

NON-INVASIVE METHODS TO ASSESS CO-OCCURRENCE OF
MAMMALIAN CARNIVORES

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ABSTRACT—During 2003–2005, I surveyed 82 sites on the North and South rims of Grand Canyon, Coconino County, Arizona, to test effectiveness of four non-invasive techniques for detecting carnivores and to assess patterns of co-occurrence among pairs of species. Techniques were not equally effective for detecting carnivores. Searches of transects for feces, tracks, and other evidence yielded the greatest number of detections; remotely triggered cameras and track plates had the greatest probabilities of detecting common species and also produced detections of smaller and rarer carnivores; and hair traps generally were ineffective. Even after accounting for variation in probabilities of use of habitats by species due to characteristics of sites, two pairs of carnivores had limited co-occurrence. Coyotes (*Canis latrans*) and bobcats (*Lynx rufus*) did not co-occur at sites on the North Rim, and coyotes and gray foxes (*Urocyon cinereoargenteus*) co-occurred less than one-half as frequently as expected in the South Rim.

RESUMEN—Entre 2003 y 2005, muestreé 82 sitios en los bordes norte y sur del Gran Canyon, condado de Coconino, Arizona, para probar la eficacia de cuatro técnicas no-invasivas para detectar carnívoros mamíferos, y evaluar los patrones de coocurrencia entre pares de especies. Las técnicas no fueron igualmente efectivas para detectar carnívoros. Búsquedas de transectos para excrementos, huellas y otra evidencia produjeron el número mayor de detecciones; cámaras de disparador-remoto y superficies preparadas para registrar huellas tuvieron la mayor probabilidad de detección de especies comunes y también produjeron detecciones de carnívoros pequeños y raros; y trampas de pelo generalmente fueron inefectivas. Aún después de tener en cuenta la variación en la probabilidad del uso de hábitat de especies debido a características del sitio, dos pares de carnívoros tuvieron coocurrencias limitadas. Coyotes (*Canis latrans*) y gatos montés (*Lynx rufus*) no coocurrieron en sitios en el borde norte, y coyotes y zorros grises (*Urocyon cinereoargenteus*) coocurrieron con frecuencia menos de la mitad de lo esperado en el borde sur.

Co-occurrence of species is important to consider when surveying for and modeling distributions of mammalian carnivores. The majority of models used to predict distributions of species are developed solely using characteristics of habitats and for one species at a time (Scott et al., 2002). However, biotic interactions in communities may influence results of field surveys and predictive models should consider the likelihood that detecting one species depends on presence of another (MacKenzie et al., 2006). Prior research on diets (Fedriani et al., 2000; Neale and Sacks, 2001a; Carvalho and Gomes, 2004) and movements of carnivores (Fedriani et al., 1999; Neale and Sacks, 2001b; Constible et al., 2006) indicates limited co-

existence and partitioning of resources among species, especially at fine spatial scales (Scognamiglio et al., 2003).

Researchers have employed several approaches to examine co-occurrence of carnivores, including analysis of use of space by radiocollared individuals (Neale and Sacks, 2001b), comparison of probability derived from predictive-occurrence models (Alexander et al., 2006), and null-model analysis of presence-absence matrices (Gotelli, 2000). Null models are used to generate random patterns of composition and distribution of species that would be expected in absence of interactions among species. These randomized distributions can then be compared with empirical data to assess how likely the observed patterns

of co-occurrence are, given hypothetical structuring mechanisms such as competition. However, an important limitation of many analyses of null models is accounting for the possibility that a species was detected imperfectly (Cam et al., 2000; MacKenzie et al., 2004).

In this study, I combined non-invasive surveys with occupancy analysis to examine patterns of co-occurrence among mammalian carnivores. Non-invasive surveying methods, which do not require animals to be directly observed or captured, are especially useful for detecting wide-ranging and elusive carnivores (Long et al., 2007a). Objectives of my research were to test relative effectiveness of four non-invasive techniques for detecting mammalian carnivores and to assess patterns of co-occurrence among pairs of species.

MATERIALS AND METHODS—I conducted this research on the North (36°18'N, 112°8'W) and South rims (36°1'N, 112°5'W) of Grand Canyon, Grand Canyon National Park, Coconino County, Arizona. The North Rim is on the Kaibab Plateau and vegetation includes ponderosa pine (*Pinus ponderosa*) and mixed conifer forests dominated by Douglas fir (*Pseudotsuga menziesii*) and white fir (*Abies concolor*), and it is the largest continuous unharvested and ungrazed forested ecosystem in Arizona (Fulé et al., 2002). The South Rim is on the Coconino Plateau. Main biotic communities are ponderosa pine forests and woodlands dominated by Colorado pinyons (*Pinus edulis*) and junipers (*Juniperus*); these have not been harvested or grazed for >100 years. Grand Canyon National Park is a popular destination in the United States and both rims have developed areas that support visitors and residents. The developmental footprint is small relative to total area of the park, and most visitors stay in areas away from study sites.

Up to 11 species of mammalian carnivores occur in forested ecosystems of Grand Canyon National Park: cougar *Puma concolor*, American black bear *Ursus americanus*, coyote *Canis latrans*, bobcat *Lynx rufus*, gray fox *Urocyon cinereoargenteus*, American badger *Taxidea taxus*, raccoon *Procyon lotor*, striped skunk *Mephitis mephitis*, western spotted skunk *Spilogale gracilis*, ringtail *Bassariscus astutus*, and long-tailed weasel *Mustela frenata*. Three species of large carnivores have been extirpated from the park; brown bears (*Ursus arctos*) and jaguars (*Panthera onca*) disappeared by the early 20th century (Hoffmeister, 1971), and the last observation of gray wolves (*Canis lupus*) was in 1935.

I surveyed sites for 2 seasons in each study area; 2003–2004 on the North Rim and 2004–2005 on the South Rim. Study areas were selected to minimize variation in habitats among monitoring sites and, thereby, increase likelihood of separating potential interspecific effects from underlying relationships of habitat. On the North Rim, I monitored 20 sites in ponderosa pine forests during each season. On the South Rim, sites were in ponderosa pine forests or

pinyon-juniper woodlands. I monitored 18 sites during the first season (2004) and 24 sites during the second (2005). Sites were selected using a geographic-information-system (GIS) database in ArcGIS 9.0 (Environmental Systems Research Institute, Redlands, California). I did not consider roads, trails, sources of water, boundary of park, or other variables when choosing sites. Sites were selected based on distance between them; sites were ≥ 2 km apart and ≥ 2 km from edges of developed areas. I selected a buffer distance of 2 km to ensure that the area between sites would exceed the average size of home range for all target species except cougars (Crooks, 2002) and, thereby, minimize the chance that the same individual would be detected at multiple sites.

Monitoring sites were 4 ha in area (200 by 200 m). I used four non-invasive techniques to maximize likelihood of detecting all target species at each site: surveys along transects, remotely triggered cameras, hair traps, and track plates. Effort was allocated to a greater number of sites to balance limitations of equipment and staff (i.e., 12 cameras, 20 track plates, and 1–2 field technicians) with low rates of occurrence of carnivores (MacKenzie and Royle, 2005). The goal was to identify the optimal suite of methods and minimal effort that would be required for repeatable, long-term monitoring (Long et al., 2007c). I surveyed sites three times during each field season (June–September), when remote regions were accessible by vehicle. Sampling interval between visits to sites was ca. 14 days.

I established four 200-m line transects, spaced 50 m apart through the interior of each site. One observer walked slowly along each transect, searching for scats (feces), tracks, burrows, or other evidence of carnivores. All evidence was measured, photographed, and identified to species based on size and visual characteristics (Murie, 1975; Halfpenny, 1986). When a scat was detected, it was collected and stored in a paper bag. I repeated surveys along transects during each of the 3 monitoring visits/season.

I installed one remotely triggered, passive-infrared, still camera (WildlifePro, Jackson, Mississippi; VanCam, Bakersfield, California; DeerCam, Park Falls, Wisconsin) in one quadrat of each monitoring site. Cameras were attached to trees 30–45 cm above the ground, facing a clearing, and stations were baited with a fresh pork rib (2003) or chicken leg (2004–2005). Although using food as bait is debated among researchers (Long et al., 2007a), the objective of my survey was to maximize probability of detecting multiple species rather than to obtain an index of abundance. Additionally, bait was secured to a stake buried firmly in the ground 3–5 m in front of the camera, reducing the probability that it would be removed. Cameras were loaded with 36-exposure 35-mm film and set to triggering intervals of 1.5–3 min and for monitoring at night to minimize detections of birds and other non-target species. In 2003, camera stations were established for two sampling intervals. In subsequent years, because total number of cameras was limited, cameras were installed during only one sampling interval at each site.

In 2003, a line of five hair-trap stations was installed, spaced 100 m apart through the center of each site. In subsequent years, one hair-trap station was established

in a second quadrat of the site. Hair-trap stations included a visual attractant (aluminum pie tin suspended from a branch on a swivel hook) and a padded hair snag (Silver Cloud Associates, Libby, Montana) or a square piece of coarse-fiber door mat. The hair trap was attached to a tree 40–50 cm above the ground and within 1–3 m of the visual attractant. The hair trap and visual attractant were baited with a non-commercial scented lure (1:1:8 ratio of propylene glycol, glycerine, and beaver castoreum, plus several drops of catnip oil and cougar or bobcat urine). I established hair-trap stations during the first monitoring visit and checked them for hair during the two subsequent visits. If hair was present, I collected the whole trap and replaced it with a new trap. If no hair was present, I rebaited the hair trap with scent lure.

In 2004, I added a track-plate station to a third quadrat of each monitoring site in an effort to better detect smaller carnivores. Track-plate stations were constructed following specifications of Zielinski (1995). A 10 by 30-cm piece of Con-Tact shelf-liner paper (Pliant, Schaumburg, Illinois) was attached, adhesive side out, to the center of each 10 by 100-cm aluminum track plate, and a mixture of carpenter chalk (IRWIN, DeWitt, Nebraska) and ethanol was poured on the ends of the track plate and allowed to evaporate. A can of tuna-flavored cat food was attached to the center of the contact paper and the track plate was placed inside a plywood box with a 30 by 30-cm opening at each end. I established track-plate stations during the first monitoring visit and checked them for tracks during the two subsequent visits. I collected and replaced the shelf paper during each visit and rebaited the station. I measured each discernable track from track-plate stations using dial calipers. I then matched diagnostic measurements to a published key to species (Taylor and Raphael, 1988).

On the North Rim, scats collected in 2003 were allowed to dry naturally and stored in paper bags at room temperature. Scats collected in 2004 were stored in glass screw-top tubes containing 96% ethanol. On the South Rim, scats were stored in 96% ethanol at -9.4°C . At the end of each monitoring season, scats were dried and subsampled (ca. 500 mg) into 1.5-ml centrifuge tubes. Additionally, individual hairs were removed from hair traps and divided into replicate subsamples of 3–4 hairs each. Both scats and hairs were stored at -20°C pending genetic analysis.

During July–August 2006, I extracted and amplified DNA from all samples of scats and hairs. DNA was extracted from scats using QIAamp DNA Stool extraction kits (Qiagen, Inc., Valencia, California). DNA was extracted from hair by boiling samples in a solution of $1 \times \text{TE}$ buffer. Preliminary tests indicated that this method was more effective for extracting DNA from hair than commercially available extraction kits. To detect whether different species may have deposited hairs on the same trap, DNA was extracted from duplicate samples from each hair trap. Follicles from three hairs were deposited in a 2-ml, screw-top, tube containing 25 μl of $1 \times \text{TE}$ buffer and boiled for 10 min at 100°C . An additional 175 μl of $1 \times \text{TE}$ buffer was then added and boiled for 10 min at 100°C ; the resulting solution was used as a DNA template for PCR amplifications.

I used the HCarn200 (Bidlack et al., 2007) and CanidL1 (Paxinos et al., 1997) primers to amplify the first 196 base pairs of the mitochondrial cytochrome-b gene. Each 20 μl PCR reaction contained 10 μl Taq PCR Master Mix (Qiagen Inc., Valencia, California), 1 μl 10 μM HCarn200 primer, 1 μl 10 μM CanidL1 primer, 6 μl deionized H_2O and 2 μl diluted (1:50) DNA template. Thermal cycling was initiated at 94°C for 2 min, followed by 40 cycles of 94°C for 1 min, 54°C for 1 min and 72°C for 2 min. All PCR reactions included at least one negative control to monitor for contamination, and electrophoresis was used to check each reaction for successful amplification. I repeated amplification of DNA for samples that failed to amplify during the first PCR reaction.

I used restriction-fragment-length polymorphisms (RFLP) to identify amplified fragments to species in a two-step process designed to distinguish DNA from cougars, coyotes, bobcats, and gray foxes (Bidlack et al., 2007). I assumed that scats from American badgers and long-tailed weasels would not be deposited above-ground and that scats from American black bears, raccoons, and skunks could be distinguished according to visual characteristics (Murie, 1975; Halfpenny, 1986). PCR products were first digested with Hpa II (New England Biolabs, Ipswich, Massachusetts), which does not cut fragments from canids (coyote and gray fox), but cuts fragments from both felids (bobcat and cougar). Samples identified as canid were digested with HpyCH4 V (New England Biolabs, Ipswich, Massachusetts), which cuts fragments from gray foxes, but not coyotes. Samples identified as felid were digested with Bsl I (New England Biolabs, Ipswich, Massachusetts), which cuts fragments from cougars, but not bobcats. Each 10 μl digest reaction contained 3.75 μl deionized H_2O , 1 μl digest buffer, 0.25 μl restriction enzyme, and 5 μl PCR product. Reactions were incubated 4–6 h at the temperature indicated by the manufacturer. Electrophoresis was used to separate products on a 1.7% agarose gel and predicted cutting patterns were visualized using ethidium bromide and UV light. I repeated failed or ambiguous digests, and I excluded digests that failed twice from further analysis.

I compared detections of species by the four methods of survey by calculating which methods produced the greatest numbers of detections, which produced detections of the greatest number of species, and which species were detected by the least number of methods. I then used a maximum-likelihood-estimation modeling approach in the program PRESENCE 3.0 (United States Geological Survey Patuxent Wildlife Research Center, Laurel, Maryland; MacKenzie et al., 2006) to compare relative effectiveness of the four methods for detecting carnivores. The program PRESENCE allows for estimation of the probability of detecting a species (p) during one visit using each method. When the effort and duration of survey are defined for each method, this approach can be used to compare probabilities of detection among methods with variable effort of survey. It also can estimate number of surveys needed to ensure detection of a species, if it is present in a site (Gompper et al., 2006; O'Connell et al., 2006; Long et al., 2007c).

Data collected from both study areas were compiled into a detection history for each species (i.e., a

sequence of detections and nondetections by method and visit) for a total of ≤ 8 sampling occasions/season (e.g., $2 \times$ hair trap + $1 \times$ camera + $2 \times$ track plate + $3 \times$ transect). I used a single-season model with a custom-design matrix that treated methods and visits as covariates of probability of detection. Models were compared for a constant p (probabilities of detection were equal among methods and visits), a method-specific p (probabilities of detection varied by method), a visit-specific p (probabilities of detection varied by visit), and a survey-specific p (probabilities of detection varied by method and visit). All models maintained a constant rate of occupancy (ψ). Sites were assumed to be closed to changes in state of occupancy during each season, and methods producing no detection for a given species were excluded from the data matrix to reduce total number of parameters in the model. I compared models using Akaike's Information Criterion with a small-sample-size adjustment (AIC_c); in this instance, effective sample size was equal to total number of sites surveyed during 3 years ($n = 82$). Probabilities of detection for each method were estimated from the model with the greatest support (minimum AIC_c) or averaged from among highly ranked competing models ($\Delta AIC_c < 2$; Burnham and Anderson, 2002).

I estimated probabilities of occupancy in the two study areas for species detected at $\geq 10\%$ of sites during ≥ 1 season. Because sites were small relative to size of home range of most species detected (Crooks, 2002), probability of occupancy can be interpreted more accurately as an indicator of use of habitat during the season studied (June–September; MacKenzie and Royle, 2005). Thus, use of habitat was used to describe the variable ψ to distinguish it from true occupancy. To examine whether rates in use of habitat by a species varied as a function of characteristics of sites, I used a GIS database in ArcGIS 9.0 to measure several covariates of sites. In addition, I took measurements of canopy cover using a spherical densitometer (Forestry Suppliers, Inc., Jackson, Mississippi) at the center and four corners of each site. Measurements of canopy cover were averaged and all covariate values were centered prior to analyses. For the North Rim and South Rim, respectively, means ($\pm SD$) of these covariates were: elevation (m), $2,504.8 \pm 88.4$ and $2,163.1 \pm 66.0$; canopy cover (%), 67.1 ± 10.3 and 45.1 ± 14.0 ; distance to water (m), $1,269.8 \pm 574.2$ and $1,606.0 \pm 853.2$; distance to boundary of park (m), $6,914.1 \pm 4,371.7$ and $2,029.9 \pm 1,573.8$; and distance to public road (m), $1,220.0 \pm 1,138.4$ and 983.3 ± 717.2 .

I used the single-season model PRESENCE 3.0 to estimate probability of use of habitat in both study areas. I used a hierarchical approach to fit these models. I fit a model of probability of detection (constant p , method-specific p , visit-specific p , or survey specific p) using a constant model of use of habitat (ψ), and then held the best model of probability of detection constant for all subsequent comparisons of use of habitat. I estimated probabilities of use of habitats by comparing models that incorporated one or two covariates of sites with a model assuming a constant probability of use of habitat among sites. Models were compared using AIC_c with an effective size of sample

equal to total number of sites surveyed in the two study areas ($n_{NR} = 40$ and $n_{SR} = 42$). Probabilities of use of habitat for each species and study area were estimated from the top-ranked model (minimum AIC_c) or averaged from among highly ranked, competing models ($\Delta AIC_c < 2$).

I used the alternate parameterization (ψ_{Ba}/r_{Ba}) of the two-species model in PRESENCE 3.0 to examine co-occurrence between pairs of species in study areas. This analysis also was limited to species detected in $>10\%$ of sites during a monitoring season. For each comparison, the species with the largest number of detections per study area was selected to be Species A. Due to the large number of parameters in the two-species models ($K \geq 8$), probabilities of detection were assumed to be constant among methods and visits. I compared models incorporating individual covariates of sites independently for Species A and Species B with a model assuming a constant probability of use of habitat among sites. I selected the set of models whose cumulative weight ($\sum w_i$) exceeded 0.95, excluding models that failed to converge. I calculated model-averaged estimates of probabilities of use of habitat for Species A (ψ_A), for Species B when A was present (ψ_{Ba}), and for Species B when A was absent (ψ_{Ba}). I then used the model-averaged values to calculate the species-interaction factor (ϕ ; MacKenzie et al., 2006) for each pair of species. I propagated model-generated estimates of error through the equations and used a t -test (Zar, 1999) to compare resulting species-interaction factors to one, which was the value that would be expected if the two species were distributed independently.

RESULTS—I detected nine species of carnivores; the cougar, coyote, bobcat, American badger, raccoon, gray fox, striped skunk, western spotted skunk, and long-tailed weasel. All nine species were detected by a minimum of two methods, but four species (raccoon, striped skunk, western spotted skunk, long-tailed weasel) were detected during ≤ 5 visits during the study. Scats collected during surveys along transects yielded the greatest number of detections ($n = 77$) and 65% of scats ($n = 50$) were identified to species in the laboratory. I identified scats from all four species targeted by DNA methods (cougar, coyote, bobcat, and gray fox) and most scats (62%) were identified as coyote. Only 39% of samples of hair were successfully identified in the laboratory, yielding detections of three species: coyote (47%), gray fox (47%), and bobcat (7%). The coyote was the species most frequently detected by remotely triggered cameras and cameras also produced detections of rarer species, including the American badger and both species of skunks. Surveys along transects and track-plate stations produced detections of the greatest number of species, especially smaller carnivores.

TABLE 1—Modeling probabilities of detection of carnivores by method for sites ($n = 82$) on North and South rims of Grand Canyon, Coconino County, Arizona. Models are compared assuming that probabilities of detection are constant ($p(\cdot)$), vary by method ($p(\text{Method})$), vary by visit ($p(\text{Visit})$), or vary by method and visit ($p(\text{Method} \times \text{Visit})$). Methods producing no detection of a species were excluded from analysis to reduce the number of parameters. Information presented for each model includes twice the negative log-likelihood ($-2l$), number of parameters (K), change in AIC_c (small-sample correction to Akaike's Information Criterion) relative to the top model (ΔAIC_c), and Akaike weight (w_i ; weight of evidence in favor of a given model). All models assume a constant probability of use of habitat (ψ) among sites.

Model	$-2l$	K	ΔAIC_c	w_i
Gray fox				
$p(\text{Method})$	233.59	5	0	0.807
$p(\text{Method} \times \text{Visit})$	227.91	9	4.03	0.108
$p(\cdot)$	245.00	2	4.77	0.074
$p(\text{Visit})$	244.50	4	8.64	0.011
Coyote				
$p(\text{Method} \times \text{Visit})$	274.36	7	0	0.566
$p(\text{Visit})$	282.28	4	0.92	0.357
$p(\text{Method})$	285.95	4	4.59	0.057
$p(\cdot)$	292.45	2	6.72	0.020
Bobcat				
$p(\cdot)$	103.37	2	0	0.491
$p(\text{Method})$	99.36	4	0.36	0.410
$p(\text{Visit})$	102.43	4	3.43	0.088
$p(\text{Method} \times \text{Visit})$	97.17	8	7.62	0.011

Low overall rates of detection limited the number of species for which probabilities of detection and use of habitat could be estimated using maximum-likelihood-estimation methods. Covariates for method were included among the top-ranked models ($\Delta\text{AIC}_c < 2$) of probability of detection for the three most common species, and detection of coyotes also was influenced by visit to sites (Table 1). Remotely triggered cameras had the greatest probability of detecting coyotes ($\bar{p} = 0.200$) and gray foxes ($\bar{p} = 0.240$) during a single visit, while track plates had the greatest probability of detecting bobcats ($\bar{p} = 0.152$) (Fig. 1).

Four species were detected frequently enough to estimate rates of use of habitat by study area: coyote, bobcat, American badger, and gray fox. Top-ranked models ($\Delta\text{AIC}_c < 2$) for all four species included covariates of sites and relationships with covariates varied by study area and species (Table 2). Distance to a public road appeared in the greatest number of models and was related positively to use of habitat by coyotes and bobcats and negatively related to use of habitat by gray foxes. Distance to boundary of park also was related positively to use of habitat

by coyotes and bobcats on the South Rim and negatively related to use of habitat by bobcats on the North Rim. Elevation was related positively to use of habitat by American badgers on the North Rim and negatively related to use of habitat by gray foxes on the South Rim. Coyotes and

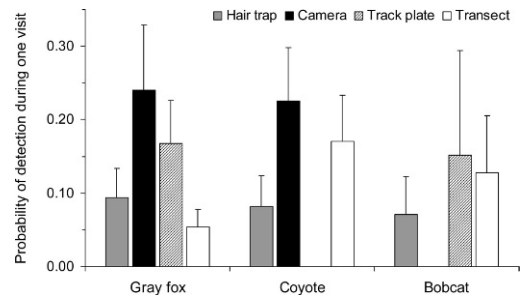


FIG. 1—Probability ($\pm SE$) of detecting a gray fox (*Urocyon cinereoargenteus*), coyote (*Canis latrans*), or bobcat (*Lynx rufus*) during one visit to a hair trap, remotely triggered camera, track plate, or survey along a transect on North and South rims of Grand Canyon, Coconino County, Arizona. Hair traps, remotely triggered cameras, and track plates were deployed for sampling intervals with a mean of 14.7 days (± 2.0 days). Transects were 800 m in length.

TABLE 2—Top-ranked models ($\Delta AIC_c < 2$) for use of habitat by species on North and South rims of Grand Canyon, Coconino County, Arizona. Models are compared assuming that probabilities of a species using a habitat (ψ) are constant among sites, or vary as a function of five characteristics of sites: elevation, canopy cover, distance to water, distance to boundary of park, or distance to a public road. Information presented for each model includes twice the negative log-likelihood ($-2l$), number of parameters (K), change in AIC_c (small-sample correction to Akaike's Information Criterion) relative to the top model (ΔAIC_c), Akaike weight (w_i ; weight of evidence in favor of a given model), and value of coefficients for one or two covariates of sites with respect to their effect on use of habitat (b_1 , b_2). Models of use of habitat maintain the same structure for probability of detection (p) indicated for the top-ranked model for each species and study area.

Model	$-2l$	K	ΔAIC_c	w_i	b_1	b_2
Coyote (North Rim)						
$\psi(\text{Road}), p(\cdot)$	88.19	3	0	0.343	1.21	
$\psi(\cdot)$	90.87	2	0.29	0.297		
$\psi(\text{Road}, \text{Canopy})$	86.72	4	1.07	0.201	1.42	0.86
$\psi(\text{Canopy})$	89.72	3	1.53	0.160	0.73	
Bobcat (North Rim)						
$\psi(\text{Road}), p(\cdot)$	56.26	3	0	0.645	1.61	
$\psi(\text{Road}, \text{Boundary})$	54.91	4	1.19	0.355	1.41	-0.87
American badger (North Rim)						
$\psi(\text{Elevation}), p(\cdot)$	50.03	3	0	0.650	-1.56	
$\psi(\text{Elevation}, \text{Canopy})$	48.73	4	1.24	0.350	-1.61	-0.69
Coyote (South Rim)						
$\psi(\cdot), p(\text{Visit})$	173.62	4	0	0.550		
$\psi(\text{Road})$	172.66	5	1.62	0.245	0.54	
$\psi(\text{Boundary})$	173.00	5	1.97	0.205	1.45	
Bobcat (South Rim)						
$\psi(\cdot), p(\cdot)$	39.71	2	0	0.536		
$\psi(\text{Boundary})$	37.68	3	0.29	0.464	0.84	
Gray fox (South Rim)						
$\psi(\cdot), p(\text{Method})$	190.65	5	0	0.486		
$\psi(\text{Road})$	188.85	6	0.94	0.304	-0.72	
$\psi(\text{Elevation})$	189.58	6	1.67	0.211	2.86	

American badgers had opposite relationships with canopy cover on the North Rim; use of habitat by coyotes increased with increasing canopy cover. Distance to water did not appear in top-ranked models for any species.

Model-estimated rates for use of habitat also varied by species and study area. Bobcats ($\bar{\psi}_{NR} = 0.287$) and American badgers ($\bar{\psi}_{NR} = 0.290$) were detected more frequently on the North Rim and coyotes ($\bar{\psi}_{SR} = 0.702$) and gray foxes ($\bar{\psi}_{SR} = 0.750$) were detected more frequently on the South Rim (Fig. 2). Raccoons and striped skunks were never detected on the North Rim and the remaining species, cougars, spotted skunks, and long-tailed weasels, were detected infrequently in both study areas.

I examined patterns of co-occurrence for pairs of the four species for which rates of use of

habitat were estimated. Coyotes and bobcats had a species-interaction factor of zero on the North Rim and coyotes and gray foxes had a species-interaction factor of significantly less than one on the South Rim ($P = 0.009$), indicating limited coexistence between these pairs of species (Table 3). I detected no evidence for limited coexistence between coyotes and American badgers, or American badgers and bobcats on the North Rim, or between gray foxes and bobcats or coyotes and bobcats on the South Rim.

DISCUSSION—I detected nine of the 11 species of mammalian carnivores believed to occur in Grand Canyon National Park. Neither American black bears nor ringtails were detected; cougars, raccoons, skunks, and long-tailed weasels were

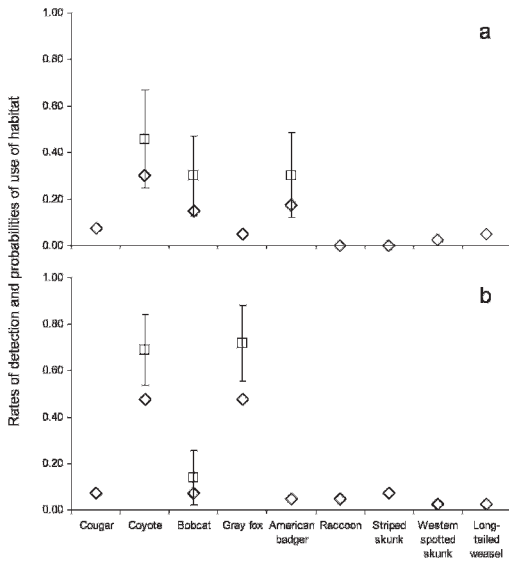


FIG. 2—Observed rates of detection expressed as a proportion of sites in which the species was detected by ≥ 1 method (\diamond) and predicted probabilities of use of habitat with *SE* indicated (\square) for nine species of carnivores on a) North (2003–2004) and b) South (2004–2005) rims of Grand Canyon, Coconino County, Arizona.

detected infrequently. American black bears are believed to persist in low densities in the park, but they are subject to hunting pressures in the adjacent Kaibab and Coconino national forests. Ringtails may be more abundant at lower elevations and in riparian areas in Grand Canyon (Poglayen-Neuwall and Toweill, 1988) and raccoons were detected only on rim regions of the park beginning in the 1990s (E. Leslie, pers. comm.). Recent research on GPS-collared animals indicates that cougars in Grand Canyon National Park have home ranges on the order of

500 km² (E. York, in litt.), making detections in the 4-ha monitoring sites relatively unlikely. The survey was not sufficiently intensive to determine true absences of undetected species or probabilities of use of habitat by rare species. Further research regarding fate of these species is warranted. Such inventory data and regional assessments of resources in Grand Canyon National Park (Parks and Harcourt, 2002).

The four surveying techniques were not equally effective for detecting carnivores. Method influenced probabilities of detection for coyotes, gray foxes, and bobcats (Table 1). Searches along transects yielded the greatest number of detections ($n = 114$) of eight species; most of these detections were scats. While rate of success for amplification was similar to other studies using DNA extracted from scats of carnivores (e.g., Long et al., 2007b), it might be improved by standardizing methods used to store scats in future research (Prugh et al., 2005). Despite the relatively large number of detections from surveys along transects, this method did not have the greatest probability of detecting the three most common species. Instead, models of probability of detection indicated that remotely triggered cameras had the greatest probability of detecting a coyote or gray fox during a single visit, and track plates had the greatest probability of detecting a bobcat (Fig. 1). Photographs provided unequivocal identifications of species and, along with the track-plate stations, detections of rarer species. Although hair traps produced the second-greatest number of detections ($n = 38$), DNA amplifications failed for most (61%) samples of hair. Nearly all (93%) samples of hair that were identified were from canids, despite using a scent

TABLE 3—Results of two-species models of co-occurrence of carnivores on North and South rims of Grand Canyon, Coconino County, Arizona. Model-averaged estimates of conditional probabilities of use of habitat by Species A and B were used to calculate the species-interaction factor (ϕ) and standard error (*SE*) for each pair of species. Results are given for a two-tailed *t*-test (*P*) comparing species-interaction factors to one, the value that would be expected if two species were distributed independently.

Study area	Species A/Species B	ϕ (<i>SE</i>)	<i>P</i>
North Rim	Coyote/Bobcat	0 (0)	<0.001
	Coyote/American badger	0.963 (0.275)	0.893
	American badger/Bobcat	0.504 (0.721)	0.496
South Rim	Gray fox/Coyote	0.401 (0.218)	0.009
	Gray fox/Bobcat	0.820 (1.233)	0.885
	Coyote/Bobcat	0.664 (1.539)	0.826

lure that was designed to attract felids. In addition, only coyotes, bobcats, and gray foxes visited hair traps and these three species were detected more frequently by other methods. These observations contribute to a growing body of evidence that hair traps are ineffective compared to other non-invasive methods (Harrison, 2006; O'Connell et al., 2006; Long et al., 2007c).

Results of this study are consistent with research from other regions of the United States, which indicates that no single non-invasive method is sufficient for community-level surveys (Gompper et al., 2006). Combined probability of detection over eight sampling occasions using the four methods (e.g., $2 \times$ hair trap + $1 \times$ camera + $2 \times$ track plate + $3 \times$ transect survey) was 0.589 for bobcats to 0.623 for coyotes to 0.633 for gray foxes. Model-estimated probabilities of detection (Fig. 1) could be used to optimize design of surveys for detection of particular species of carnivores. For example, a survey for gray foxes might emphasize use of cameras and track plates. To detect the full suite of species in communities of carnivores in western North American, pairing searches of transects with a greater number of remotely triggered cameras and track-plate stations may be the most reliable and efficient approach. In addition, efficiency and precision of surveys along transects could be improved through use of scat-detection dogs (Smith et al., 2003; Long et al., 2007b).

Despite the relatively intensive monitoring program, on average, model-estimated probabilities of use of habitat were 71.5% greater than observed rates of detection for species on the North Rim and 64.6% greater than observed on the South Rim (Fig. 2). These differences indicate the importance of accounting for imperfect detection in surveys for mammalian carnivores. Relying solely on pattern of detections of species could underestimate area used by a particular species. Incorporating false absences into regression models of distributions of species could lead to misestimation of relationships of habitat, especially where probabilities of detection covary with habitat (Tyre et al., 2003; Gu and Swihart, 2004; McKenzie et al., 2006). Additionally, large differences in detection of species and rates of use of habitat between study areas on the North and South rims highlight the importance of conducting

site-specific surveys of communities of carnivores.

Although study areas were selected to minimize variation in habitat among sites, evidence from PRESENCE models indicated that most probabilities of use of habitat varied as a function of characteristics of sites (Table 2). For example, probabilities of use of habitat by coyotes and bobcats were related positively to distance from public roads on the North Rim, whereas use of habitat by coyotes was related positively and use of habitat by gray foxes was related negatively to distance from public roads on the South Rim. These covariate relationships can, in turn, influence patterns of co-occurrence (MacKenzie et al., 2004). If coyotes and bobcats avoided public roads on the North Rim, they would be more likely to co-occur at monitoring sites than if they were distributed randomly. Conversely, the opposite response of coyotes and gray foxes to public roads on the South Rim could make them less likely to co-occur. Thus, it is important to model co-occurrence and habitat relationships simultaneously.

The two-species model indicated that two pairs of carnivores had limited co-occurrence in Grand Canyon National Park: coyotes and bobcats, and coyotes and gray foxes (Table 3). Coyotes and bobcats never co-occurred at sites on the North Rim and had a species-interaction factor of zero, indicating complete avoidance or exclusion. Coyotes and gray foxes had a species-interaction factor of 0.401 on the South Rim. This means that coyotes and gray foxes were less than one-half as likely to co-occur at monitoring sites than if they were distributed independently. Although I did not detect evidence for patterns of co-occurrence between the remaining pairs of species, it is important to note that estimates of error were relatively high for species-interaction factors in most of these comparisons (Table 3). Surveying a larger number of sites, or targeting methods to particular species, could help reduce uncertainty in the two-species models and confirm the results.

These observations are consistent with other studies that revealed spatial disassociation between sympatric carnivores. Studies using radio-telemetry have detected limited overlaps between locations of home range of different species. For example, in Venezuela, <15% of locations of cougars overlapped those for jaguars (*Panthera onca*; Scognamiglio et al., 2003). Simi-

larly, an average of 16% of the area of home ranges of bobcats overlapped with home ranges of coyotes in northern California (Neale and Sacks, 2001*b*). Comparison of empirically derived, distribution models also indicated that sympatric carnivores partition prey and habitat in space and time (Alexander et al., 2006). Given the limited area surveyed by each monitoring site (4 ha) in my study relative to sizes of home ranges of carnivores reported elsewhere (Crooks, 2002), it is reasonable to expect that species with segregated home ranges rarely would be detected at the same sites.

Quantifying spatial patterns of co-occurrence among species is important for a variety of biodiversity monitoring and conservation applications. Researchers need to explicitly account for spatial patterns of co-occurrence in the design of non-invasive surveys and incorporate distributions of co-occurring species as independent variables in development of models for distribution of species. Prior to conducting surveys, pairs of species that have the potential to interact spatially should be identified and previous research or pilot surveys should be used to estimate magnitude of the interaction. A two-species model using pilot or simulated data could be used to determine how much effort would be needed to accurately detect patterns of co-occurrence in the field.

If patterns of co-occurrence are likely to affect distributions of target species, then data on co-occurring species should be collected in the same manner as data on habitat or other independent variables. Temporal and spatial scales of multispecies surveys should be optimized for the whole community, so that probabilities of detection and rates of use of habitat can be estimated accurately for all target species. Additionally, variability among characteristics of monitoring sites should be minimized or controlled; especially, those factors that are likely to affect species differently (e.g., strong gradients in habitat). These steps are necessary to separate potential interspecific effects from other environmental factors and improve accuracy and utility of non-invasive surveys and associated spatial models.

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