppress the current rabies outbreak. Wildlife Services will continue to enhance rabies surveillance, distribute ORV baits, conduct density studies, and trap post-ORV to monitor program effectiveness. Wildlife Services will continue to communicate closely and coordinate field work with Quebec officials to maximize efforts to contain, and explore strategies to eliminate, the raccoon variant of the rabies virus.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM VIRGINIA 2006

### BACKGROUND

In 1977, raccoon (*Procyon lotor*) rabies was first introduced to the mid-Atlantic region of the United States with the translocation of infected raccoons from Florida to Hardy County, West Virginia and Shenandoah County, Virginia. From these counties, the disease spread rapidly along the east coast and has now become enzootic in all of the East Coast states, as well as Alabama, Pennsylvania, Vermont, West Virginia, and eastern Ohio. Wildlife Services (WS) has been involved in an oral rabies vaccination (ORV) program to control raccoon rabies in Virginia since 2002. The ORV program in Virginia is part of the Appalachian Ridge (AR) ORV project and of a larger, cooperative effort to stop the westward spread of the raccoon variant of rabies in the eastern United States.

The raccoon variant of rabies occurs throughout Virginia, with the exception of 3 counties (Dickenson, Lee, and Wise) on the southwestern border with Kentucky (Figure 1). According to the Virginia Department of Health (VDH), Office of Epidemiology, infected raccoons accounted for 311 (48.8%) of the 637 animal rabies cases reported in Virginia during 2006. During the past 9 years, January 1998-December 2006, raccoons have accounted for 2,759 (55.8%) of the 4,945 laboratory confirmed cases of animal rabies statewide. Wildlife Services is working in cooperation with the VDH, the Virginia Department of Game and Inland Fisheries (VDGIF), and local animal control officers to provide as many rabies surveillance specimens as possible.

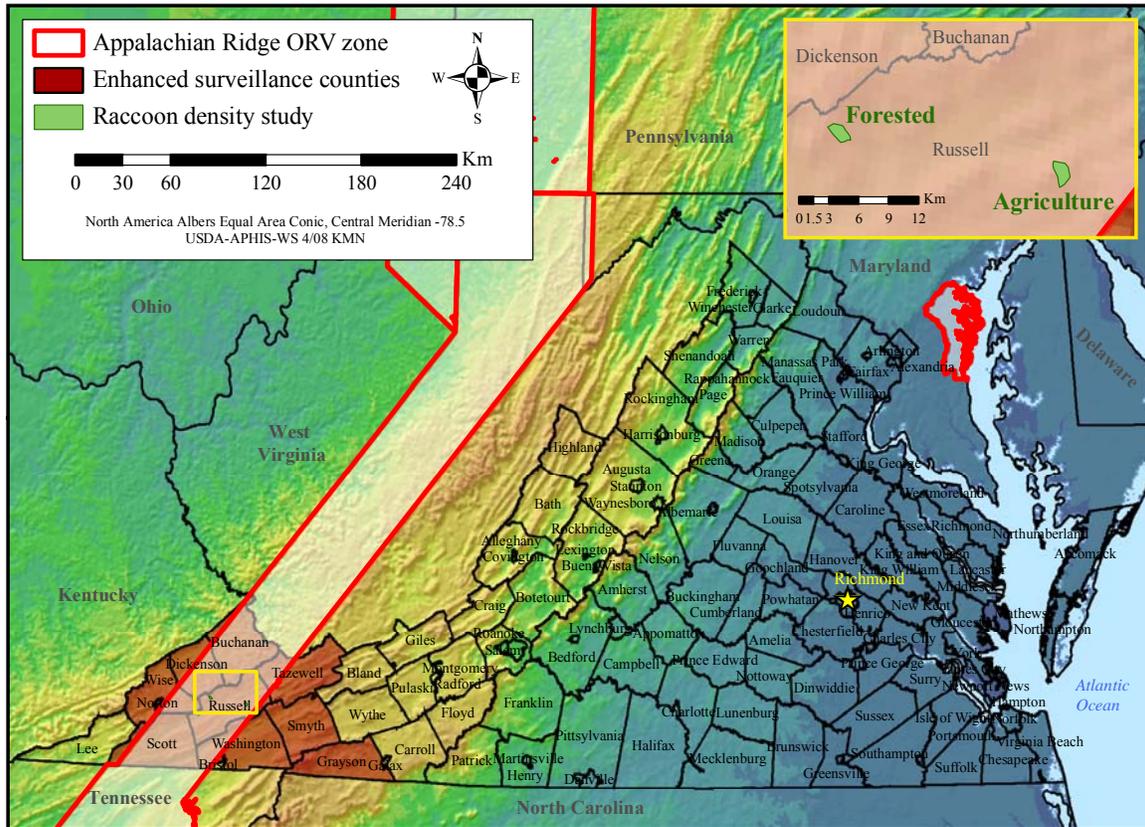


Figure 1. Wildlife Services cooperative rabies management program activities in Virginia, 2006.

### ORV PROGRAM 2006

#### Bait Distribution

For the fifth consecutive year, WS participated in bait distribution efforts in southwestern Virginia as part of the larger AR ORV zone; 320,617 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia,

USA) were distributed over 5,024 km<sup>2</sup> (1,940 mi<sup>2</sup>) in 2006 (Figure 1). Since its program inception in 2002, WS has cooperated to distribute 1,786,441 ORV baits in Virginia.

From 7-16 August 2006, 309,848 fishmeal-coated sachet (CS) baits were distributed over 8 counties in southwestern Virginia. Fixed-wing aircraft were provided by Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA), while WS personnel served as navigators and flight crew in the rear of the plane. Planes flew at approximately 150-300 meters altitude and flight lines were spaced approximately 500 meters apart. Baits are distributed via ground operations (hand and vehicle) in areas too populated to bait by air. From 29-31 August, WS distributed 10,769 fishmeal polymer (FMP) ORV baits in several urban and suburban areas of southwest Virginia: Castlewood, Cedar Bluff, Claypool Hill, Coeburn, Gate City, Grundy, Lebanon, Raven, Richlands, St. Paul, and Weber City.

### Enhanced Surveillance

During 2006, WS cooperated with the VDH, the Virginia Department of Transportation, the VDGIF, county animal control officers, and local trappers to collect 196 animals from counties in or adjacent to the ORV zone to enhance rabies surveillance in southwestern Virginia. These samples were from road killed, strange-acting, injured, or nuisance animals and from carcasses collected from fur trappers that otherwise would not have been tested through the public health surveillance system. The Virginia Department of General Services – Division of Consolidated Laboratory Services (VDCLS) tested 9 of these animals: 5 raccoons and 1 striped skunk (*Mephitis mephitis*) from Russell County, 2 raccoons from Tazewell County, and 1 striped skunk from Scott County. One raccoon each from Russell and Tazewell Counties tested positive for rabies.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

In 2006, WS tested 187 of the 196 enhanced surveillance samples (95.4%) using the dRIT and 1 raccoon tested positive for rabies (Table 1).

Table 1. Animals tested for rabies by Wildlife Services using the direct rapid immunohistochemistry test (dRIT) from counties within or adjacent to the oral rabies vaccination (ORV) zone in Virginia, 2006 (rabies positives in parentheses).

County	Raccoon	Striped Skunk	Coyote	Red fox	Gray fox	Bobcat	Total
Buchanan	3						3
Dickenson	9		2				11
Grayson			4				4
Russell	40	1	1	1	2		45
Scott	10		1				11
Smyth	47(1) <sup>a</sup>	16	9	11	5	1	89 (1)
Tazewell	9	1					10
Washington	5	1				1	7
Wise	6			1			7
Total	129 (1)	19	17	13	7	2	187 (1)

<sup>a</sup> Collected east of the ORV zone.

All positives and 10% of all negative samples were sent to the CDC for confirmation and strain typing. The CDC (using the dFA test) had 100% agreement with the WS dRIT results for positive and negative samples. Antigenic typing conducted by the CDC determined that the rabies virus in the positive raccoon from Smyth County was consistent with the raccoon variant found in the eastern U.S. Wildlife Services will continue to use the dRIT in 2007 to enhance surveillance of suspect rabid animals in Virginia.

## Population Monitoring

In 2006, WS conducted 2 raccoon density studies in Russell County using the National Rabies Management Program standard protocol of 50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights (Figure 1 inset). Both studies were conducted in areas representative of habitat found throughout the Virginia ORV zone; one on an agricultural area and one on a forested area. Over 1,000 trap nights, 53 unique raccoons were captured and blood and tooth samples were collected from most of them (Table 2). During the study on the agricultural area, 33 raccoons were immobilized, processed and released, while 8 raccoons were euthanized at the landowner's request or because of injuries/wounds. Brainstem samples were collected from the 8 raccoons and tested for rabies by WS using the dRIT (Table 1). All samples tested negative for the presence of rabies. During the study on the forested area, all raccoons were immobilized, processed and released. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with American Veterinary Medical Association's Panel on Euthanasia recommendations.

Table 2. Index to raccoon densities in Russell County, Virginia, 2006.

	Russell	Russell
Time of study	6-16 July	19-29 July
Macrohabitat	Agriculture	Forested
Target trap nights	500	500
Unique raccoons	41	12
Recaptured raccoons	6	4
Non-target captures	24	7
Area (km <sup>2</sup> )	2.76	2.42
Raccoon density index <sup>a</sup>	14.9	5.0

<sup>a</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

## Post-ORV Monitoring

During September and October 2006, WS conducted trapping activities 4-7 weeks post-aerial ORV bait distribution in Buchanan, Dickenson, Russell, Scott, and Wise Counties. Over 1,241 trap nights, 194 unique raccoons were captured on public and private lands (187 and 175 serum and tooth samples were collected, respectively). Four raccoons were found dead at trap sites, 18 raccoons were euthanized due to injury or at the request of landowners, and 173 raccoons were immobilized, processed and released.

## Non-target Captures

Non-target animals captured and released by WS in 2006 included: 69 opossums (*Didelphis virginiana*), 5 Eastern box turtles (*Terrapene carolina*), 3 domestic/feral cats (*Felis catus*), 2 domestic/feral dogs (*Canis familiaris*), 2 Eastern cottontails (*Sylvilagus floridanus*), 2 snapping turtles (*Chelydra serpentina*), and 1 beaver (*Castor Canadensis*).

Non-target animals that were captured and euthanized by WS in 2006 included: 10 opossums and 9 woodchucks (*Marmota monax*).

## Laboratory Cooperation

The ORV program in Virginia cooperates with the VDCLS and the CDC.

*Virginia Division of Consolidated Laboratory Services.*--The VDCLS tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure). They also work with WS to test a limited number of enhanced surveillance samples (specimens not involved in an exposure) from areas within and adjacent to the ORV zone. In 2006, the VDCLS tested 4,180 samples for the rabies virus and confirmed 637 cases of rabies (Table 3).

Raccoons, skunks, foxes, and bobcats are of priority interest to WS and cooperators involved in ORV. These carnivores are common rabies vectors throughout the U.S. and the animals most frequently collected and

submitted by WS to enhance rabies surveillance. Many additional species, when involved in human or domestic animal exposures to the rabies virus, are of priority interest to public health agencies. This explains why 74.2% of the animals tested for rabies in Virginia in 2006 are reported by WS as “other.” For a full listing of rabid animals from Virginia in 2006 please visit the VDH’s rabies website: <http://www.vdh.virginia.gov/epidemiology/DZEE/Rabies/>

Table 3. Animals tested for rabies by the Virginia Department of General Services – Division of Consolidated Laboratory Services in Virginia, 2006.

	Statewide		Within and adjacent to Virginia ORV <sup>a</sup> zone <sup>b</sup>	
	Number tested	Number rabid	Number tested	Number rabid
Raccoons	659	311 (47.2%)	27 (4.1%)	6
Skunks	223	156 (70.0%)	4 (1.8%)	0
Foxes	190	80 (42.1%)	3 (1.6%)	0
Bobcats	6	6 (100%)	0	0
Other <sup>c</sup>	3,102	84 (2.7%)	127 (4.1%)	0
Total	4,180	637 (15.2%)	161 (3.9%)	6

<sup>a</sup> ORV=oral rabies vaccination.

<sup>b</sup> Samples from Buchanan, Dickenson, Lee, Russell, Scott, Smyth, Tazewell, Washington, and Wise Counties and the cities of Bristol and Norton.

<sup>c</sup> Other animals included: bats, cats (domestic/feral), cows, dogs (domestic/feral), goats, horses, and woodchucks (groundhogs).

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA). Virginia WS submitted 238 blood serum samples for rabies VNA analysis to the CDC in 2006. This represents a 16.5% decrease from the 285 samples submitted by WS in 2005. The Virginia ORV program anticipates similar numbers of serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology, and prevention and control) on a national level please visit the CDC’s rabies homepage: <http://www.cdc.gov/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year’s monitoring to provide time for laboratory analyses. At the time of this report however, the current year’s (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005, WS live-trapped 92 raccoons during raccoon density studies conducted prior to annual ORV in ORV naïve areas (never before treated with ORV) and 196 raccoons during fall post-ORV evaluation activities (Table 4). In 2006, WS live-trapped 247 raccoons during raccoon density studies conducted prior to annual ORV and fall post-ORV evaluation activities (Table 4). In both years, blood and tooth samples were collected from most of these animals and serum samples were sent to the CDC, while tooth samples were sent to Matson’s Laboratory LLC (Milltown, Montana, USA).

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during cooperative rabies management program activities in Virginia, 2005 and 2006.

	Density studies 2005	AR <sup>a</sup> evaluation 2005	Density studies 2006	AR evaluation 2006
Sample collection timeframe	6-29 Jul.	12 Sep.-7 Oct.	6-29 Jul.	11 Sep.-6 Oct.
Weeks post-ORV <sup>a</sup>	n/a <sup>b</sup>	4-7	44-47	4-7
ORV bait type/ distribution method	n/a	CS <sup>3</sup> /fixed-wing & FMP/hand	CS/fixed-wing & FMP/hand	CS/fixed-wing & FMP/hand
Unique raccoons	92	196	53	194
<b>Serology</b>				
Testable blood samples	91	194	51	187
Positive rabies antibody response (≥0.05 IU)	0	63 (32.5%)	28 (54.9%)	107 (57.2%)
<b>Tetracycline biomarker</b>				
Testable tooth samples	92	192	44	175
Presence of tetracycline biomarker	11 (12.0%)	68 (35.4%)	6 (13.6%)	37 (21.1%)

<sup>a</sup> AR=Appalachian Ridge; ORV=oral rabies vaccination; CS=coated sachet; FMP=fishmeal polymer.

<sup>b</sup> Samples were collected in an ORV naïve area (never before treated with ORV).

*Age Results.*--In 2005 and 2006, 283 and 219 raccoon teeth, respectively, were aged using premolars of live-captured animals collected during summer density studies and fall post-ORV trapping activities (Figure 2). Age results are typical, with the populations dominated by young of the year juveniles (<1 y.o.) and 1 and 2 y.o. animals.

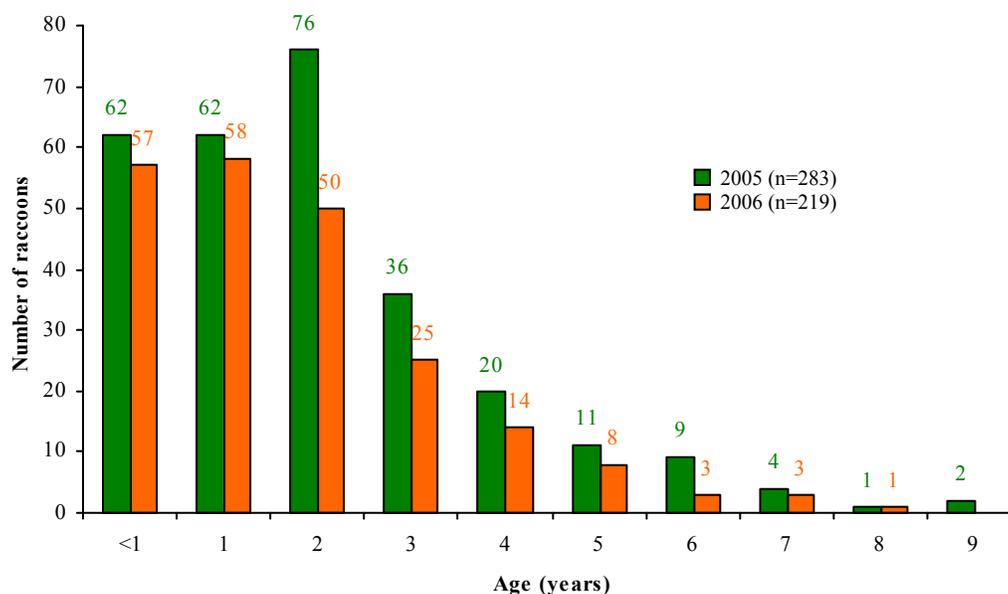


Figure 2. Age class distribution of 283 and 219 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in Virginia, 2005 and 2006.

## SUMMARY

During 2006, WS completed its sixth year of cooperative participation in rabies management in Virginia, and its fifth year conducting ORV bait distribution activities. Other activities in 2006 included: enhancing surveillance of raccoon rabies by collecting and testing road killed, strange-acting and nuisance animals from counties within and adjacent to the ORV zone; and trapping post-ORV bait distribution to monitor and evaluate raccoon serology and biomarker uptake in southwestern Virginia. In 2007, WS will continue to conduct raccoon density studies and enhanced rabies surveillance within and adjacent to the current AR ORV zone. Future ORV baiting strategies in Virginia will continue to be tied to national and international planning efforts to contain, and explore strategies to eliminate, the raccoon variant of the rabies virus in North America.



The West Virginia ORV program is coordinated by the WV Department of Agriculture (WVDOA), while surveillance is being conducted by the WV Department of Health and Human Resources (WVDHHR), WS, and County Departments of Health. Wildlife Services provides funding and operational support, including coordination of ORV bait distribution activities, raccoon population monitoring, and program evaluation through live trapping efforts. The West Virginia Division of Natural Resources (WVDNR) provides permits for handling wildlife, access to state owned property, and expertise in raccoon ecology and management.

## **ORV PROGRAM 2006**

### **Bait Distribution**

For the sixth consecutive year, WS participated in bait distribution efforts along the AR in West Virginia via 2 distinct baiting efforts (a south and north campaign); 1,503,365 baits containing Raboral V-RG® vaccine (Merial Limited, Athens, Georgia, USA) were distributed over 22,534 km<sup>2</sup> (8,701 mi<sup>2</sup>) in 2006 (Figure 1). Since its program inception in 2001, WS has cooperated to distribute 8,697,702 ORV baits in West Virginia. Ground support for aerial baiting was provided by: the WVDHHR, the WVDOA, the WVDNR, the Ohio National Guard, the Ohio Department of Health, and WS employees from West Virginia, Virginia, Tennessee, Kentucky, Ohio, and Pennsylvania.

*Appalachian Ridge South.*--In 2006, the southern portion of the West Virginia AR ORV zone covered 18,125 km<sup>2</sup> (6,998 mi<sup>2</sup>) in 24 counties (Figure 1). From 7-16 August, 1,190,316 ORV baits were distributed; 890,144 fishmeal-coated sachet (CS) and 300,172 fishmeal polymer (FMP) baits via fixed-wing aircraft. From 25-29 September, 17,012 FMP baits were distributed via ground operations (hand baiting). Fixed-wing aircraft and pilots were provided by Dynamic Aviation Group Inc. (Bridgewater, Virginia, USA), while WS personnel served as navigators and flight crew in the planes.

*Appalachian Ridge North.*--In 2006, the northern portion of the West Virginia AR ORV zone covered 4,409 km<sup>2</sup> (1,702 mi<sup>2</sup>) in 11 counties (Figure 1). From 5-21 September, 285,546 CS baits were distributed via fixed-wing aircraft. From 25-29 September, 10,491 FMP baits were distributed via hand baiting. Fixed-wing aircraft were provided by the Ontario Ministry of Natural Resources, while WS personnel served as navigators and flight crew in the rear of the plane.

### **Enhanced Surveillance**

In 2006, WS enhanced surveillance of raccoon rabies by collecting and submitting 338 animals from 41 counties in or adjacent to the West Virginia ORV zone (Table 1). Of the animals collected, 76 were euthanized due to abnormal behavior (73 raccoons, 1 gray fox (*Urocyon cinereoargenteus*), 1 woodchuck (*Marmota monax*), and 1 bat (*Chiroptera* spp.). The other 262 animals were collected during road kill surveillance. Eight of 338 animals (2.4%) tested positive for rabies; they were all raccoon variant. All animals captured by WS in 2006 were handled according to the American Society of Mammalogists, Animal Care and Use Committee guidelines and all animals euthanized were done so in accordance with the American Veterinary Medical Association's Panel on Euthanasia recommendations.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the direct fluorescent antibody (dFA) test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Wildlife Services personnel attended dRIT training in March 2005 and December 2006 at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia. Wildlife Services began implementation of the dRIT in January 2006 and by year's end had tested all 338 enhanced surveillance specimens.

Table 1. Animals tested for rabies by Wildlife Services using the direct rapid immunohistochemistry test (dRIT) from counties within or adjacent to the oral rabies vaccination (ORV) zone in West Virginia, 2006 (rabies positives in parentheses).

County	Raccoon	Skunk	Red fox	Gray fox	Coyote	Bobcat	Bat	Opossum	Woodchuck	Total
Barbour	17									17
Boone	5	1			1					7
Braxton	2									2
Brooke	8									8
Cabell <sup>a</sup>	1									1
Calhoun <sup>a</sup>	9									9
Clay			1							1
Doddridge	3									3
Fayette	1									1
Gilmer <sup>a</sup>	11									11
Grant <sup>a</sup>							1			1
Hancock	2									2
Hardy <sup>a</sup>	1									1
Harrison	1									1
Jackson <sup>a</sup>	6									6
Kanawha	7	1								8
Lewis	15		1		1					17
Lincoln <sup>a</sup>	3									3
Logan	1				1					2
Marion	2									2
Marshall	52	1		1				2		56
Mingo	1									1
Monongalia	2									2
Nicholas	3									3
Ohio	3									3
Pendleton <sup>a</sup>	11 (6)	1 (1)		1 (1)						13 (8)
Pleasants <sup>a</sup>	7									7
Preston	26								1	27
Raleigh	5									5
Randolph	11	1		1						13
Ritchie	25		1							26
Roane <sup>a</sup>	5	1								6
Taylor	1									1
Tyler	4									4
Upshur	25			1						26
Wayne <sup>a</sup>	2	1								3
Webster	1									1
Wetzel	5		1							6
Wirt <sup>a</sup>	5						1			6
Wood <sup>a</sup>	22	2	1							25
Wyoming		1								1
<b>Total</b>	<b>311 (6)</b>	<b>10 (1)</b>	<b>5</b>	<b>4 (1)</b>	<b>3</b>		<b>2</b>	<b>2</b>	<b>1</b>	<b>338 (8)</b>

<sup>a</sup> ORV not applied in this county.

## Population Monitoring

In 2006, WS conducted 5 raccoon density studies (1 each in Hardy, Greenbrier, Barbour, Wetzel, and Jackson Counties) using the National Rabies Management Program (NRMP) standard protocol of 50 cage traps set on a target study area of 3 km<sup>2</sup> for 10 consecutive nights (Figure 1). The Barbour and Wetzel studies were each conducted on wildlife management areas, managed by the WVDNR, and were representative of habitat found throughout much of the state with forested rolling hills and permanent streams; the Barbour site also had 3 large corn food plots. The Hardy and Jackson study sites were mostly agricultural (composed of forest, pasture, and corn fields). The Greenbrier study site was composed of pasture and forest lands. The Barbour and Wetzel studies were conducted in the ORV zone, while the Hardy, Greenbrier, and Jackson studies were conducted outside of the bait zone in ORV naïve areas (never previously treated with ORV). Raccoon density estimates are critical for use in future plans to potentially shift the ORV zone to the east. During the 5 studies, 140 unique raccoons were captured and density indices ranged from 4.1-14.2 raccoons/km<sup>2</sup> (Table 2). All raccoons were immobilized, processed and released.

Table 2. Index to raccoon densities in Hardy, Greenbrier, Barbour, Wetzel, and Jackson Counties, West Virginia, 2006.

	Hardy <sup>a</sup>	Greenbrier <sup>a</sup>	Barbour	Wetzel	Jackson <sup>a</sup>
Time of study	12-22 June	10-20 July	11-21 July	11-21 July	24 July-3 Aug.
Macrohabitat	Agriculture	Agriculture	Agriculture	Forested	Agriculture
Target trap nights	500	500	500	500	500
Unique raccoons	26	39	29	12	34
Recaptured raccoons	11	17	19	9	11
Non-target captures	53	14	21	10	14
Area (km <sup>2</sup> )	2.95	2.94	3.27	2.92	2.39
Raccoon density index <sup>b</sup>	8.8	13.3	8.9	4.1	14.2

<sup>a</sup> ORV not applied in this county.

<sup>b</sup> Raccoon density index (raccoons/km<sup>2</sup>) = unique raccoons ÷ area.

## Post-ORV Monitoring

From September-November 2006, WS conducted trapping activities 4-11 weeks post-ORV bait distribution in Barbour, Brooke, Harrison, Lewis, Monongalia, Nicholas, Preston, and Upshur Counties. As a result, biological samples were collected from 239 animals: 224 raccoons and 15 striped skunks (*Mephitis mephitis*). Of these, 102 raccoons and 3 skunks were euthanized at the landowner's request, while the remaining animals were released. Of the 105 animals euthanized, 18 animals were tested by WS and were negative for rabies. WS will continue collecting brainstem samples in 2007 on all euthanized animals.

## Other Rabies Management Program Activities

*Habitat Characteristics Study.*--For the fifth consecutive year, WS collected habitat data to determine the relationship of habitat composition and elevation to raccoon relative abundance (Table 3). Habitat characteristics are important variables that need further research because the historical spread of rabies through West Virginia appeared to be slowed by the higher elevations of the Appalachian Mountains. Several habitat characteristics were measured at 100 locations during raccoon trapping activities. At each trap location a 7.97 m (26.3 ft) diameter plot was sampled, with the trap centered in each plot. Wildlife Services will continue to collect these data in 2007.

Table 3. Mean habitat characteristics measured at trap locations that captured and did not capture raccoons in West Virginia, 2006.

Habitat Characteristics	Capture site	Non-capture site
Basal area <sup>a</sup> (ft <sup>2</sup> )	4.31	5.34
Trees >11" dbh <sup>b</sup>	2.61	3.92
Trees <11" dbh	8.29	9.00
Ground vegetation cover (%)	71.85	78.55
Canopy cover (%)	64.60	68.95
Distance (m) to water-intermittent	8.83	14.50
Distance (m) to water-permanent	41.86	33.38
Slope (%)	4.95	6.29
Elevation (ft)	1521	1,939
Den sites per plot	0.42	0.26
Number of tree/shrub species per plot	2.39	2.19
Red Oak ( <i>Quercus rubra</i> )	3.44	3.29
White Oak ( <i>Quercus alba</i> )	8.67	6.75
Beech ( <i>Fagus grandifolia</i> )	0	1.00
Hickory ( <i>Carya spp.</i> )	2.56	3.00
Black walnut ( <i>Juglans nigra</i> )	2.50	2.00
Common apple ( <i>Malus sylvestris</i> )	0	1.50
Black cherry ( <i>Prunus serotina</i> )	1.40	2.50
Flowering dogwood ( <i>Cornus florida</i> )	1.00	1.00
Blackberry ( <i>Rubus spp.</i> )	7.67	1.00
Autumn Olive ( <i>Elaeagnus umbellata</i> )	1.00	2.00
Multiflora Rose ( <i>Rosa multiflora</i> )	2.80	4.43

<sup>a</sup> Calculated using a 20-factor prism.

<sup>b</sup> dbh=diameter at breast height.

### Non-target Captures

Non-target animals captured and released by WS in 2006 included: 107 opossums (*Didelphis virginiana*), 11 Eastern cottontails (*Sylvilagus floridanus*), 11 woodchucks, 9 domestic cats (*Felis catus*), 4 fox squirrels (*Sciurus niger*), 2 domestic dogs (*Canis familiaris*), 1 gray squirrel (*Sciurus carolinensis*), 1 sparrow (*Passeridae* spp.), and 1 turtle (*Emydidae* spp.).

Non-target animals that were captured and euthanized by WS in 2006 included: 26 woodchucks, 15 opossums, and 1 fox squirrel. These animals were euthanized at the request of landowners.

### Rabies Laboratory Cooperation

The ORV program in West Virginia cooperates with the WVDHHR and the CDC.

*West Virginia Department of Health and Human Services.*--The WVDHHR tests animal brainstems for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure usually submitted by conservation officers, veterinarians, and the public) and enhanced surveillance (specimens not involved in an exposure and usually submitted by WS). In 2006, the WVDHHR tested 1,250 samples for the rabies virus and confirmed 118 cases of rabies in West Virginia. Including the rabid animals from dRIT, 126 cases of rabies were confirmed in West Virginia in 2006: 79 raccoons, 23 striped skunks, 9 foxes, 7 bats, 3 cats, 1 bobcat (*Lynx rufus*), 1 cow (*Bos taurus*), 1 dog, 1 goat (*Capra aegagrus hircus*), and 1 woodchuck.

Of the 1,250 samples tested by the WVDHHR, 497 were tested because of human exposures, 239 were tested because of domestic animal exposures, and 514 were tested for other reasons. The 2006 samples were submitted from all 55 counties throughout the state. For a full listing of rabid animals from West Virginia by species and location from 2002-2006 please visit the WVDHHR's rabies website:

<http://www.wvdhhr.org/idep/a-z/a-z-rabies.asp>

*Centers for Disease Control and Prevention.*--The CDC analyzes wildlife blood serum samples (submitted by WS) for levels of rabies virus neutralizing antibodies (VNA). West Virginia WS submitted 382 blood serum samples for rabies VNA analysis to the CDC in 2006. This represents a 9.5% increase from the 349 samples submitted by WS in 2005. The West Virginia ORV program anticipates similar numbers of serum sample submissions to the CDC in 2007. For more information about the rabies virus (its natural history, diagnosis, epidemiology on a national scale, and prevention and control) please visit the CDC's rabies homepage: <http://www.cdc.gov/ncidod/dvrd/rabies/>

## ORV PROGRAM 2005 and 2006 – EVALUATION

Typically, WS presents ORV program evaluation data (serology, tetracycline, and age results) from the previous year's monitoring to provide time for laboratory analyses. At the time of this report however, the current year's (2006) evaluation data were available so they have been included here as well.

### Serology, Tetracycline Biomarker, and Age Results

*Serology and Biomarker Results.*--Raccoon blood sera are analyzed to detect rabies VNA (or rabies vaccination levels) and the tooth is analyzed to determine animal age and bait uptake (when appropriate: FMP baits contain a chemical biomarker, tetracycline, which stains teeth/bone and can be detected under microscope; CS baits do not contain this biomarker). Presence of tetracycline in a tooth may indicate that the animal consumed part of the FMP bait matrix (outer portion of the bait). However, presence of tetracycline does not confirm that the vaccine sachet was punctured or consumed, thus the need for sera evaluation as well.

In 2005 and 2006, WS live-trapped 355 and 372 raccoons, respectively, during summer density studies and fall post-ORV evaluation activities (Table 4). Blood and tooth samples were collected from most of these animals and serum samples were sent to the CDC, while tooth samples were sent to Matson's Laboratory LLC (Milltown, Montana, USA). Post-ORV trapping in 2006 included serum samples from the AR North zone as well (included in Table 4): 14 of 47 (29.8%) demonstrated a positive rabies antibody response.

Table 4. Serology and tetracycline biomarker results of raccoon biological samples collected by Wildlife Services during cooperative rabies management program activities in West Virginia, 2005 and 2006.

	2005			2006		
	Density studies (ORV naïve) <sup>a</sup>	Density studies (prior to ORV) <sup>b</sup>	Fall AR <sup>c</sup> south (post-ORV)	Density studies (ORV naïve) <sup>a</sup>	Density studies (prior to ORV) <sup>d</sup>	Fall AR (post-ORV)
Sample collection timeframe	6-16 June	11-28 July	13 Sep.-14 Oct.	13 June -30 July	11-20 July	20 Sep.-3 Nov.
Weeks post-ORV	n/a	39-42	4-8	n/a	48-49	4-11
ORV bait type	n/a	FMP <sup>c</sup>	FMP & CS <sup>c</sup>	n/a	FMP & CS <sup>c</sup>	CS & FMP
Unique raccoons	21	55	279	97	51	224
<b>Serology</b>						
Testable blood samples	21	52	275	95	50	212
Positive rabies antibody response (≥0.05 IU)	1 (0.05%)	18 (34.6%)	51 (18.5%)	17 (17.9%)	24 (48.0%)	76 (35.8%)
<b>Tetracycline biomarker</b>						
Testable tooth samples	15	38	239	72	30	175
Presence of tetracycline biomarker	0	28 (73.7%)	112 (46.9%)	0	16 (53.3%)	32 (18.3%)

<sup>a</sup> Density studies conducted in Hardy County (2005) and Greenbrier, Hardy, and Jackson Counties (2006) were ORV naïve (never before treated with ORV).

<sup>b</sup> Density studies conducted in Barbour, Wetzel, and Wyoming Counties were last treated with ORV in September 2004.

<sup>c</sup> AR=Appalachian Ridge; CS=coated sachet; FMP=fishmeal polymer.

<sup>d</sup> Density studies conducted in Barbour and Wetzel Counties were last treated with ORV in August 2005.

*Age Results.*--In 2005 and 2006, 271 and 268 raccoon teeth were aged using premolars of live-captured animals collected during summer density studies and fall post-ORV trapping activities (Figure 2). Age results were typical in both years, with the populations dominated by animals  $\leq 1$  y.o.

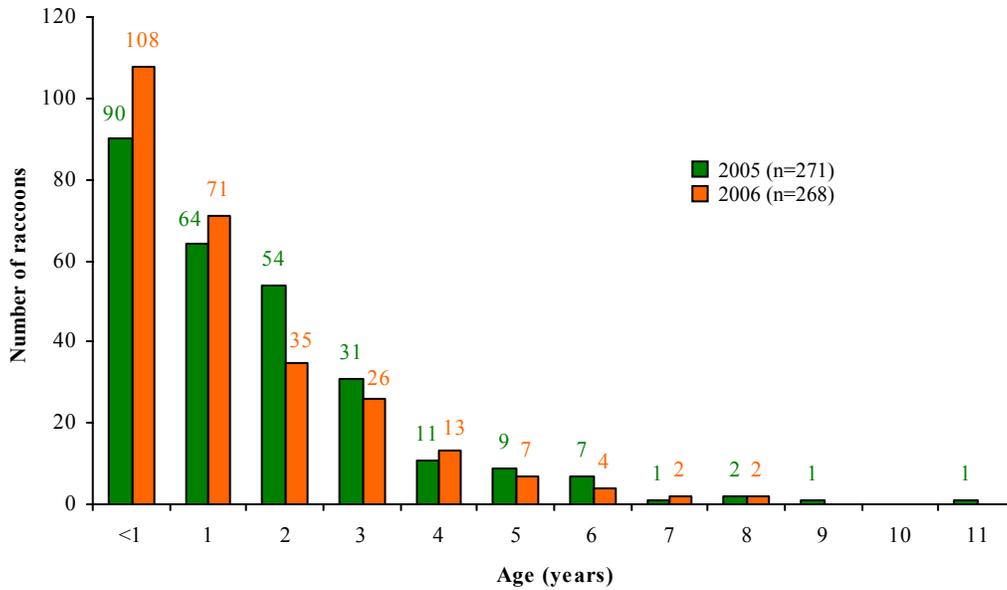


Figure 2. Age class distribution of 271 and 268 raccoon tooth samples collected by Wildlife Services during the cooperative rabies management program in West Virginia, 2005 and 2006.

## SUMMARY

During 2006, WS completed its sixth year of cooperative participation in rabies management in West Virginia including ORV bait distribution activities. Other activities in 2006 included: enhancing surveillance of raccoon rabies by collecting and testing road killed, strange acting and nuisance animals from counties within and adjacent to the ORV zone; conducting raccoon population density studies within the AR ORV zone and in ORV naïve areas east of the zone; and trapping post-ORV bait distribution to monitor and evaluate raccoon serology and biomarker uptake throughout West Virginia’s portion of the AR ORV zone.

In 2007, WS will continue to: conduct enhanced rabies surveillance within and adjacent to the current AR ORV zone; investigate the influence of elevation and habitat by collecting pertinent habitat data at each trap location; conduct raccoon density estimates at sites >1,067 meters in elevation; and monitor and evaluate ORV program efficacy by collecting raccoon serology.

## WILDLIFE SERVICES COOPERATIVE RABIES MANAGEMENT PROGRAM WYOMING 2006

### BACKGROUND

The striped skunk (*Mephitis mephitis*) is the primary reservoir of terrestrial rabies in Wyoming. The North Central skunk variant of the rabies virus is endemic in the northeastern counties of Wyoming and typically occurs throughout the Missouri River drainage system (Figure 1). A bat variant of the rabies virus, common in big brown bats (*Eptesicus fuscus*), occurs throughout the state.

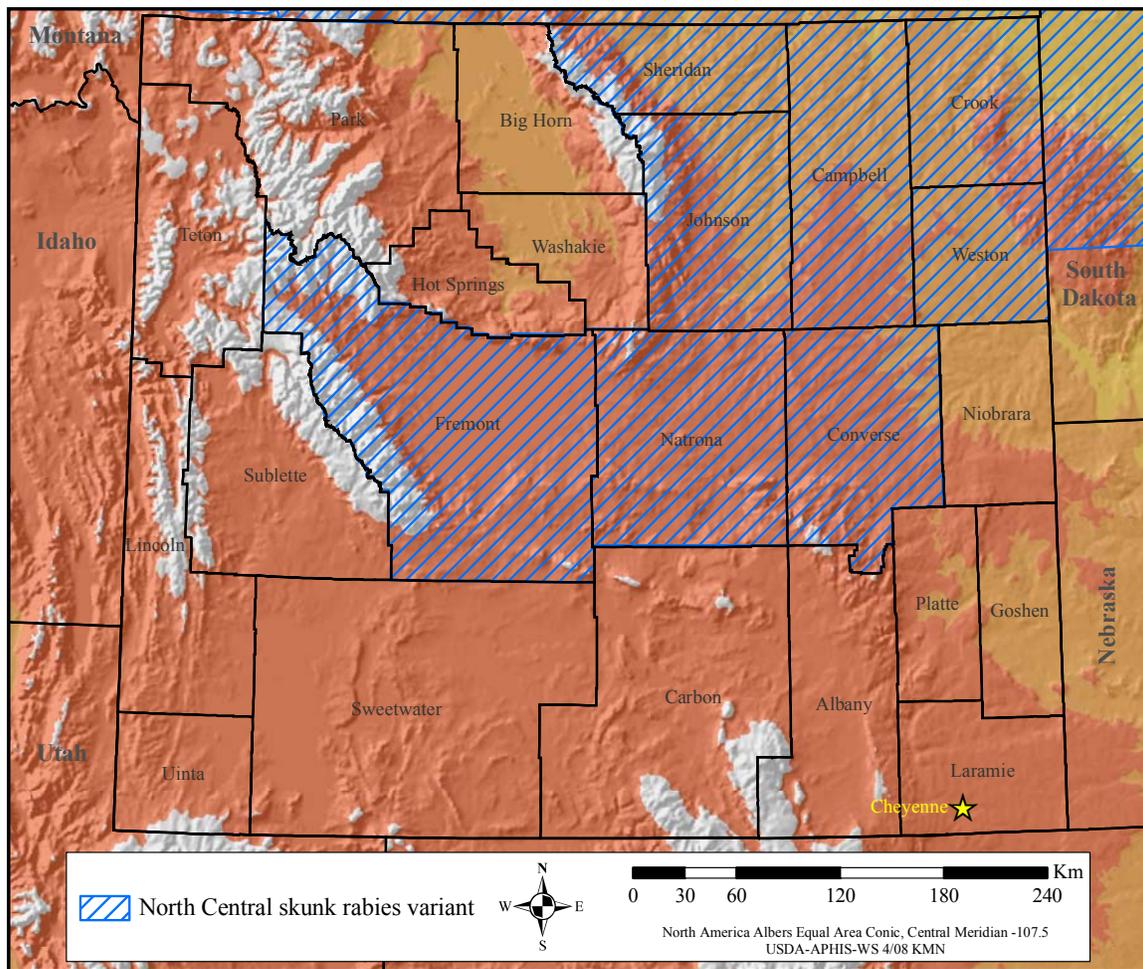


Figure 1. Wildlife Services cooperative rabies management program activities in Wyoming, 2006.

In 2002, the Wyoming Legislature directed the Wyoming Animal Damage Management Board (ADMB) to develop and implement a wildlife rabies management program. The Board, as directed by statute, was charged with promulgating rules pertaining to rabies prevention in wildlife including: surveillance; public education and prevention; vaccination protocol; post-exposure procedures and quarantines; and entering into agreements with law enforcement agencies to carry out quarantine provisions. To comply with this directive, the ADMB signed a memorandum of understanding with Wildlife Services (WS) to provide specimens (primarily skunks) from across the state to the Wyoming State Veterinary Laboratory (WSVL) in Laramie for rabies testing.

In 2003 and 2004, Wyoming WS assisted in a multi-state, long-term study coordinated by WS' National Wildlife Research Center to evaluate bait acceptance of oral rabies vaccine (ORV) placebo baits by skunks. The ultimate goal is to pair an attenuated virus vaccine with a bait matrix highly preferred by skunks for use in a broad-based ORV bait distribution program in the western U.S.

A history of rabies in northeast Wyoming and the availability of additional special funds once again enabled the Wyoming WS program to hire a seasonal (180-day) employee to conduct enhanced rabies surveillance

in Campbell County in 2006. Additional rabies surveillance was conducted in 3 counties in eastern Wyoming (Goshen, Niobrara, Platte, and Weston) as an adjunct to a coyote (*Canis latrans*) plague serosurvey funded by the ADMB in cooperation with the Wyoming Department of Health (WDH).

## **RABIES MANAGEMENT PROGRAM 2006**

### **Bait Distribution**

There is currently no ORV bait distribution program in Wyoming.

### **Enhanced Surveillance**

In 2006, WS assisted the ADMB with statewide rabies surveillance for the third consecutive year. Specimens were submitted by WS specialists and other entities (city and town governments, veterinary clinics, animal shelters, animal control officers, and public health departments). WS specialists in the state live trapped skunks from February through September in the course of performing their normal work duties. Of 254 samples (heads or carcasses) submitted by WS (144) and other entities/individuals (110), 248 were suitable for rabies testing. These included 101 skunks (not identified to species), 66 bats (not identified to species), 56 coyotes (see *Plague Survey* below), 24 raccoons (*Procyon lotor*), 4 coyotes (*Canis latrans*), 1 red fox (*Vulpes vulpes*), 1 feral cat (*Felis catus*), and 1 squirrel (not identified to species). Two bats (1 each from Natrona and Albany Counties) were the only submissions to test positive for rabies by the WSVL using the direct fluorescent antibody (dFA) test.

*Skunk Rabies Surveillance.*--The seasonal employee hired to conduct enhanced rabies surveillance targeting skunks, submitted a total of 17 skunk (not identified to species) to the WSVL for rabies testing from Campbell (n=13) and Converse (n=4) Counties. The modest number of samples he submitted is attributed to a family emergency which terminated his employment shortly after he started. None of the 17 skunks tested positive for rabies. The numbers reported here are included in the statewide totals listed in the preceding paragraph.

*Plague Seroprevalence Survey.*--In 2005, the Wyoming WS program, in cooperation with the WDH, through funding supplied by the ADMB, initiated a plague seroprevalence survey in 4 eastern Wyoming counties (Goshen, Niobrara, Platte, Weston). The justification for this project was the concern that plague is spreading in eastern Wyoming, as evidenced by a human plague case in Goshen County coupled with the detection of plague for the first time in prairie dogs in adjacent western South Dakota. The acquisition of baseline plague data is important to the WDH to better address potential health concerns which could impact its citizens. This project continued in 2006, with WS collecting and submitting 56 coyote heads from Platte (53) and Weston (3) Counties to the WSVL for rabies testing. None of these samples tested positive for rabies. This project concluded in 2006.

*Direct Rapid Immunohistochemistry Test (dRIT).*--The dRIT is an unlicensed procedure designed for consideration as a potential confirmatory measure of the dFA test (the test most frequently used to diagnose rabies). In addition, the dRIT may be used to enhance field surveillance among suspect wildlife, particularly in support of ORV programs. The dRIT may be used in remote locations to improve sample turnaround and not overburden rabies laboratories, but it is not to be used for public health surveillance. Animals involved in potential or actual rabies exposures with humans or domestic animals will continue to be processed by public health experts at established local, state, or federal laboratories.

Wyoming WS has not implemented the dRIT because the WSVL is readily meeting enhanced surveillance testing needs.

### **Rabies Laboratory Cooperation**

The WSVL in Laramie, Wyoming conducts all rabies testing on animals collected in the state. In addition to enhanced surveillance testing, the lab tests brain stems from mammals for rabies via routine public health surveillance (specimens involved in a potential or confirmed exposure). In 2006, the WSVL tested 751 animals for rabies and confirmed 11 (1.5%) rabid animals including 8 bats and 3 skunks. The positive bats originated from the following counties: Albany (1), Fremont (1), Hot Springs (1), Laramie (1), Natrona (2), Park (1) and Sheridan (1). All 3 positive skunks came from one location in Campbell County. Statewide prevalence of rabies in skunks decreased from 15 cases in 2005 (n=129 samples tested).

A continuing challenge faced by the WSVL is acquiring an adequate and representative number of surveillance samples from each Wyoming county. In 2007, the WSVL expects continued funding from the Wyoming Department of Agriculture to maintain the statewide surveillance program. For more information on rabies in Wyoming, please visit: <http://www.wyorabies.org/index.htm>

## **SUMMARY**

In 2006, WS focused on a general statewide surveillance program targeting skunks, with increased emphasis in the northeast portion of the state. Wyoming WS completed a special surveillance project in eastern Wyoming to test for rabies in coyotes, but did not conduct any ORV placebo bait studies. In 2007, WS will continue to cooperate with the ADMB and the WSVL to enhance rabies surveillance in Wyoming.

## NATIONAL WILDLIFE RESEARCH CENTER CONTROLLING WILDLIFE VECTORS OF RABIES 2006

### BACKGROUND

The National Wildlife Research Center (NWRC), located in Fort Collins, Colorado, is the research arm of Wildlife Services (WS). To assist WS' operational National Rabies Management Program (NRMP), oral rabies vaccination (ORV) program, the NWRC, Wildlife Disease Research Program (WDRP), was requested to conduct a second five year research project from FY 2006-2010. The objectives of this project are to: 1) determine the significance of demography, behavior, movements, and dispersal of raccoons (*Procyon lotor*) and striped skunks (*Mephitis mephitis*) as they may relate to the transmission and rabies virus trafficking across ecosystems; 2) develop and/or evaluate methods and technologies for use by the ORV program to increase effectiveness in vaccinating free-ranging wildlife against rabies which may reduce or eliminate the transmission of rabies from wildlife to humans, livestock, and other wildlife; 3) obtain information on the ecology of gray foxes (*Urocyon cinereoargenteus*) for possible development of improved baiting strategies for the ORV program in Texas; and 4) evaluate long-term efficacy of Raboral V-RG® (V-RG) (Merial Limited, Athens, Georgia, USA) vaccine and factors that may interfere with or reduce rabies vaccination rates in free-ranging raccoons. Several studies are designed to obtain basic information on rabies in wildlife reservoirs and vectors. Pen and laboratory studies were conducted at the NWRC and Colorado State University, while field studies were conducted in Alabama, Ohio, Pennsylvania, and Texas, all states with current ORV programs (Figure 1). Studies include research on: bait development to better deliver V-RG to raccoons and skunks; ecology of raccoons and gray foxes in rural and urban areas; better use of biomarkers to evaluate vaccine uptake by target and non-target wildlife; the evaluation of geographic barriers for wildlife dispersal that may affect the spread of rabies; and determining long-term efficacy of V-RG vaccine in raccoons.

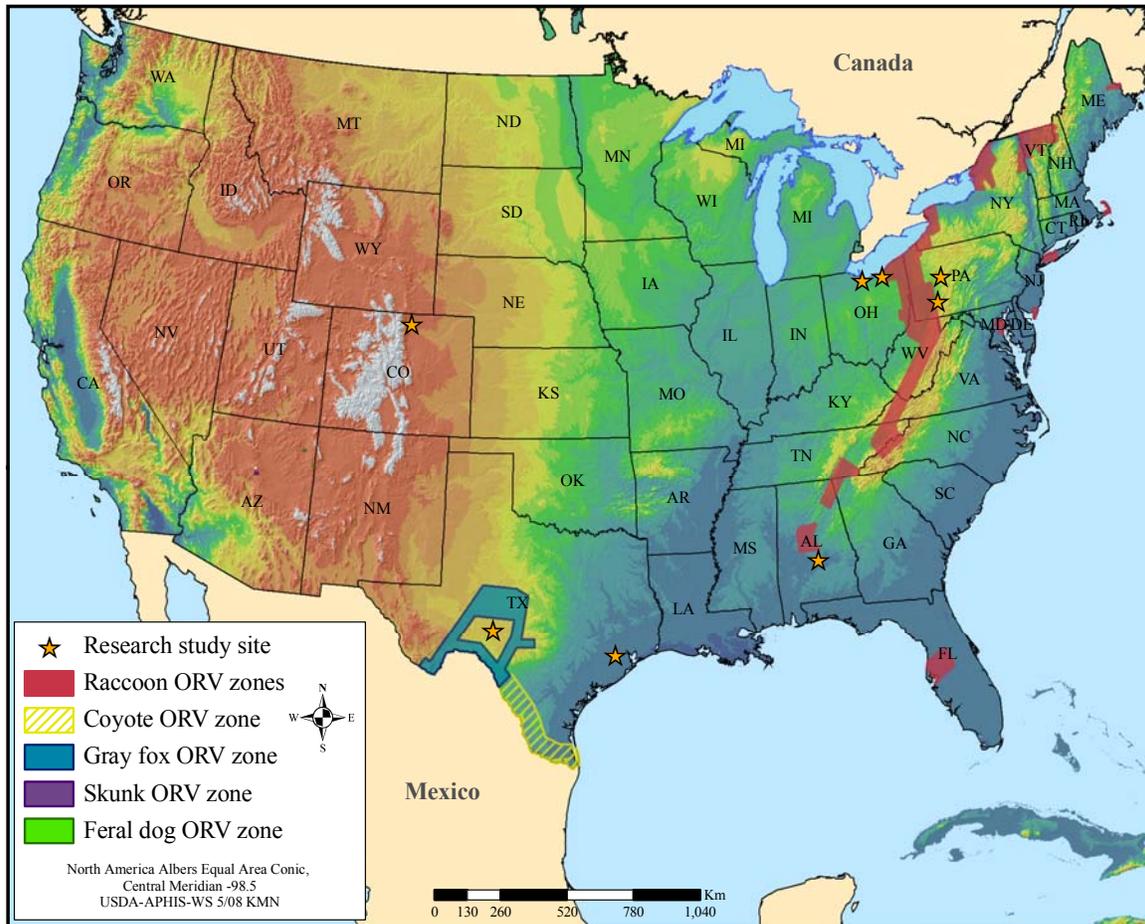


Figure 1. Wildlife Services' (WS) National Wildlife Research Center cooperative rabies research sites and WS' operational oral rabies vaccination (ORV) zones in the United States, 2006.

## RESEARCH PROGRAM 2006

### NWRC, WDRP Research Studies

*“The determination of the immunological response to an oral rabies vaccine in raccoons by characterizing the persistence of the protective antibody titer and challenge of the protection” (QA 1218).*--This study was completed in late 2006 and the graduate student is completing his MS thesis in cooperation with Colorado State University. The objectives of the study were to: 1) determine the humoral antibody response after the oral administration of 1 vs. 2 doses of V-RG; 2) determine the duration of protective antibody up to 18 months; 3) determine if 2 concurrent doses of V-RG produces more protection than 1 dose; and 4) determine if the administration of a booster of V-RG 1 year after the single dose gives more protection compared with a single dose or 2 concurrent doses. In this study antibody response was followed for 18 months and vaccinated raccoons were challenged at 6, 12, and 18 months. The V-RG vaccine is deployed in baits in attempts to immunize wild populations of raccoons and other terrestrial carnivores against the rabies virus. To better understand the duration of immunity elicited by the vaccine, groups of raccoons were immunized with different dosages of vaccine, with and without booster vaccination, and challenged with a non-passaged raccoon rabies virus at varying times post-immunization. Blood samples were collected at regular intervals after immunization and challenge for assay of rabies virus neutralizing antibodies (VNA). Initial serologic testing indicated that 70, 70, and 80% of the animals in groups A, B and C successfully generated rabies VNA by the time of challenge, and that survival rates had a high correlation with antibody production (Table 1). Only 1 of 51 animals that had produced antibodies in response to vaccination succumbed to rabies virus infection following challenge. Two-dose groups had a similar response to vaccination with a wide range of antibody production. The survival rates of this challenge period had a direct correlation with antibody production, in that every animal that produced an antibody titer survived challenge, with the 12 and 18 month groups having an 80% and 100% survivability rate.

Table 1. Summary of rabies survival in raccoons treated with Raboral V-RG® (Merial Limited, Athens, Georgia, USA) and control raccoons, National Wildlife Research Center, 2006.

Group	Number of immunizations and doses of V-RG®	Time of challenge post-vaccination	Survival of rabies challenge	
			Vaccinates	Controls
A	1 dose at 0 months	6 months	9/10 (90%)	2/2 (100%)
B	1 dose at 0 months	12 months	7/10 (70%)	1/4 (25%)
C	1 dose at 0 months	18 months	8/10 (80%)	0/3
D	2 doses at 0 months	12 months	8/10 (80%)	1/4 (25%)
E	2 doses at 0 months	18 months	10/10 (100%)	0/3
F	1 dose at 0 and 12 months	18 months	10/10 (100%)	0/3

*“Ecology of the gray fox in relation to oral rabies vaccination programs in Texas” (QA 1223).*--This study began in January 2005 and will be completed in late 2007. The objectives are to: 1) evaluate movements, dispersal, and home range of gray foxes in and near the present ORV zone in west-central Texas; 2) analyze gene flow in the fox population; 3) evaluate survival and population densities; and 4) use GIS to examine habitat use and other parameters. Monitoring locations of gray foxes are continuing on 3 study sites. Presently, >40 gray foxes have been trapped and fitted with VHF/GPS radio collars and are being monitored for locations and movements. Preliminary data indicate that male foxes move greater distances compared with females. Long distance movements by males include 13 km in Kerr County within the ORV zone and 8 km in Sutton County inside the circle of the ORV zone. More recently, additional long-distance movements of gray foxes have been noted, including 1 male that putatively moved >100 km and movements of 19 and 14 km by 2 female gray foxes. The information obtained in this study will be used in the determination of future ORV baiting strategies to prevent the spread of rabies in foxes in Texas. In addition, a population genetic analysis has been completed and a manuscript for this work is being written. These preliminary genetic analyses also indicate long distance movements occur in gray fox populations, which has obvious implications for ORV zones. Field work should be completed in late FY 07 and data analysis should be completed in early FY 08.

*“Use of genetic analyses to determine the effects of the Alabama River in Central Alabama as a natural barrier on preventing the western spread of rabies”*.--In late FY 06, the rabies project completed funding and support of a graduate student in an MS program at Auburn University who was studying raccoon movements and dispersal in relation to the Alabama River in collaboration with WS staff in Alabama and another NWRC scientist. This research determined that a few (3 of 70) of the raccoons in the study did cross the Alabama River and that the river did not constitute a complete barrier (see section under “collaborative university-based research”). Since that time, scientists at the NWRC in Ft. Collins, tested tissue samples from raccoons collected from the same 3 study sites as was used by the graduate student and conducted a genetic analysis using those samples. Preliminary results indicate that the Alabama River in central Alabama does not constitute a natural barrier to raccoon movement and dispersal. However, a further study site west of the river appears genetically separated from both study sites on either side of and adjacent to the river. This analysis is currently underway and should be completed in late 2007.

*“Movements of raccoons within the ridge-and-valley system of central Pennsylvania” (QA 1359)*.--This study was initiated in late 2006. One term biological science technician was hired to conduct the majority of the field work for this study and is stationed in Bolivar, Pennsylvania. Thus far, most raccoons have been fairly sedentary. No valley-to-valley movements have been observed, but 1 ridge-to-valley movement covering about 6 miles in straight-line distance was observed. A second set of transmitters will be deployed during the spring of 2007 with a focus on sub-adult males. In addition, a landscape genetics approach is being used to assess allelic differences among multiple populations of raccoons at 5 to 10 study sites associated with ridges and valleys. Additional genetic samples will be collected during the spring and summer of 2007 and genetic analyses will be initiated when DNA sampling is complete. Field work will be completed in FY 07 and reports will be completed in FY 08.

*“Evaluation of raccoon movements, dispersal, habitat, and population genetics to predict the probability of the westward spread of rabies in northern Ohio” (QA 1375)*.--This study was initiated in September 2006. One term biological science technician was hired to conduct the study and is stationed in Cleveland, Ohio. At present, trapping and radio-collaring of raccoons is taking place and the biologist is conducting radio telemetry. Approximately 12-15 animals are radio-collared. Many animals trapped initially were too small (about 4.4 kg) to place radio-collars on; this is due to their just coming out of their winter dens. Collection of GIS information is just beginning. Samples for a landscape genetic analysis are in the initial phase of collection. Genetic analysis of samples will be conducted in collaboration with Kansas State University. Data analysis has not yet begun. The study is on schedule and should be completed sometime in FY 08.

*“Vitrification of Raboral V-RG for Improved Vaccine Stability” (QA 1334)*.--This protocol has been developed and approved by the NWRC. The Materials Transfer Agreement (MTA) and Cooperative Research and Development Agreement (CRADA) are awaiting approval by Merial Limited pharmaceutical company who has license for the V-RG rabies vaccine and who manufactures the vaccine. This process, if proven successful, should further stabilize the current vaccine used in the ORV program which should increase vaccination rates in raccoons and other free-ranging wildlife, including gray foxes. The excellent collaboration between the PDP and the WDRP at the NWRC will allow this new and exciting process of thermal stabilization of vaccines to not only produce more stable vaccines but allow the development of more efficient oral bait delivery systems due to the dry formulation of the rabies and other vaccines.

*“The utility of Rhodamine B as a biomarker in raccoons.” (QA 1410)*.--This study, which was approved by the NWRC in 2006, will attempt to determine if Rhodamine B, a dye, can be incorporated into baits used by the ORV program and be effective and safe for use as a biomarker. The possible benefits of the use of Rhodamine B will be that it can be visualized by direct observation in raccoon whiskers under a polarized light by persons without a great deal of training, it is inexpensive, it is non-toxic to animals at doses used in the ORV program, it does not contaminate the environment with an antibiotic (e.g., tetracycline), and it is non-toxic to the vaccinia virus found in the rabies vaccine V-RG. It should prove to be a much less costly biomarker to have evaluated compared with tetracycline, presently being used, as well as have a faster turn-around time for evaluation. Studies should begin early in 2007.

*“Effects of naturally occurring orthopoxviruses on successful Raboral V-RG® vaccination in raccoons” (QA 1354)*.--This study will attempt to determine if raccoons, having been experimentally given a commercial pox virus, will develop no, or less, antibody response to a rabies vaccine which uses a pox virus (*vaccinia*). The theory

is that because the V-RG rabies vaccine currently being used in the ORV program contains *vaccinia*, antibodies to naturally occurring pox viruses will interfere with vaccination using V-RG. The study is currently being conducted. The test animals for the study have been captured and are in their quarantine phase. It is anticipated that the animals will be in testing in late 2007.

*“Investigation of serologic crossreactivity to rabies virus in wild raccoons” (QA 1377).*--This study is a spin-off from the study conducted on Plum Brook Station, Ohio, where it was inadvertently found that raccoons had a high prevalence of rabies titers (48%) in the spring, before experimental vaccination, compared with a low prevalence (8%) after field vaccination. This study will attempt to determine to which rabies variant the rabies titer was generated and why did the titer fall in the raccoon population after vaccination. The technology that will be used initially is the Ouchterlony test (a double gel diffusion test). Later, the Western Blot test will be used. This could have far-reaching implications in the ORV program if we could determine that antibodies generated by contact with certain rabies variants may interfere with antibodies generated by V-RG. It should also be noted that this phenomenon has occurred in other states and areas as well as Ohio. This study will begin as soon as Kansas State University can furnish the NWRC with various rabies antigens. The aim of this study is to answer two questions: 1) what is the source of antibody titers to rabies in raccoons residing in a non-enzootic area? and 2) why are titers higher in the spring than in the autumn even when vaccination takes place in the summer? To address these questions, two hypotheses have been formulated for testing: 1) rabies antibodies in raccoons in non-enzootic areas can occur due to crossreactivity with skunk or bat variants of rabies that are not lethal to the raccoons, but which can infect and induce an immune response in the animals; and 2) female raccoons that have been previously exposed and developed rabies VNA have a resurgence of antibody production during pregnancy in order to protect their offspring by passive transfer. This accounts for an increase in titer prevalence and frequency in the spring. This study protocol was approved in 2006 and studies will begin soon.

#### **NWRC, WDRP Collaborative University-Based Research**

*Pennsylvania State University.*--The NWRC continues to fund a PhD graduate student at Pennsylvania State University who is studying the zoogeography of raccoons in Pennsylvania by evaluating the home range, density fidelity, movement patterns, and dispersal rates of raccoons in forested and mixed-agriculture habitats. This study has reached some preliminary conclusions that raccoon movement patterns shift depending on food supply and onset of furbearer trapping season. Also, the student determined that landscape in the study area does not inhibit a raccoon's movement. This information will be applied to management recommendations that will soon be made to the NRMP concerning baiting and vaccinating raccoons for rabies in rural environments. This study is nearing completion and funding by the NWRC was terminated in late 2006. A completed report is anticipated in early 2007.

*Texas A & M University, Kingsville.*--The rabies project continues to fund and assist a PhD graduate student at Texas A & M University, Kingsville, Texas in her research study, “Behavioral ecology of striped skunks within urban and suburban areas.” The objectives of this study are to evaluate movements, habitat utilization, and seasonal food habits of skunks in urban and suburban areas of Houston, Texas and compare those with skunks in rural areas. Preliminary results show that skunks prefer short grass areas for feeding, breeding, and teaching young. Also, skunks can be born from late May until September and skunks are not active when ambient temperatures drop below 45° F. This researcher believes that distribution of rabies vaccine in the fall would be more beneficial for the program. The purpose of this study is to be able to make informed recommendations concerning a possible ORV program that might be developed for skunks if an efficacious oral rabies vaccine is developed for skunks. Data analysis is nearing completion. This study will conclude in late 2006 and results will be reported in 2007.

*Auburn University.*--The NWRC is nearing completion of funding an MS student at Auburn University who is studying raccoon movements and dispersal in relation to the Alabama River, as well as evaluating raccoon home range, habitat use, and survivorship of raccoons. Seventy of 127 raccoons captured were radio collared for telemetry. Only 3 of the 70 collared raccoons crossed the Alabama River, all of which were males. Preliminary data indicate that males appear to have larger home ranges compared with females in the same habitat type. Home range sizes ranged from a mean for females of 79.6 hectares in riverine habitat types to 396.8 hectares for males in managed forests. Survival, both annual and during the fall, did not differ between habitats for males or females. Mortality was low and hunting may have been the main cause of mortality. Data analysis is currently underway. This study will be completed in 2006 and results should be reported in FY 2007.

## SUMMARY

Scientists at the NWRC have determined that: the V-RG vaccine is efficacious at least up to 18 months following vaccination; movement of gray foxes in Texas was much greater than anticipated and foxes move outside the present ORV zone; raccoons inhabiting the ridge-and-valley system in Pennsylvania do not move between valleys; and the Alabama River in Central Alabama does not constitute a natural barrier to raccoon movement and dispersal in a westward direction. Several studies were initiated in 2006 which, when completed, should have direct benefits to the ORV program. These in-progress studies will attempt to: determine quantitatively the effect of the Alabama River as a barrier to raccoon movement and dispersal by use of sophisticated genetic analyses; determine if and how raccoon movements in Cleveland, Ohio could traffic rabies through the urban and adjacent rural areas of Ohio; develop a more stabilized V-RG vaccine by use of the vitrification process; use Rhodamine B as an alternative to tetracycline as a biomarker for use in the ORV program; determine if naturally occurring pox viruses interfere with vaccination of raccoons with V-RG; and determine why rabies antibody titers in Ohio raccoons change seasonally and possibly interfere with vaccination with V-RG. The NWRC continues to fund graduate students at three universities in an attempt to answer important questions that will eventually benefit the ORV program including better understanding of raccoon ecology in Pennsylvania and Alabama and skunk ecology in urban and rural areas of central Texas in relation to the ORV program. These accomplishments by NWRC research scientists over the past year, which were funded by the WS operational NRMP have assisted the ORV program in being more efficient in vaccinating free-ranging raccoons against rabies at reduced costs to the program. The NWRC will continue to conduct research and collaborate with others to assist the NRMP in carrying out its mission of containing and eventually eliminating terrestrial rabies from the United States.