

## Comparison of Suburban Vaccine Distribution Strategies to Control Raccoon Rabies

Authors: Boulanger, Jason R., Bigler, Laura L., Curtis, Paul D., Lein,

Donald H., and Lembo, Arthur J.

Source: Journal of Wildlife Diseases, 44(4): 1014-1023

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-44.4.1014

BioOne Complete (complete.BioOne.org) is a full-text database of 200 subscribed and open-access titles in the biological, ecological, and environmental sciences published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Complete website, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at <a href="https://www.bioone.org/terms-of-use">www.bioone.org/terms-of-use</a>.

Usage of BioOne Complete content is strictly limited to personal, educational, and non - commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

BioOne sees sustainable scholarly publishing as an inherently collaborative enterprise connecting authors, nonprofit publishers, academic institutions, research libraries, and research funders in the common goal of maximizing access to critical research.

## Comparison of Suburban Vaccine Distribution Strategies to Control Raccoon Rabies

Jason R. Boulanger, <sup>1.5</sup> Laura L. Bigler, <sup>2</sup> Paul D. Curtis, <sup>3</sup> Donald H. Lein, <sup>2</sup> and Arthur J. Lembo, Jr. <sup>4</sup> <sup>1</sup>Cornell University, College of Agriculture and Life Sciences, Department of Natural Resources, 106 Rice Hall, Ithaca, New York 14853, USA; <sup>2</sup>Cornell University, College of Veterinary Medicine, Department of Population Medicine and Diagnostic Sciences, 777 Warren Road, Ithaca, New York 14850, USA; <sup>3</sup>Cornell University, College of Agriculture and Life Sciences, Department of Natural Resources, 114 Fernow Hall, Ithaca, New York 14853, USA; <sup>4</sup>Cornell University, College of Agriculture and Life Sciences, Department of Crop and Soil Science, 1001 Bradfield Hall, Ithaca, New York 14853, USA; <sup>5</sup>Corresponding author (email: jrb69@cornell.edu)

ABSTRACT: Helicopters and hand baiting are commonly used to distribute vaccine-laden baits to help control raccoon (*Procyon lotor*) rabies in suburban landscapes, but these techniques may be labor intensive, costly, or unavailable in some areas. We tested conventional baiting strategies against polyvinyl-chloride (PVC) bait stations in Erie County (New York, USA) during July and August 2003-05. Hand, helicopter, and bait station treatments were randomly assigned to six 25-km<sup>2</sup> suburban study sites. To estimate the proportion of raccoons that ingested baits, tooth and blood samples from 954 raccoons were collected and examined for tetracycline biomarker and rabies-neutralizing antibodies, respectively. Overall, 38% (358/954) of the raccoons in Erie County tested positive for tetracycline; 16% (155/954) tested seropositive for rabies virus. Year of study significantly impacted biomarker prevalence; fewer raccoons tested positive for tetracycline in 2004. Probability of seropositivity increased with raccoon age. No statistically significant differences existed between baiting strategies and frequencies of biomarker and antibody-positive raccoons across all years combined. Thus, bait stations could be used as part of an integrated rabies control strategy.

*Key words:* Bait station, New York, oral rabies vaccination, *Procyon lotor*, PVC, rabies, raccoon, suburban landscapes.

Mid-Atlantic raccoon (*Procyon lotor*) rabies in the United States had spread to Erie County, New York, by 1992 and has become enzootic in subsequent years (Trimarchi, 1992). Suburban landscapes such as those found in Erie County often support elevated raccoon densities (Prange et al., 2003), which may increase the risk of rabies transmission in these areas (Wolf et al., 2003). An oral rabies vaccination (ORV) program was initiated in 2002 to control the raccoon variant of

rabies and thus help protect human and animal health in Erie County. As part of this ongoing control effort, polyvinyl-chloride (PVC) bait stations were compared to conventional hand baiting and helicopter distribution. To compare baiting strategies, tetracycline biomarker and rabies-virus-neutralizing antibodies (VNAs) were used to estimate the proportion of free-ranging raccoons that consumed vaccine-laden baits.

An orally administered vaccinia-rabies glycoprotein (V-RG) recombinant vaccine was demonstrated effective for immunizing raccoons (Rupprecht et al., 1986) and found safe for over 50 vertebrate species (Hanlon et al., 1998). Although still in developmental stages, ORV programs have shown promise in controlling terrestrial rabies through the mass distribution of V-RG baits across the landscape. In the United States, over 40 million vaccine baits have been distributed to mitigate the western migration of terrestrial rabies (Johnston et al., 2005), and by 2003 ORV efforts had taken place in 15 states (Rupprecht et al., 2004). In Erie County fixed-wing aircraft have been utilized to distribute baits in rural landscapes, but this system is not suitable for most suburban environments. Thus, hand baiting and helicopter distribution at similar densities have been used for ORV distribution in suburban areas. Large residential areas, however, preclude the use of hand baiting because it requires a high labor component (Johnston et al., 1988).

We tested PVC bait stations as a novel alternative to conventional suburban bait

distribution techniques. Raccoons are known to exploit concentrated food sources as they become available in the environment (Dalgish and Anderson, 1979). In suburban landscapes, raccoons readily feed on an abundance of refuse (Prange et al., 2003). Vegetable gardens, compost piles, ornamental plants, bird feeders, and pet foods are also examples of abundant food sites. Accordingly, the establishment of artificial feeding stations at defined intervals throughout the residential environment could provide a supplemental, cost-effective alternative to hand baiting and helicopter distribution. Raccoons may then obtain the vaccineladen baits contained within the bait stations at will.

Examining tooth samples for tetracycline biomarker and blood serum for rabies-neutralizing antibodies are methods used to estimate the proportion of raccoons that consume vaccine-laden baits. Tetracycline has been used extensively for monitoring bait consumption in freeranging animals (Johnston et al., 2005). Tetracycline is incorporated in the bait matrix and deposited in the teeth and bone of raccoons after bait consumption. Teeth are sectioned and viewed under ultraviolet illumination; fluorescent yellow emissions of tetracycline indicate an animal that has consumed the bait (Olson et al., 2000). Tetracycline deposits may appear as soon as 2 days after bait consumption (Hanlon et al., 1989) and may last for the life of the animal (Johnston et al., 1987). Virus-neutralizing antibodies may result from ingestion of V-RG or natural immunity. In a laboratory setting, rabies antibodies resulting from V-RG ingestion persisted in raccoons for over 6 mo (Rupprecht et al., 1986). In another study, raccoons that were captured in the wild following a raccoon rabies epizootic and held for observation remained seropositive for 2 yr (Bigler et al., 1983).

The results reported here represent 3 yr (2003–05) of data from an ongoing

ORV program in Erie County managed by the Cornell University Animal Health Diagnostic Center. Based on the evident desire to develop more effective ORV strategies, this study was conducted to determine whether bait stations could be used as a viable alternative to conventional hand baiting and helicopter distribution in a suburban landscape. Additionally, we wished to determine factors that help explain the presence of biomarker and antibody response in this highly suburbanized county.

Erie County (42°53'N: 78°47'W), located in western New York, is highly urbanized and includes the City of Buffalo. Approximately 591 km<sup>2</sup> (67%) of suburban landscape within the greater Buffalo area is suitable for suburban ORV control. Within this area, six 25-km<sup>2</sup> suburban study sites were chosen based on their relative homogeneity and quantified by human population density, housing density, and land use (Table 1). Human population and housing densities were derived from census data (United States Department of Commerce United States Census Bureau Geography Division, 2001). We determined land use using National Land Cover Dataset (Vogelmann et al., 2001), aerial digital orthophotos (United States Department of Agriculture Farm Services Agency Aerial Photography Field Office, Salt Lake City, Utah, USA), and ground observation. Overstory vegetation in our study areas was characterized by a predominance of elm (*Ulmus* spp.), red maple (Acer rubrum), and northern hardwoods (Dickenson, 1983; Alerich and Drake, 1995).

Baits consisted of a fishmeal polymer matrix (3.25 cm  $\times$  3.25 cm  $\times$  2 cm) that included 1% tetracycline biomarker (Bait-Tek Inc., Beaumont, Texas, USA). Plastic sachets containing 2 ml of RABORAL V-RG® (Merial Limited, Athens, Georgia, USA) recombinant rabies vaccine were embedded in the baits and sealed with wax.

Baits were distributed during late summer (July–August) 2003–05. Hand, heli-

Site <sup>b</sup>			Land use (%)			
	Persons/km <sup>2</sup>	Housing units/km <sup>2</sup>	For.	Res.	Ag.	
1 (43°01′N: 78°45′W)	1,038	320	28	34	35	
2 (43°01′N: 78°42′W)	870	312	27	39	33	
3 (42°53′N: 78°42′W)	1,375	559	28	58	13	
4 (42°50′N: 78°42′W)	746	283	25	37	38	
5 (42°45′N: 78°46′W)	596	232	32	33	35	
6 (42°45′N: 78°50′W)	859	353	32	47	19	

Table 1. Human population density (persons/km²), housing density (housing units/km²), and surrounding land use (%) segregated by study sites in Erie County, New York, USA.<sup>a</sup>

copter, and bait station treatments were randomly assigned to the six study sites so that each treatment was replicated twice (Fig. 1). Each site was comprised of a uniform, square grid containing 25 1-km<sup>2</sup> cells. Final bait densities were determined by dividing the total number of baits distributed by baiting treatment (50

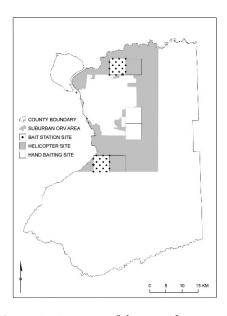


FIGURE 1. Locations of the six study sites within suburban Erie County, New York. Bait station sites depict approximate bait station distribution. \*A dense residential development and a golf course prevented placement of these two bait stations because of a lack of suitable habitat and poor station concealment.

km<sup>2</sup>). The minimum bait density goal was set at 75 baits/km<sup>2</sup> for all study sites.

Each year volunteers distributed 3,750 baits (75 baits/km<sup>2</sup>) across hand baiting sites in areas that avoided human and pet disturbance and in areas that maximized uptake by raccoons. Because of its large size, most of the greater Buffalo area was treated with a helicopter (New York State [NYS] Police Aviation Unit, Albany, New York, USA). A helicopter flew 1-km transects across study sites while broadcasting baits at a rate of 75 baits/km<sup>2</sup> in vegetated areas between housing and commercial developments. In addition, the helicopter flew along stream, railroad, highway, and power line corridors, broadcasting baits at a similar rate. Approximately 5,900 baits (118 baits/km<sup>2</sup>) were distributed across helicopter study sites. In each bait station site, 17 PVC bait stations were uniformly distributed in a staggered formation (n=34; Fig. 1) and maintained for 10 days. ArcMAP 9.1 (ESRI, Redlands, California, USA) geographic information system (GIS) software and a Garmin GPS 12XL (Olathe, Kansas, USA) global positioning system receiver were used to establish and position bait stations, respectively. All bait stations were placed in wooded areas, as close as possible to predetermined GIS locations. A dense residential development and a golf course prevented placement of two

<sup>&</sup>lt;sup>a</sup> Human population and housing densities derived from United States Census data (United States Department of Commerce United States Census Bureau Geography Division 2001). Land use determined using National Land Cover Dataset (Vogelmann et al., 2001). Ag = agricultural, For = forested, Res = residential/commercial.

<sup>&</sup>lt;sup>b</sup> Study site centroids.

bait stations because of a lack of suitable habitat and poor station concealment. Bait stations were monitored daily and refilled when necessary. Approximately 4,651 (93 baits/km²), 2,960 (59 baits/km²), and 4,269 (85 baits/km²) baits were dispensed from stations in 2003, 2004, and 2005, respectively. Design and construction of bait stations are presented in Boulanger et al. (2006).

Raccoon capture for sample collection commenced 4–5 wk postbait distribution. Tomahawk box traps (Tomahawk Live Trap Co., Tomahawk, Wisconsin, USA) baited with Fur King (Blackie's Blend, Glenmont, Ohio, USA), a commercial raccoon sweet paste, were used to trap raccoons during September-October 2003–05. Because reported home ranges for urbanized raccoons are generally less than 1 km<sup>2</sup> (Prange et al., 2004), we provided a 1-km buffer zone around each study site to help prevent cross-contamination of raccoons traveling between adjacent treatment areas. Therefore, 36 traps were restricted to the nine 1-km<sup>2</sup> grid cells located in the center of each study site; four traps (in sets of two) were placed in each 1-km<sup>2</sup> grid cell. Trap locations were selected to maximize capture rate and to avoid disturbance from people, pets, and other wildlife. If no raccoons were captured at a trap site within three nights, the two traps were moved  $\geq 100$  m within the respective cell. Traps were maintained for nine or 10 nights and subsequent trapping sessions continued until ≥100 unique raccoons were captured per treatment each year. Nontarget species, were released at the trap site each morning. We recorded the distance between the two farthest points of capture to estimate mean linear raccoon movement among recaptured individuals.

Raccoons were sedated with a 10:1 dilution of ketamine:xylazine (10 mg/kg; Phoenix Scientific, St. Joseph, Missouri, USA) and treated with Puralube® (E. Fougera & Co., Melville, New York, USA), an ophthalmic ointment used to

prevent corneal desiccation. Blood samples (10 ml) were collected from femoral blood vessels in vacutainer tubes (BD, Franklin Lakes, New Jersey, USA) for serologic assays of VNA. The first upper premolar was extracted for biomarker and cementum age analyses. All raccoons were marked with individually numbered Monel #3 ear tags (National Band and Tag Company, Newport, Kentucky, USA). Sex, relative age, and weight of each raccoon were recorded. Raccoons were released at the site of capture after a recovery period. This research conforms to the requirements of Cornell University's Institutional Animal Care and Use Committee (Protocol No. 95-79-01).

Blood samples were refrigerated (4 C) for 24 hr, and serum was separated from clotted blood. Sera were aliquoted into 2-ml skirted screw-top tubes (Laboratory Products Sales, Rochester, New York, USA) and stored in a freezer (-20 C) for subsequent testing. The New York State Department of Health Rabies Laboratory at the Wadsworth Center (Slingerlands, New York, USA) conducted an in vitro virus neutralization test to detect rabies antibodies (Trimarchi et al., 1996); the minimum level of detectable antibody titer was 0.125 IU/ml.

Matson's Laboratory (Milltown, Montana, USA) conducted tetracycline biomarker testing. A Buehler low-speed saw (Buehler Ltd., Lake Bluff, Illinois, USA) was used to cut 100-µm sections from extracted tooth samples. Sections were mounted on glass slides using glycerol and a glass coverslip and then examined with an epi-fluorescence microscope for the presence of biomarker (Matson and Kerr, 1998). At least one fluorescent yellow band in a tooth section indicated a tetracycline-positive sample.

Pearson's chi-square test was conducted to discern differences between biomarker and antibody-positive raccoons for treatment types and within individual study sites. Stepwise logistic regression was performed to determine variables that

Treatment	Tetracycline biomarker <sup>a</sup>			Titer $(\ge 0.125 \text{ IU/ml})^{\text{b}}$				
	Negative		Positive		Negative		Positive	
	%	n	%	n	%	n	%	n
Helicopter	60.0	197	40.0	131	83.0	272	17.0	57
Bait station	67.5	210	32.5	101	83.0	259	17.0	54
Hand baiting	60.0	189	40.0	126	86.0	268	14.0	44
Γotal		596		358		799		155

Table 2. Overall baiting treatment type segregated by frequencies of tetracycline biomarker and rabies-neutralizing antibody titer in Erie County, New York, 2003–05.

best explained presence of biomarker and titer. All data were analyzed using SAS 9.1 (SAS Institute Inc., 2004); alpha was set at P=0.05.

We captured 965 unique raccoons; 85 raccoons were recaptured in the last 2 yr of this study. The mean linear distance traveled between the two farthest points of capture was 0.52 km (SE=0.05). Most (n=77) raccoons were recaptured within less than 1 km from the first capture location. Six raccoons traveled between 1 and 2 km, and two raccoons traveled greater than 2 km.

Tooth sections from 958 raccoons were examined for tetracycline deposits. Four individuals were removed because of questionable biomarker results, resulting in 954 usable samples. Overall, 38% (358/954) of the raccoons in suburban Erie County tested positive for biomarker. In 2003 biomarker was detected in 35% (107/302) of sampled raccoons. Biomarker presence decreased to 32% (103/327) in 2004 but increased to 46% (148/325) in 2005 ( $\chi^2_2$ =14.46, P<0.01).

We determined the nature of the relationship between tetracycline-positive raccoons and baiting treatment. Treatment types differed marginally ( $\chi^2_{2}$ = 6.28, P=0.04) relative to biomarker frequencies in 2003; the proportion of tetracycline-positive raccoons ranged from 27% (27/101) among bait station sites to 44% (44/101) among hand baiting sites. We detected no differences between

biomarker frequencies and treatment area during 2004 ( $\chi^2_2$ =3.37, P=0.19) and 2005 ( $\chi^2_2$ =4.87, P=0.09). No difference ( $\chi^2_2$ =5.02, P=0.081) existed between treatment type and biomarker frequencies across all years combined (Table 2).

There was no difference between biomarker frequencies within individual study sites during 2003 ( $\chi^2_5$ =6.42, P=0.27) and 2005 ( $\chi^2_5$ =10.04, P=0.07). However, study sites differed ( $\chi^2_5$ =12.99, P=0.02) relative to biomarker frequencies in 2004; the proportion of tetracycline-positive raccoons ranged from 18% (13/73) in the northern hand baiting site to 45% (17/38) in the northern bait station site. Across all years combined, no difference ( $\chi^2_5$ =8.55, P=0.13) existed between individual study sites and biomarker frequencies.

Stepwise logistic regression was used to determine variables that best explained the presence of biomarker. Initial variables included treatment year, raccoon sex and age, treatment type, and individual study site. Treatment year (P < 0.01) alone best explained the presence of biomarker. The odds of biomarker prevalence in 2003 were 1.2 times the odds in 2004; however, the confidence interval (CI) included one, so the difference was not significant (OR=0.84, 95% CI [0.60-1.17]). The odds of biomarker prevalence in 2003 were 0.68 times the odds in 2005 (OR=1.48, 95% CI [1.07–2.04]). Interaction terms added no significance to the

<sup>&</sup>lt;sup>a</sup> Overall  $\chi^2_2 = 5.02$ , P = 0.08.

<sup>&</sup>lt;sup>b</sup> Overall  $\chi^2_2 = 1.57$ , P = 0.46.

model. Although the model's overall chisquare was significant at P<0.01, the area under the receiving operating characteristic (ROC; 0.56), a measure of predictive accuracy, was low (SAS Institute Inc., 2004).

Sera from 962 raccoons were examined for VNA. Eight individuals were removed because of hemolysis, resulting in 954 acceptable samples. Overall, 16% (155/954) of the raccoons in suburban Erie County tested positive for rabies-neutralizing antibody titers. In 2003, 19% (57/295) of sampled raccoons had detectable levels of antibodies. Antibody responses were 13% (43/329) in 2004 and 16% (55/330) in 2005. These differences, however, were not statistically significant ( $\chi^2_2$ =4.58, P=0.10).

During each year we detected no differences between antibody response and treatment type (2003:  $\chi^2_2$ =0.38, P=0.83; 2004:  $\chi^2_2$ =1.21, P=0.55; 2005:  $\chi^2_2$ =2.41, P=0.30) and individual study sites (2003:  $\chi^2_5$ =6.07, P=0.30; 2004:  $\chi^2_5$ =6.52, P=0.26; 2005:  $\chi^2_5$ =4.98, P=0.42). Overall, no difference existed between antibody response and treatment type ( $\chi^2_2$ =1.57, P=0.46; Table 2) and study sites ( $\chi^2_5$ =2.34, P=0.80).

We used stepwise logistic regression to determine variables that best explained the presence of VNA. Initial variables included treatment year, raccoon sex and age, individual study area, and treatment type. Raccoon age (P<0.01) was the only variable retained in the model. A yearly increase in raccoon age was associated with a 24% increase in the predicted odds of antibody response (OR=1.24, 95% CI [1.14–1.35]). Interaction terms added no significance to the model. Although the model's overall chi-square was significant at P<0.01, the area under the ROC (0.63) was low (SAS Institute Inc., 2004).

Without a formal radio-telemetry study of home ranges, the mean distance between the two farthest points of capture (Schinner and Cauley, 1974; Hoffman and Gottschang, 1977) was used to determine whether cross-contamination of raccoons traveling between adjacent treatment areas was a confounding factor. In this study only 9% (n=8) of recaptured raccoons traveled distances greater than buffer zones separating treatments (1 km). In the future increasing the distance of buffer zones may help alleviate the possibility of movement between study areas.

Despite variation in bait densities, no statistically significant differences existed between baiting strategies and frequencies of biomarker and antibody-positive raccoons across all years combined. In 2003, however, the proportion of biomarkerpositive raccoons in the bait station treatment ranked lowest when compared to conventional baiting treatments. In addition, differences existed between biomarker-positive raccoons and study site in 2004. These differences are likely the result of varying bait and raccoon densities, weather events, environmental variability, and alternative food availability in study sites. Hand baiting is also more likely to target good raccoon habitat.

Overall, 38% (358/954) of the raccoons in suburban Erie County tested positive for biomarker, which fell within the lower range of previously reported placebo and vaccine trials (30–85%; Perry et al., 1989; Hable et al., 1992; Hanlon et al., 1993; Roscoe et al., 1998; Olson et al., 2000). Overall, 16% (155/954) of the raccoons in suburban Erie County tested positive for rabies-neutralizing antibody titers, which was less than the lowest range of previously reported field trials (30–77%; Hanlon et al., 1996, 1998; Robbins et al., 1998; Roscoe et al., 1998). Direct comparisons among studies are difficult because of differences in land use, raccoon densities, tooth and bone samples used for biomarker testing, seroconversion levels, bait type, distribution method, and bait density. Despite relatively low proportions of biomarker and antibody-positive raccoons, we noted a fivefold reduction in mean terrestrial rabies cases in Erie County

after ORV distribution commenced (Boulanger et al., 2008).

Results from this study indicated that treatment year influenced biomarker rates. In 2004 the proportion of raccoons testing positive for biomarker and rabies antibody was lowest. In addition, a 10-day interval was not sufficient to achieve a minimum density of 75 baits/km² in the bait station sites during that year. These discrepancies are likely the result of heavy rains that caused flooding in the study sites in 2004, which may have impeded raccoon foraging in some areas. Alternatively, natural food availability may have been greater in 2004, compared to other years.

A yearly increase in raccoon age was associated with a 24% increase in the predicted odds of antibody response. Because results in this study are cumulative, older raccoons most likely had a greater chance of consuming VR-G or acquiring "natural" rabies immunity in this rabies endemic county during previous years. We recognize that VNA by itself may not be a suitable indicator of vaccination success; our minimum level of detectible antibody (0.125 IU/ml) was used only for treatment comparison. To our knowledge, a minimum protective level of antibody titer for successful raccoon immunization has not been defined. In addition, raccoons produce virusneutralizing antibodies in areas of rabies outbreaks (Winker and Jenkins, 1991) and in areas without prior enzootic raccoon rabies (Hill et al., 1992). We are unaware of any published accounts of background levels of rabies virus antibody in New York. However, given the rapid turnover of raccoon populations, background titers do not likely have much influence on ORV titers where rabies has been virtually eliminated because of vaccine application.

Biomarker rates were generally higher than seroconversion rates in this study. Differences between these rates may be due to different sample populations, time of consumption of bait relative to sampling, and ingestion of the bait matrix but not V-RG (Roscoe et al., 1998). Other differences may be caused by degradation of V-RG due to environmental influences, raccoon immunity, and dilution of V-RG by concurrent consumption of other food and water (Sidwa et al., 2005). The results of a recent study that investigated tetracycline stability in fishmeal polymer V-RG baits suggest that approximately 40% of the target quantity of biomarker was unavailable for absorption, possibly resulting in low biomarker detection rates (Johnston et al., 2005). In that study, the decrease in marking potential was linked to the conversion of tetracycline to epitetracycline during the manufacturing process of V-RG baits (Johnston et al., 2005).

Normally, background tetracycline in the environment from sources other than ORV efforts would not likely be an issue in our study area. Based on a study in Ontario in an area free of ORV control efforts, only 0.2% of raccoons were found to have tetracycline-like fluorescence in their teeth (Nunan et al., 1994). Reliable tetracycline dating could not be performed for our study, resulting in an inability to discern biomarker among years. Thus, the overall nature of our results may only be interpreted as cumulative.

A paucity of research addressing vaccine delivery to raccoons via bait stations precludes direct comparison among studies. Andelt and Woolley (1996) evaluated a device to deliver liquid rabies vaccine to raccoons in an urban landscape. This device, however, was limited to a single dose and needed recharging after each use. In another study, initial trials with a bait station that dispensed multiple vaccine-laden baits to raccoons were promising (Frantz, 1994). Boulanger et al. (2006) designed and tested the broad-scale-use PVC bait stations to dispense multiple V-RG baits to suburban raccoons in Erie County and quantified their use with infrared-triggered cameras. In that study bait stations were easy to assemble, lightweight, weatherproof, durable, reusable, and relatively inexpensive (\$10–15/station) and minimized nontarget acquisition of baits. Raccoons comprised 90% of all animals photographed at bait stations (Boulanger et al., 2006).

When helicopters and volunteers for hand baiting are available for bait distribution, managers specializing in ORV efforts will likely decide on rabies control methods based on cost and terrain. A benefit of hand baiting is that distribution can precisely target raccoon habitat. Some of the costs associated with hand baiting include the number of personnel, their salaries, their abilities, vehicle availability, and the suburban terrain, to name a few. Based on the salaries of technicians during the time of this study, and assuming an 8hr work day, it would have taken one technician approximately 18.75 days to bait 50 km<sup>2</sup> at an approximate cost of \$1,406. Expansive suburban landscapes benefit from helicopter distribution because large numbers of baits can be broadcast quickly, albeit at a higher cost. Costs generally associated with helicopter distribution include fuel, helicopter maintenance, two salaried pilots, one to five personnel per flight, and air-traffic control. In 2006 New York State Police helicopter baiting in Erie County cost approximately \$800/hr. State helicopters, however, are not always available for ORV control. HeloAir Inc. (Richmond, Virginia, USA), a private aviation contractor that provided ORV services on Long Island, New York, in 2006, conservatively estimated a cost of \$1,375/hr (2007 pricing) and 2 hr of flight time to bait 50 km<sup>2</sup>. In addition to cost concerns, dense housing developments may restrict helicopter bait application in areas lacking baitable green spaces. In contrast, 36 reusable bait stations at \$15/station cost approximately \$540. In addition, one technician setting up (one day) and monitoring (10 days) the stations at the time of this study cost approximately \$825. Although beyond the scope of this paper, cost-benefit analyses should be conducted to further discern differences among baiting strategies.

Bait station evaluation will continue on Long Island, New York. August 2004 marked the first reported case of raccoon rabies in Nassau County (Rudd, 2004), followed by viral spread to neighboring Suffolk County in March 2006. Information from this study, in part, has led to continued research through the broadscale implementation of modified PVC bait stations in suburban areas of New York and Massachusetts, to supplement traditional ORV control efforts during 2006 and 2007.

Suburban landscapes support elevated raccoon densities that may increase the risk of rabies transmission to people and domestic pets in these areas. Moreover, housing growth in the United States has increased in recent decades (Radeloff et al., 2005). As a result, the need for improved ORV methods in these areas will increase. Although bait stations may have potential for dispensing V-RG to raccoons, further experimentation is needed to refine their use. The effects of bait, bait station, baiting interval, and raccoon density on the proportion of vaccinated raccoons need to be determined. Bait stations appear to be a statistically comparable, less expensive tool that could be used as part of an integrated rabies control strategy.

This research was funded by New York State Department of Agriculture and Markets, United States Department of Agriculture Wildlife Services, and the Ontario Ministry of Natural Resources. We thank the New York State Police Aviation Unit, New York State Rabies Laboratory, Elephant Watch Limited, Merial Limited, Erie and Niagara County Departments of Health, AmeriCorps, New York State Department of Correctional Services, and Bippert Farms for their support and cooperation. We also thank Jodie Jarvis from the New York State Rabies Laboratory for serological analysis. Valuable field assistance was

provided by M. Antonik, J. Brennan, S. Carlin, P. Corp, S. Hauver, E. Maynard, K. Mueller, and F. Purkert. P. J. Sullivan reviewed an earlier version of this manuscript.

## LITERATURE CITED

- ALERICH, C. L., AND D. A. DRAKE. 1995. Forest statistics for New York, 1993. United States Forest Service, Northeastern Forest Experiment Station, Newtown Square, Pennsylvania, USA.
- Andelt, W. F., and T. P. Woolley. 1996. Responses of urban mammals to odor attractants and a bait dispensing device. Wildlife Society Bulletin 24: 111–118.
- BIGLER, W. J., G. L. HOFF, J. S. SMITH, R. G. McLean, H. A. Trevino, and J. Ingwersen. 1983. Persistence of rabies antibody in freeranging raccoons. Journal of Infectious Diseases 148: 610.
- Boulanger, J. R., L. L. Bigler, P. D. Curtis, D. H. Lein, and A. J. Lembo, Jr. 2006. A PVC bait station for dispensing rabies vaccine to raccoons in suburban landscapes. Wildlife Society Bulletin 34: 1206–1211.
- ——, ——, ——, AND ———. 2008. Evaluation of an oral vaccination program to control raccoon rabies in a suburbanized land-scape. Human-Wildlife Conflicts 2: 212–224.
- Dalgish, J., and S. Anderson. 1979. A field experiment on learning by raccoons. Journal of Mammalogy 60: 620–622.
- Dickenson, N. R. 1983. Physiographic zones of south and western New York. New York State Department of Environmental Conservation, Albany, New York.
- FRANTZ, S. C. 1994. Vaccine bait stations and enhanced baits—additional tools for delivering oral rabies vaccine to raccoons. *In Proceedings* of the fifth annual international meeting of rabies in the Americas, Rabies Research Unit Ontario Ministry of Natural Resources, Niagara Falls, Ontario, Canada, 16–19 November, 13 pp.
- Hable, C. P., A. N. Hamir, D. E. Snyder, R. Joyner, J. French, V. Nettles, C. Hanlon, and C. E. Rupprecht. 1992. Prerequisites for oral immunization of free-ranging raccoons (*Procyon lotor*) with a recombinant rabies virus vaccine: Study site ecology and bait system development. Journal of Wildlife Diseases 28: 64–79.
- HANLON, C. A., D. E. HAYES, A. N. HAMIR, D. E. SNYDER, S. JENKINS, C. P. HABLE, AND C. E. RUPPRECHT. 1989. Proposed field evaluation of a rabies recombinant vaccine for raccoons (*Procyon lotor*): Site selection, target species characteristics, and placebo baiting trials. Journal of Wildlife Diseases 25: 555–567.
- —, J. R. Buchanan, E. Nelson, H. S. Niu, D.

- DIEHL, AND C. E. RUPPRECHT. 1993. A vaccinia-vectored rabies vaccine field trial: Ante- and post-mortem biomarkers. Revue Scientifique et Technique de l'Office International des Epizootics 12: 99–107.
- ——, A. WILLSEY, B. LANIEWICZ, C. TRIMARCHI, AND C. E. RUPPRECHT. 1996. New York State oral wildlife rabies vaccination: first evaluation for enzootic raccoon rabies control. *In* Proceedings of the seventh annual international meeting on advances towards rabies control in the Americas, Centers for Disease Control and Prevention, Atlanta, Georgia, 9–13 December, 26 pp.
- ——, M. Niezgoda, A. N. Hamir, C. Schumacher, H. Koprowski, and C. E. Rupprecht. 1998. First North American field release of a vaccinia-rabies glycoprotein recombinant virus. Journal of Wildlife Diseases 34: 228–239.
- HILL, R. E., G. W. BERAN, AND W. R. CLARK. 1992. Demonstration of rabies virus-specific antibody in the sera of free-ranging Iowa raccoons (*Procyon lotor*). Journal of Wildlife Diseases 28: 377–385.
- HOFFMANN, C. O., AND J. L. GOTTSCHANG. 1977. Numbers, distribution, and movements of a raccoon population in a suburban residential community. Journal of Mammalogy 58: 623–636.
- JOHNSTON, D. H., D. G. JOACHIM, P. BACHMANN, K. V. KARDONG, R. E. A. STEWART, L. M. DIX, M. A. STRICKLAND, AND I. D. WATT. 1987. Aging furbearers using tooth structure and biomarkers. In Wild furbearer management and conservation in North America, M. Novak, J. A. Baker, M. E. Obbard, and B. Malloch (eds.). Ontario Trappers Association, North Bay, Ontario, Canada. pp. 228–243.
- , D. R. Voict, C. D. Macinnes, P. Bachmann, K. F. Lawson, and C. E. Rupprecht. 1988. An aerial baiting system for the distribution of attenuated or recombinant rabies vaccines for foxes, raccoons, and skunks. Review of Infectious Diseases 10: 660–664.
- JOHNSTON, J. J., T. M. PRIMUS, T. BUETTGENBACH, C. A. FURCOLOW, M. J. GOODALL, D. SLATE, R. B. CHIPMAN, J. L. SNOW, AND T. J. DELIBERTO. 2005. Evaluation and significance of tetracycline stability in rabies vaccine baits. Journal of Wildlife Diseases 41: 549–558.
- Matson, G. M., and K. D. Kerr. 1998. A method for dating tetracycline biomarkers in black bear cementum. Ursus 10: 455–458.
- Nunan, C. P., C. D. Macinnes, P. Bachmann, D. H. Johnston, and I. D. Watt. 1994. Background prevalence of tetracycline-like fluorescence in teeth of free ranging red foxes (Vulpes vulpes), striped skunks (Mephitis mephitis), and raccoons (Procyon lotor) in Ontario, Canada. Journal of Wildlife Diseases 30: 112–114.
- Olson, C. A., K. D. MITCHELL, AND P. A. WERNER. 2000. Bait ingestion by free-ranging raccoons

- and nontarget species in an oral rabies vaccine field trial in Florida. Journal of Wildlife Diseases 36: 734–743.
- Perry, B. D., N. Garner, S. R. Jenkins, K. McCloskey, and D. H. Johnston. 1989. A study of techniques for the distribution of oral rabies vaccine to wild raccoon populations. Journal of Wildlife Diseases 25: 206–217.
- PRANGE, S., S. D. GEHRT, AND E. P. WIGGERS. 2003. Demographic factors contributing to high raccoon densities in urban landscapes. Journal of Wildlife Management 67: 324–333.
- ———, AND ———. 2004. Influences of anthropogenic resources on raccoon (*Procyon lotor*) movements and spatial distribution. Journal of Mammalogy 85: 483–490.
- RADELOFF, V. C., R. B. HAMMER, S. I. STEWART, J. S. FRIED, S. S. HOLCOMB, AND J. F. McKeefry. 2005. The wildland-urban interface in the United States. Ecological Applications 15: 799–805.
- Robbins, A. H., M. D. Borden, B. S. Windmiller, M. Niezgoda, L. C. Marcus, S. M. O'Brien, S. M. Kreindel, M. W. McGuill, A. Demaria, C. E. Rupprecht, and S. Rowell. 1998. Prevention of the spread of rabies to wildlife by oral vaccination of raccoons in Massachusetts. Journal of the American Veterinary Medical Association 213: 1407–1412.
- Roscoe, D. E., W. C. Holste, F. E. Sorhage, C. C. Campbell, M. Niezgoda, R. Buchannan, D. Diehl, H. S. Nlu, and C. E. Rupprecht. 1998. Efficacy of an oral vaccinia-rabies glycoprotein recombinant vaccine in controlling epidemic raccoon rabies in New Jersey. Journal of Wildlife Diseases 34: 752–763.
- Rudd, R. J. 2004. 2004 rabies annual report. New York State Department of Health, Wadsworth Laboratory, Albany, New York.
- RUPPRECHT, C. E., T. J. WIKTOR, D. H. JOHNSTON, A. N. HAMIR, B. DIETZSCHOLD, W. H. WUNNER, L. T. GLICKMAN, AND H. KOPROWSKI. 1986. Oral immunization and protection of raccoons (*Procyon lotor*) with a vaccinia-rabies glycoprotein recombinant virus vaccine. Proceedings of the National Academy of Science 83: 7947–7950.
- ———, C. A. HANLON, AND D. SLATE. 2004. Oral vaccination of wildlife against rabies: Opportunities and challenges in prevention and control. Developmental Biology Basel 119: 173–184.

- SAS Institute Inc. 2004. Statistical software. Version 9.1. SAS Institute Inc., Cary, North Carolina.
- Schinner, J. R., and D. L. Cauley. 1974. The ecology of urban raccoons in Cincinnati, Ohio. *In* Wildlife in an urbanizing environment, J. H. Noyes and D. R. Progulske (eds.). Planning and resource development series number 28, Holdsworth Natural Resources Center, Amherst, Massachusetts, pp. 125–130.
- SIDWA, J. T., P. J. WILSON, G. M. MOORE, E. H. OERTLI, B. N. HICKS, R. E. ROHDE, AND D. H. JOHNSTON. 2005. Evaluation of oral rabies vaccination programs for control of rabies epizootics in coyotes and gray foxes: 1995–2003. Journal of the American Veterinary Medical Association 227: 785–792.
- Trimarchi, C. V. 1992. 1992 rabies annual report. New York State Department of Health, Wadsworth Laboratory, Albany, New York, USA.
- ——, R. D. Rudd, and M. Safford, Jr. 1996. An in vitro virus neutralization test for rabies antibody. In Laboratory techniques in rabies, F. X. Meslin, M. M. Kaplan, and H. Koprowski (eds.). World Health Organization, Geneva, Switzerland. pp. 193–199.
- United States Department of Commerce United States Census Bureau Geography Division. 2001. Census blocks, New York State (Shapefile: 2001). United States Department of Commerce United States Census Bureau Geography Division, Washington, DC.
- VOGELMANN, J. E., S. M. HOWARD, L. YANG, C. R. LARSON, B. K. WYLIE, AND N. VAN DRIEL. 2001. Completion of the 1990s National Land Cover Data Set for the conterminous United States from Landsat Thematic Mapper data and ancillary data sources. Photogrammetric Engineering and Remote Sensing 67: 650–652.
- WINKLER, W. G., AND S. R. JENKINS. 1991. Raccoon rabies. *In* Natural history of rabies. CRC Press, Boca Raton, Florida. pp. 325–340.
- Wolf, K. N., F. Elvinger, and J. L. Pilcicki. 2003. Infrared-triggered photography and tracking plates to monitor oral rabies vaccine bait contact by raccoons in culverts. Wildlife Society Bulletin 31: 387–391.

Received for publication 28 February 2007.