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## Population Monitoring in Support of a Rabies Vaccination Program for Skunks in Arizona

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ABSTRACT: Three population monitoring methods were evaluated in support of a trap/vaccinate/release program for controlling a bat variant of rabies virus in skunks (Mephitis mephitis) in Flagstaff, Arizona (USA). Skunks were the primary species targeted for population monitoring during the program, but feral cats were also monitored as they represented an abundant secondary vector species capable of rabies transmission. Skunks were vaccinated and released, except for a subset tested for rabies. All captured cats were placed in the local animal shelter. Spotlight surveys essentially did not detect skunks, and were not able to detect reductions in the cat population. Catch-per-unit-effort marginally tracked population trends, but a passive track index adapted for an urban setting was most sensitive for detecting changes in skunk and cat populations. Mark-recapture population estimates could not be validly calculated from the data on captures and recaptures due to multiple violations of analytical assumptions.

*Key words:* Catch rate, mark-recapture, passive track index, population index, population monitoring, spotlight index, trap/vaccinate/ release.

A trap, vaccinate, and release (TVR) program based on Rosatte et al. (1992) was conducted in Flagstaff, Arizona (USA) in response to an outbreak of a bat variant of rabies virus in striped skunks (Mephitis mephitis) during early 2001 (Christensen and Bergman, 2001; Smith et al., 2001). This TVR program offered an opportunity to evaluate population indexing methods (e.g., Caughley, 1977) that might be practical for similar TVR programs, while providing valid quantitative results. Such an index could provide valuable information on relative population abundances, population changes, and the spatial distribution of the target animals, as well as the same population information for species co-occupying the TVR area that might impact the TVR program. An inherent difficulty in Flagstaff was that most wildlife monitoring methods were not designed for application in an urban setting. We describe the results from testing three potential monitoring methods that could be used in conjunction with a similar urban TVR program.

The TVR program was focused on the south side of Flagstaff where the rabid skunks were found (south of Interstate 40, Fig. 1). The habitat prior to human development would have been Rocky Mountain (petran)-montane conifer forest (Brown, 1994) dominated by stands of ponderosa pine (*Pinus ponderosa*). Today, the habitat in the area varies considerably. Much of the area has been developed into a typical urban/suburban setting, with single family homes on adjacent lots having manicured lawns and yards. Condominium and apartment complexes are also present, as are shopping areas and a golf course. Parts of the area also have homes placed in the surrounding ponderosa pine forest with natural, rather than manicured lots. Remnant patches of pine forest also are dispersed through the area.

Skunks were live-trapped, vaccinated (Imrab3, Merial Ltd., Athens, Georgia, USA), uniquely marked with ear tags, and released. A random subset of 19 skunks was tested for rabies (fluorescent antibody test on brainstem). Raccoons (*Procyon lotor*) and grey fox (*Urocyon cinereoargenteus*) were also marked, vaccinated, and released. Feral cats were removed from the population by placing them in the local animal shelter.

Skunks, as the primary rabies vector, were the main target animals for monitoring. All identified rabid animals were

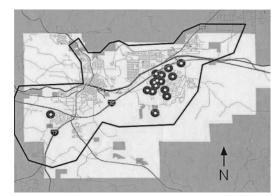


FIGURE 1. Map showing the area of a trap-vaccinate-release program for skunks in Flagstaff, Arizona.

striped skunks. Feral cats represented an abundant secondary species capable of carrying rabies. They also were an abundant animal that could reduce capture rates for skunks through occupation of traps.

Three population indexing methods were considered: catch rate, spotlight surveys, and a passive track index (PTI). All population monitoring methods were applied throughout the TVR area.

Catch per unit effort has long been used as an index for animal abundance (e.g., Caughley, 1977), and these data were available as part of the TVR program. Trapping was carried out in three 10-day sessions, which we identify as: early May, late May/early June, and mid-June. Captures were made using Tomahawk live traps (Tomahawk, Wisconsin, USA). The catch rate index was calculated at the end of each of the three sessions as the number of captures of each species during that session divided by the number of available trap nights (TN) during the session (each session exceeded 1900 TN).

Spotlight surveys were conducted at the end of each week during the 6 wk TVR program. Ten 1.6 km transects were established in the TVR area. Each transect was at least 1 km from any portion of the other transects. Surveys began 1 hr after sunset. Vehicles were driven  $\leq 16$  km/hr. Spotlight observations were made from only one side of the roads and remained constant through all surveys. The weather was mild and clear for all surveys. Numbers of each species observed along each transect were recorded each week. The spotlight index was calculated for each species each week as the mean number observed per transect.

The PTI was applied immediately prior to and after the TVR program. The PTI was based on the methods in Engeman et al. (2000, 2001a, 2001b). However, in each of those applications tracking plots were placed on dirt roads because they were used as travel pathways by target animals. The large majority of roads in the TVR area were paved, and dirt roads in the area received heavy traffic, making tracking plots on roads futile. Engeman et al. (2003) demonstrated in a much different setting that animals could be monitored using tracking plots without dirt roads, if their routes of travel could be predicted. We identified alternative potential skunk travel corridors in this urban setting. We stationed 22 plots at sites such as culvert entrances, natural draws, and openings in fences.

In contrast to many tracking plot methods, observations recorded at each plot were not binary (presence/absence). Rather, the number of track sets (number of intrusions into the plot) by skunks and cats were recorded for three consecutive days at both assessments. The number of plot intrusions has been well-documented to provide superior sensitivity to differences or changes in index levels over binary measures (Allen et al., 1996; Engeman et al., 2000). The substrate at all plot sites provided an excellent tracking surface for identifying species and distinguishing the number of intrusions. After 24 hr, plots were examined for spoor and resurfaced (tracks erased and surface smoothed) for the next day's observations. Fair weather conditions prevailed during each of the assessments. The number of sets of tracks found on the ith plot on the jth day, x<sub>ii</sub>, were represented as a linear model:

 $x_{ij} = \mu + P_i + D_j + e_{ij}$ , where the term  $\mu$  is the overall mean number of sets of tracks per plot per day for the area being assessed.  $D_i$  is a random effect due to the day on which an observation was made, with j=1, 2, or 3 in our case.  $P_i$  is a random effect due to the ith plot with  $i=1,2,3...p_i \le 22$ representing the number of plots contributing data on the jth day. The  $e_{ij}$  represent random error associated with each plot each day. Neither the plots nor the days were assumed to be independent for calculation of estimates (variance calculations were based on a nonzero covariance structure among plots and among days). The number of plots contributing data for the calculations was allowed to differ between days. This data structure permitted calculation of a passive tracking index (PTI), components of variance, and variance estimates using the methods in Engeman et al. (1998) with, the PTI defined mathematically as:

$$PTI = \frac{1}{3} \sum_{j=1}^{3} \frac{1}{p_j} \sum_{i=1}^{p_j} x_{ij}.$$

and its variance estimate calculated according to the following formula:

$$\operatorname{var}(PTI) = \frac{\sigma_p^2}{3} \sum_{j=1}^3 \frac{1}{p_j} + \frac{\sigma_d^2}{3} + \frac{\sigma_e^2}{3^2} \sum_{j=1}^3 \frac{1}{p_j}$$

where the  $\sigma_{\rm p}^2$ ,  $\sigma_{\rm d}^2$ , and  $\sigma_{\rm e}^2$  are, respectively, the variance components (Searle et al., 1992) for plot-to-plot variability, daily variability, and random observational variability associated with each plot each day. The procedure SAS PROC VARCOMP, with a restricted maximum likelihood estimation procedure (REML) (SAS Institute, 1996) was used to calculate these variance components.

The TVR program allowed the sensitivities of the monitoring methods to be evaluated, because all cats and 19 skunks were removed from the populations. There were 174 skunk captures over the 6 wk TVR program, representing 133 individuals with 41 recaptures. Nineteen striped skunks were removed from the population through rabies testing (all negative) and

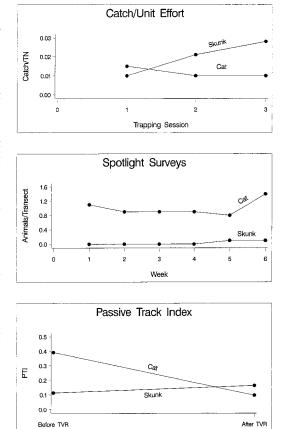


FIGURE 2. The change in index values for skunks and cats over the course of a 6 wk rabies trap/vaccinate/release program using three population monitoring methods.

Before TVR

114 were vaccinated, tagged, and released. A total of 76 cats were captured through the TVR program and removed from the habitat. No other species were captured in sufficient quantity to merit additional monitoring.

Figure 2 summarizes results from the three monitoring methods. Even though the TVR program removed 19 skunks for rabies testing, the catch rate for skunks increased across the three trapping sessions of the TVR program. Catch rate seemed to detect the reduction of cats in the area, decreasing from 0.015 to 0.010 cats/TN over the course of the TVR. Spotlight surveys rarely detected skunks, as only two sightings were made during the total 60 transect runs. Cats were readily spotted,

but the spotlight surveys indicated a stable, possibly increasing cat population at the same time the TVR program was removing 76 cats. The PTI for skunks resulted in a post-TVR index similar to that of the pre-TVR (Z=0.56, P=0.58), while the post-TVR for cats was substantially less than the pre-TVR (Z=2.05, P=0.04). Examination of the components of variance used in calculating the variance estimate for the PTI revealed that the plot effect comprised a much greater proportion of the total variability than the day effect, implying that emphasis should be placed on maximizing the number of plots for observation to achieve greatest sensitivity in future surveys. For our situation, total effort could be held constant by reducing the number of observation days to two, while increasing the number of plots.

The catch rate index for skunks, while relatively low both pre- and post-TVR, showed an increase. These results indicated increased skunk activity, likely due to greater foraging ranges for the adults as young of the year matured during the TVR period. The catch rate showed a small decrease in cat capture rates, but remained constant through the final two trapping sessions, implying that this index was not particularly sensitive to the removal of 76 cats.

Spotlight surveys did not appear useful for skunks or cats, but for different reasons. Spotlight surveys were ineffective at detecting skunks, therefore providing no potential to detect population changes or differences. The spotlight survey did not indicate an overall decrease in cat numbers, and the final survey had the highest index of all, which is a result contradictory to the removal of 76 cats.

The results for the PTI also fell in line with possible increases in skunk activity, as the index increased slightly. The PTI was the most sensitive of the three to removal of the cats and showed a steep decline. Each of the three monitoring methods held the potential to provide information on spatial distributions and abundances of animals, but the PTI provided the most logical results for both species across the TVR program. For future TVR efforts in similar urban circumstances, the PTI appears to provide a sensitive addition to the population monitoring methods.

Even though skunks were being captured, tagged, and released, we did not use mark-recapture methods (e.g., Otis et al., 1978) to estimate the initial population available for vaccination. Mark-recapture population estimates are predicated on a set of assumptions that when violated nullify the validity of the resulting estimate (Otis et al., 1978; Liedloff, 2000). Over the course of the 6 wk program an assumption that the skunk population was closed would be presumptuous, because there were no barriers to emigration or immigration and rabies, a fatal disease, was present in the population. In addition, the skunk population demography available for trapping likely changed during the TVR as juveniles entered the population. Therefore, the same mark-recapture model would have been unlikely to apply throughout the course of the TVR. Lastly, our recapture data made it clear that heterogeneity existed in individual skunk capture probabilities, because some individuals were readily recaptured while most were never recaptured. These issues nullify assumptions required for mark-recapture estimates.

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