



Research Article

A Multi-Method Hierarchical Modeling Approach to Quantifying Bias in Occupancy from Noninvasive Genetic Tagging Studies

JASON T. FISHER,¹ *Alberta Innovates—Technology Futures, Ecosystems Management Unit, #3-4476, Markham Street, Victoria, British Columbia Canada, V8Z 7X8*

STEVE BRADBURY, *Environment and Sustainable Resource Development, Government of Alberta, 2nd Floor Provincial Building, 111-54 Street, Edmonton, Alberta Canada, T7E 1T2*

ABSTRACT Detecting ecological change in mammalian communities requires large volumes of data collected across large landscapes. Noninvasive genetic tagging (NGT) can provide such data and is a common method of estimating species occupancy, distribution, and abundance. Methods to correct errors in genetic analysis exist, but estimating and accounting for detection error in NGT studies—specifically, detection error during hair trapping—has received less attention. If detectability varies with an ecological characteristic that is instead attributed to occupancy, detection error may lead to spurious conclusions. We demonstrate how multi-method occupancy models informed by camera-trapping data can quantify error, partition sources of variability, and estimate occupancy from NGT studies. We surveyed marten (*Martes americana*), fisher (*Pekania pennanti*), and wolverine (*Gulo gulo*) occurrence via hair trapping and camera trapping for 3 repeated monthly surveys at 66 sites in the Rocky Mountains of Alberta, Canada. Camera traps photographed the hair traps and surrounding sampling site. We used multi-method occupancy models to estimate site occupancy, probability of detection, and conditional occupancy at a hair trap. We tested the predictions that detection error in NGT studies could be induced by temporal variability and behavioral variability arising from the presence of heterospecifics. Models indicated that NGT via hair trapping consistently underestimated occupancy of marten, fisher, and wolverine compared to cameras. The magnitude of bias varied, but at worst underestimated occupancy by half. The presence of a heterospecific affected a species' detection at hair traps. Wolverine presence increased marten detection, whereas marten decreased fisher detection at NGT traps. For all species, detection error was reduced through successive monthly surveys, although heterogeneity induced by other mustelids remained. Detection error from multiple sources can bias NGT occupancy estimates, with implications for population estimation, ecological inference, species' legal status assessments, and conservation decisions. We recommend that NGT studies quantify and correct for detection error using independent survey methods and multi-method occupancy models, to improve the ability to answer ecological questions and make conservation decisions in the face of ecological change. © 2014 The Wildlife Society.

KEY WORDS camera trapping, monitoring, mustelids, occupancy estimation, spatial distribution.

Ecological changes from climate warming and anthropogenic landscape development are globally pervasive phenomena threatening biodiversity (Vitousek 1997, Walther et al. 2002). The need to understand and manage these changes has placed a steeply increasing premium on ways to maximize data collected per unit effort over large spatial scales, and over multiple biotic communities. Remote survey methods can collect large volumes of data for multiple species (Yoccoz et al. 2001, Lindenmayer and Likens 2010) and are cost- and time-efficient (e.g., MacKay et al. 2008, O'Connell et al. 2010). For mammals, noninvasive genetic tagging (NGT) yields low-cost and high-yield data on species

occurrence (Waits 2004, Kendall and McKelvey 2008). Trapped hair follicles can yield DNA samples, which can be amplified and identified to species by mitochondrial analysis. Microsatellite analysis can be applied further to identify individuals (McKelvey and Schwartz 2004a, b). Noninvasive genetic tagging data inform estimates of population size (Williams et al. 2009), density (Gardner et al. 2010), habitat selection (e.g., Fisher et al. 2011), and landscape genetics—the landscape-scale analysis of population connectivity and gene flow (Manel et al. 2003). Noninvasive genetic tagging has been used successfully for these purposes in many mammal species including grizzly bears (*Ursus arctos*; Mowat et al. 2005, Kendall et al. 2009), marten (*Martes americana*), fisher (*Pekania pennanti*; Mowat and Paetkau 2002, Fisher et al. 2012), and wolverines (*Gulo gulo*; Hedmark and Ellegren 2007, Fisher et al. 2013). Noninvasive genetic tagging is an important tool for ecological inquiry, and

Received: 4 April 2013; Accepted: 12 May 2014
Published: 7 July 2014

¹E-mail: jason.fisher@albertainnovates.ca

resulting data are frequently used in conservation decisions (e.g., Alberta Grizzly Bear Recovery Plan 2008).

Genetic analysis for NGT has known potential pitfalls, but controlled analytical methods and careful interpretation can overcome these (Taberlet et al. 1999, Mills et al. 2000, Waits and Leberg 2000, Paetkau 2004, Kendall et al. 2009). One notable problem with NGT surveys, as with many wildlife surveys, is that of false absences. When a species is not detected at a site, it is either absent, or it is present but goes undetected (MacKenzie et al. 2002). The problem of detection error in NGT sampling requires more consideration than it currently receives. Detection error is known to occur in NGT sampling (Campbell et al. 2008) but rarely has it been quantified, despite NGT's widespread use for conservation decisions and ecological inference. Without quantification, the degree of error in estimated population parameters remains unknown, potentially leading to spurious conclusions and misinformed management decisions.

Capture–recapture analysis of marked individuals accounts for false absences by using probability theory to estimate the present but undetected component of a sample population (Amstrup et al. 2005). Similarly, detection–nondetection data from unmarked animals can be analyzed in an occupancy framework, wherein species occupancy (ψ), or the probability that a site has been occupied by a species during the sampling frame, is modeled in conjunction with its probability of detection (p), or the probability of detecting that species when it is, in fact, present (MacKenzie et al. 2002, 2006). Because p is often less than 1, naïve occurrence measures are negatively biased. A species' probability of detection may vary among surveys, habitats, and seasons (MacKenzie et al. 2006). Estimated values of p are also likely to vary among survey methods because the efficacy of each method may differ (Nichols et al. 2008).

Noninvasive genetic tagging via hair trapping is known to underestimate species' occurrence (Long et al. 2007, Campbell et al. 2008). Hair trapping for NGT requires that an animal 1) encounter a site, 2) interact with a trap, and 3) leave a hair sample from which DNA is extracted. Each step inherently has variability and a potential for error. However, the extent of underestimation in NGT studies is typically only qualitatively assessed, or estimated based on p (Campbell et al. 2008). The present but undetected fraction from NGT occupancy studies is rarely independently validated because the true occupancy of an area is rarely known. Unmodeled heterogeneity in detection can bias estimated occupancy and, until recently, quantifying this error has been difficult. For example, 2 potentially common sources of detection error in NGT sampling are temporal variability, where detection probability changes through time (e.g., Fisher et al. 2013), and site-induced variability, where responses to local conditions may induce detection error or variability in occupancy (MacKenzie et al. 2006).

Our objective was to demonstrate how detection error in NGT studies could be quantified using multi-method occupancy models (Nichols et al. 2008) when the NGT hair trap, and the area around it, is sampled using a second method: infrared remote cameras. Cameras are widely used

for surveying mammals (Gompper et al. 2006; O'Connell et al. 2006, 2010), and have been used to characterize species density (Trolle and Kéry 2003), diversity (Tobler et al. 2008), habitat selection (Fisher et al. 2011), spatial interspecific interactions (Fisher et al. 2012), and to estimate population and community changes through time (Karanth et al. 2006, O'Brien et al. 2010). Cameras detect species' occurrence within an area defined by the size of their detection cone (Rowcliffe et al. 2008), which in our illustration includes both the hair trap and the area immediately surrounding that trap.

We used repeat-surveys at baited sites for 3 sympatric mustelid species—wolverine, fisher, and marten—at baited survey sites in the Rocky Mountains of Alberta, Canada. We obtained the data from ongoing research on the response of mammalian distribution to landscape structure and disturbance (Fisher et al. 2011, 2012, 2013). We tested 4 hypotheses: 1) the detection of an individual at a site by hair collection is lower than detection by a camera; 2) these differences in detection are expected to vary among species; 3) detection probabilities are expected to vary monthly through a study period in winter; and 4) interference among mustelid species is expected to decrease detection probabilities and probability of occupancy at a site. Interference may occur in 2 ways. An individual may avoid a baited hair trap because the bait was previously eaten or marked by a heterospecific (reduced detectability) or may avoid a site altogether (reduced occupancy). Our objective was to quantify detection error using hierarchical multi-method occupancy models and to examine if the presence of heterospecifics can affect species occurrence as measured by NGT.

STUDY AREA

We surveyed mustelid occupancy in the Willmore Wilderness Park of the Rocky Mountains of Alberta, Canada (Fig. 1). The Willmore Wilderness is a 4,600 km² conservation area that is located on the Front Ranges of the Rocky Mountains. It is part of the Western Cordilleran system, and is protected from anthropogenic disturbance and motorized recreation. Topography was rugged, with high peaks over 2,500 m, steep-sloped ridges, and valley bottoms. Coniferous forest 80–120 years old (*Pinus contorta*, *Picea glauca*, *Picea mariana*, and *Abies balsamea*) dominated this landscape, with some small deciduous (*Populus tremuloides*, *Populus balsamifera*) stands occurring throughout. Small stands of black spruce (*Picea mariana*) with forest floors dominated by Labrador tea (*Ledum groenlandicum*) and mosses (*Sphagnum* spp.) occurred in low-lying areas. Pine and mixed stands were often fairly open, with a sparse alder (*Alnus crispa*) understory. Younger fire-origin stands were embedded within this matrix. Marten, fisher, and wolverine occurred throughout this region (Fisher et al. 2012, 2013) as well as grizzly bears and black bears (*Ursus americanus*), cougar (*Puma concolor*), moose (*Alces alces*), deer (*Odocoileus virginianus* and *O. hemionus*), bighorn sheep (*Ovis canadensis*), and a diversity of other carnivores and rodents (Fisher

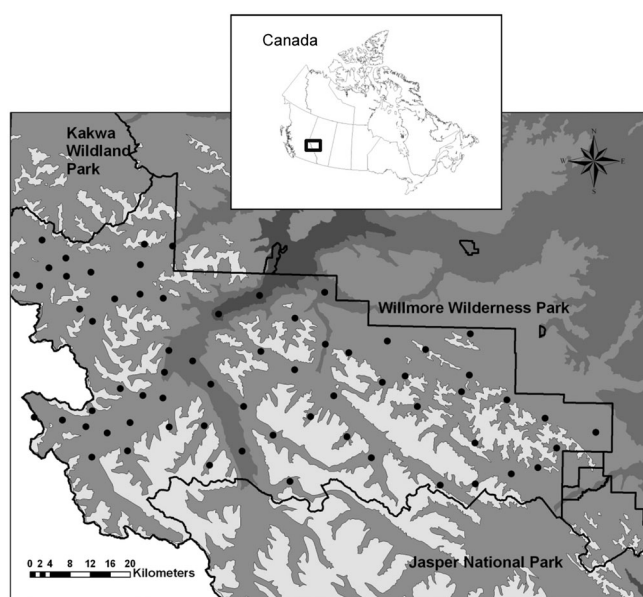


Figure 1. Location of 66 camera- and hair-trapping stations (black dots) throughout the 4,600-km² Willmore Wilderness Area in west-central Alberta, Canada, 2006–2008. This mountain landscape in the Front and Main Ranges of the Rocky Mountains includes herbaceous alpine patches (light gray), mid-elevation subalpine forests (medium gray), and lower-elevation montane and foothills forests (dark gray).

et al. 2011). Registered traplines occur throughout this area and all three mustelid species may be legally trapped.

METHODS

We set carcass-baited sampling sites in December and monitored and re-baited them monthly through March when food for mustelids was scarce and species were most likely to be attracted to the bait. We located bait sites using a systematic sampling design constrained by helicopter access and avalanche risk (Fisher et al. 2011, 2012). We placed sites ($n = 66$) an average of 5,727 m apart ($SD = 1,574$ m) in a roughly rectangular grid that covered most of the Park (Fig. 1) to accommodate the large home-range sizes of wolverines, the largest of the 3 species (Fisher et al. 2013). We sampled 30 sites in 2006–2007 and 36 different sites in 2007–2008, with sites selected randomly between years.

Sampling Species Occurrence

We used 2 concurrent methods to sample mustelid occupancy: NGT via hair sampling and camera trapping (Fig. 2). Hair traps consisted of Gaucho[®] barbed wire (Bekaert, Brussels, Belgium) wrapped around a tree baited 2-m up the bole with a large (approx. 15-kg) skinned beaver carcass and O'Gorman's LDC Extra scent lure (O'Gorman's Co., Broadus, MT, USA). Hair could be snagged on barbed wire as animals climbed the tree for bait. We collected hair samples monthly (Jan–Mar) using sterile techniques. Wildlife Genetics International (WGI; Nelson, British Columbia, Canada) analyzed DNA from hairs to identify species. They extracted DNA from hairs using Qiagen[®] DNEasy[™] Tissue Kits (Qiagen, Hilden, Germany) and analyzed DNA to identify species using sequence-based

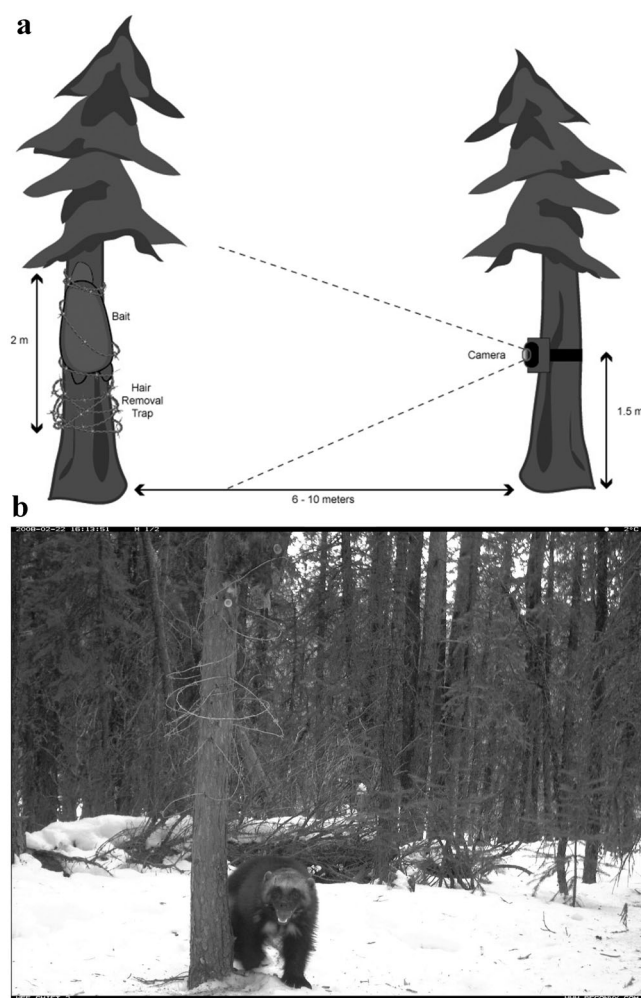


Figure 2. We surveyed occupancy of Rocky Mountain wolverines, marten, and fisher at each of 66 sites using 2 methods in 2006–2008: hair trapping for noninvasive genetic tagging (NGT), and camera trapping. (a) The hair trap consisted of barbed wire loosely wrapped around a baited tree. The Reconyx[™] passive infrared-triggered digital camera was positioned on a tree 6–10 m away to photograph the hair trap and the area around it. (b) If the animal was present at the site but did not leave hair, it was photographed.

analysis of the 16S rRNA gene of mitochondrial DNA (mtDNA; sensu Johnson and O'Brien 1997) that they then compared against a DNA reference library of all known mammal species in the region.

At each station, we also deployed 1 Reconyx[™] RM30 (2006–2007) or PM30 (2007–2008) infrared-triggered digital camera (Reconyx, Holmen, WI, USA). We placed the camera approximately 6–10 m from the tree such that the camera's detection cone and field of view included the NGT hair trap and the area surrounding it (Fig. 2). We downloaded camera data monthly in conjunction with hair data collection. We analyzed images and summarized species presence within each month (Jan–Mar). The final data frame was comprised of 66 sites with 3 surveys and 2 methods per site.

Statistical Analysis

We used the single-season, multi-method model of Nichols et al. (2008), an extension of occupancy models (MacKenzie

et al. 2002, 2006). Multi-method models allow simultaneous use of data from 2 or more detection methods for inference about method-specific detection probabilities. The models address dependence of detections within a sampling occasion, and capitalize on this dependence to inform inference about scale-specific occupancy (Nichols et al. 2008). Occupancy models assume that sites are closed to changes in occupancy at the species level within the sampling season, or for mobile animals, that changes are random among sites and among surveys. Mustelids are long-lived animals with stable territories, so this study likely met model assumptions. For mobile animals, occupancy models assume that a species inhabiting a surveyed area has a non-zero probability of being present at the survey site within the sampling season; we designed month-long surveys to satisfy this condition. We pooled the 2 years of data into a single season, because we expected no net difference in occupancy between sites between years (e.g., Fisher et al. 2014). A species would have to be removed entirely from a site (non-randomly with respect to time and covariates) to violate this assumption, and although species are trapped, they are not trapped differentially among years. Each month constituted a survey (repeated 3 times), and the winter study period was the single sampling season (cf. MacKenzie et al. 2002). For example, if we detected marten with camera traps in January, February, and March, but with hair traps only in January, then the detection histories for marten at that site would be 111 (cameras) and 100 (hairs).

In our multi-method survey protocol, animals at the hair trap were fully exposed to the camera trap (Fig. 2), and the detection area of the cameras was greater than point-detection at the hair trap. Therefore, we considered NGT hair traps as the immediate sample location (cf. Nichols et al. 2008) and the combination of cameras and NGT traps as the larger-scale sample location, wherein: $\psi = \text{Pr}(\text{sampling unit occupied})$; $p_t^s = \text{Pr}(\text{detection at survey } t \text{ by method } s |$

sample unit occupied and species present at immediate sample location); $\theta_t = \text{Pr}(\text{species occupying NGT site at occasion } t | \text{sample unit occupied})$; $\psi \cdot \theta_t = \text{Pr}(\text{occupancy at NGT site})$; $s = 2$ sampling devices, and $t = 3$ monthly surveys.

These definitions differ slightly from the original formulation wherein each detection device constituted the immediate scale, and the larger scale was not tied to a particular method but rather a sum of all devices (Nichols et al. 2008). In our design, cameras explicitly sampled NGT hair traps, and data show a near-zero probability that a species could be sampled at a hair trap without being sampled by a camera; no species detections via NGT hair trap were missed by the cameras. Therefore, the larger scale corresponds to the camera-imaged sampling area, within which is embedded the immediate scale point-sampling hair trap. In this study, θ_t is the parameter of particular interest; it refers to the conditional probability that a species estimated to occupy a site (from camera-trap data) will also occupy a hair trap (i.e., climb a tree and leave a hair with identifiable DNA). Therefore, $1 - \theta_t$ estimates the probability that a species present at a site will not be detected by NGT, and so estimates the degree of observed bias.

We estimated parameters using multi-method hierarchical models in program PRESENCE ver. 4.9 (Hines 2006), which employs maximum likelihood estimation for generalized linear models. For each of the 3 species, we constructed multiple competing models to weigh evidence in support of each hypothesis (Tables 1–3). These included 1) mustelid avoidance may prevent a species from occupying a site, where site occupancy (ψ) was either constant (.) or varied with the occurrence of each heterospecific (marten, fisher, or wolverine); 2) probability of detection (p) may be constant (.) or vary through the winter among surveys as the bait becomes more known or more valuable relative to available resources or p may be unique at each survey and device; 3) another

Table 1. Multi-method occupancy models of wolverine camera- and hair-trapping data collected in the winters 2006–2008 in the Rocky Mountains of Alberta, Canada. Occupancy (ψ) was either constant (.) or varied with the occurrence of marten or fisher. Conditional occupancy varied through time irrespective of other species (t), or varied through time and with the occurrence of marten or fisher. Probability of detection (p) was either constant (.) among methods (camera or hair) and through time, constant among methods but varying through time (t), or unique at each method and survey as $p(s, t)$.

| Model | AIC ^a | ΔAIC | w_i^b | Model L^c | K^d | $-2 \log L^e$ |
|---|------------------|--------------------|---------|-------------|-------|---------------|
| $\psi(.), \theta(t), p(s, t)$ | 452.39 | 0.00 | 0.34 | 1.00 | 10 | 432.39 |
| $\psi(\text{marten}), \theta(t), p(s, t)$ | 453.56 | 1.17 | 0.19 | 0.56 | 11 | 431.56 |
| $\psi(.), \theta(\text{marten}), p(s, t)$ | 453.58 | 1.19 | 0.19 | 0.55 | 11 | 431.58 |
| $\psi(.), \theta(\text{fisher}), p(s, t)$ | 453.97 | 1.58 | 0.15 | 0.45 | 11 | 431.97 |
| $\psi(\text{fisher}), \theta(t), p(s, t)$ | 454.15 | 1.76 | 0.14 | 0.41 | 11 | 432.15 |
| $\psi(.), \theta(t), p(t)$ | 498.19 | 45.80 | 0.00 | 0.00 | 7 | 484.19 |
| $\psi(\text{marten}), \theta(t), p(t)$ | 499.36 | 46.97 | 0.00 | 0.00 | 8 | 483.36 |
| $\psi(.), \theta(t), p(.)$ | 499.40 | 47.01 | 0.00 | 0.00 | 5 | 489.40 |
| $\psi(.), \theta(\text{marten}), p(t)$ | 499.91 | 47.52 | 0.00 | 0.00 | 8 | 483.91 |
| $\psi(.), \theta(\text{fisher}), p(t)$ | 499.93 | 47.54 | 0.00 | 0.00 | 8 | 483.93 |
| $\psi(\text{fisher}), \theta(t), p(t)$ | 499.95 | 47.56 | 0.00 | 0.00 | 8 | 483.95 |
| $\psi(\text{marten}), \theta(t), p(.)$ | 500.57 | 48.18 | 0.00 | 0.00 | 6 | 488.57 |
| $\psi(.), \theta(\text{marten}), p(.)$ | 501.09 | 48.70 | 0.00 | 0.00 | 6 | 489.09 |

^a Akaike's Information Criterion.

^b AIC weight.

^c Likelihood.

^d Number of parameters in the model.

^e $-2 \log$ likelihood.

Table 2. Multi-method occupancy models of fisher camera- and hair-trapping data collected in the winters 2006–2008 in the Rocky Mountains of Alberta, Canada. Occupancy (ψ) was either constant (.) or varied with the occurrence of marten or wolverine. Conditional occupancy varied through time irrespective of other species (t), or varied through time and with the occurrence of marten or wolverine. Probability of detection (p) was either constant among methods (camera or hair) and through time, constant among methods but varying through time (t), or unique at each method and survey as $p(s,t)$.

| Model | AIC ^a | Δ AIC | w_i ^b | Model L^c | K^d | $-2 \log L^e$ |
|---|------------------|--------------|--------------------|-------------|-------|---------------|
| $\psi(.), \theta(\text{marten}), p(s,t)$ | 165.13 | 0.00 | 0.62 | 1.00 | 11 | 143.13 |
| $\psi(.), \theta(t), p(s,t)$ | 168.80 | 3.67 | 0.10 | 0.16 | 10 | 148.80 |
| $\psi(.), \theta(\text{marten}), p(.)$ | 169.23 | 4.10 | 0.08 | 0.13 | 6 | 157.23 |
| $\psi(.), \theta(\text{wolverine}), p(s,t)$ | 169.81 | 4.68 | 0.06 | 0.10 | 11 | 147.81 |
| $\psi(\text{wolverine}), \theta(t), p(s,t)$ | 170.56 | 5.43 | 0.04 | 0.07 | 11 | 148.56 |
| $\psi(\text{marten}), \theta(t), p(s,t)$ | 170.77 | 5.64 | 0.04 | 0.06 | 11 | 148.77 |
| $\psi(.), \theta(\text{marten}), p(t)$ | 172.13 | 7.00 | 0.02 | 0.03 | 8 | 156.13 |
| $\psi(.), \theta(t), p(.)$ | 172.46 | 7.33 | 0.02 | 0.03 | 5 | 162.46 |
| $\psi(.), \theta(\text{wolverine}), p(.)$ | 173.48 | 8.35 | 0.01 | 0.02 | 6 | 161.48 |
| $\psi(\text{wolverine}), \theta(t), p(.)$ | 174.22 | 9.09 | 0.01 | 0.01 | 6 | 162.22 |
| $\psi(\text{marten}), \theta(t), p(.)$ | 174.43 | 9.30 | 0.01 | 0.01 | 6 | 162.43 |
| $\psi(.), \theta(t), p(t)$ | 175.28 | 10.15 | 0.00 | 0.01 | 7 | 161.28 |
| $\psi(.), \theta(\text{wolverine}), p(t)$ | 176.31 | 11.18 | 0.00 | 0.00 | 8 | 160.31 |

^a Akaike's Information Criterion.

^b AIC weight.

^c Likelihood.

^d Number of parameters in the model.

^e $-2 \log$ likelihood.

species scavenging the bait may prevent heterospecifics from scavenging the carcass and therefore interacting with the hair trap. Conditional occupancy may either vary through time irrespective of other species (t), or vary both through time and with the occurrence of heterospecifics.

We ranked models using an information-theoretic approach based on Akaike's Information Criterion (AIC) scores and their normalized AIC weights (w_i), which describe the weight of evidence in support of each model (Burnham and Anderson 2002). We summed w_i and calculated evidence ratios (ER; Anderson 2008) to summarize the overall importance of each variable in explaining wolverine occurrence. For example, $ER=2$ suggests the evidence for inclusion of an explanatory variable is 2 times greater than for its exclusion. We obtained model-averaged ψ

estimates from model sets by weighting each model's contribution to the parameter estimate and its standard error by w_i (Burnham and Anderson 2002). From per-survey estimates of p , we calculated the probability of false absence (PFA) for a given survey duration as $[1 - p]^t$ (Long and Zielinski 2008) with $t=3$ independent surveys.

RESULTS

We collected 489 hair samples across the 66 sites from multiple species. Of these, 458 (94%) contained enough roots to merit extraction; 34 samples (7%) failed during the mtDNA analysis. Of successfully identified samples, 162 were wolverine, 183 were marten, and 36 were fisher. We also detected cougar (7 samples), coyote (*Canis latrans*; 2),

Table 3. Multi-method occupancy models of marten camera- and hair-trapping data collected in the winters 2006–2008 in the Rocky Mountains of Alberta, Canada. Occupancy (ψ) and conditional occupancy (θ) were either constant (.) or varied with the occurrence of fisher or wolverine. Probability of detection (p) was either constant (.) among methods (camera or hair) and through time, constant among methods but varying through time (t), or unique at each method and survey as $p(s,t)$.

| Model | AIC ^a | Δ AIC | w_i ^b | Model L^c | K^d | $-2 \log L^e$ |
|---|------------------|--------------|--------------------|-------------|-------|---------------|
| $\psi(.), \theta(\text{wolverine}), p(s,t)$ | 394.71 | 0.00 | 0.87 | 1.00 | 11 | 372.71 |
| $\psi(.), \theta(t), p(s,t)$ | 400.23 | 5.52 | 0.06 | 0.06 | 10 | 380.23 |
| $\psi(\text{wolverine}), \theta(t), p(s,t)$ | 401.31 | 6.60 | 0.03 | 0.04 | 11 | 379.31 |
| $\psi(\text{fisher}), \theta(t), p(s,t)$ | 402.20 | 7.49 | 0.02 | 0.02 | 11 | 380.20 |
| $\psi(.), \theta(\text{fisher}), p(s,t)$ | 402.23 | 7.52 | 0.02 | 0.02 | 11 | 380.23 |
| $\psi(.), \theta(\text{wolverine}), p(t)$ | 502.15 | 107.44 | 0.00 | 0.00 | 8 | 486.15 |
| $\psi(.), \theta(t), p(t)$ | 503.00 | 108.29 | 0.00 | 0.00 | 7 | 489.00 |
| $\psi(\text{wolverine}), \theta(t), p(t)$ | 504.11 | 109.40 | 0.00 | 0.00 | 8 | 488.11 |
| $\psi(.), \theta(\text{fisher}), p(t)$ | 504.95 | 110.24 | 0.00 | 0.00 | 8 | 488.95 |
| $\psi(\text{fisher}), \theta(t), p(t)$ | 504.96 | 110.25 | 0.00 | 0.00 | 8 | 488.96 |
| $\psi(.), \theta(\text{wolverine}), p(.)$ | 506.01 | 111.30 | 0.00 | 0.00 | 6 | 494.01 |
| $\psi(.), \theta(t), p(.)$ | 508.89 | 114.18 | 0.00 | 0.00 | 5 | 498.89 |
| $\psi(\text{wolverine}), \theta(t), p(.)$ | 510.01 | 115.30 | 0.00 | 0.00 | 6 | 498.01 |

^a Akaike's Information Criterion.

^b AIC weight.

^c Likelihood.

^d Number of parameters in the model.

^e $-2 \log$ likelihood.

lynx (*Lynx canadensis*; 6), and red fox (*Vulpes vulpes*; 3). From camera traps, we obtained 115,488 photographs containing mammal species ranging in size from red squirrels (*Tamiasciurus hudsonicus*) to cougars (see Fisher et al. 2011). As expected, no DNA detections were missed by the cameras.

The probability of detecting a species when present varied by device and by survey for wolverines ($\Sigma w_i = 1.0$; Table 1), fisher ($\Sigma w_i = 0.85$; ER = 5.8; Table 2), and marten ($\Sigma w_i = 1.0$; Table 3). Wolverine, fisher, and marten detectabilities were consistently lower at hair traps than at camera traps (Fig. 3). The probability of detecting a species at a hair trap ranged from 0.48 to 0.67 for wolverines, 0.43–0.71 for fisher, and 0.38–0.71 for marten (Fig. 3). After 3 months of sampling across both years, the probability of false absence was 0.10 for wolverine, 0.07 for fisher, and 0.09 for marten. We found an approximately 0.90 probability of detecting these species via hair trapping over the 3 months. Camera traps had very high probability of detecting both fisher and marten ($p \sim 1.0$; PFA < 0.001).

The conditional probability that wolverines would occupy a hair trap given they were present at cameras (θ) was only 0.54 in the first January survey, indicating that NGT detection error can underestimate site occupancy by about half. Estimated θ increased through time to 0.90 by the final March survey (Fig. 4), and was not influenced by the presence of marten (ER = 0.19) or fisher (ER = 0.15). Wolverine occupied 0.90 (SE = 0.04) of the sites in this mountain landscape; site occupancy was not influenced by the presence of marten (ER = 0.23) or fisher (ER = 0.16).

Noninvasive genetic tagging produced biased estimates of fisher occupancy, and the degree of bias varied markedly with marten presence. Marten presence decreased fisher conditional occupancy at a hair trap (ER = 2.53). Fisher, when present, were expected to always leave hair where marten were absent ($\theta_{\text{fisher}} = 1.0$), but did so with a probability of only 0.32 when marten were also present at a site ($\theta_{\text{fisher}} = 0.32$; SE = 0.13). Wolverines had no effect on conditional occupancy of fisher at hair traps (ER = 0.08). Site occupancy by fisher was less than other mustelids, with a probability of occupying a site of only 0.30 (model-averaged $\psi = 0.30$; SE = 0.09). Site occupancy was not influenced by the presence of marten (ER = 0.05) or wolverine (ER = 0.05).

Marten conditional occupancy at a hair trap was not affected by the presence of fisher. However, we found a strong effect of wolverine occupancy ($\Sigma w_i = 0.87$; ER = 6.8). Contrary to expectations, the conditional probability that marten would occupy a hair trap given they were detected by cameras was greater when wolverine also occupied that site. At the start of sampling in January, marten were almost twice as likely to go up the tree and deposit hair at sites with wolverines ($\theta = 0.83$) than at sites without wolverines ($\theta = 0.49$). This difference was also marked in February (0.87 vs. 0.56) but closed by the end of sampling in March (Fig. 4). Across this landscape, marten were the most common species detected (model-averaged $\psi = 0.94$; SE = 0.03); marten site-occupancy was not influenced

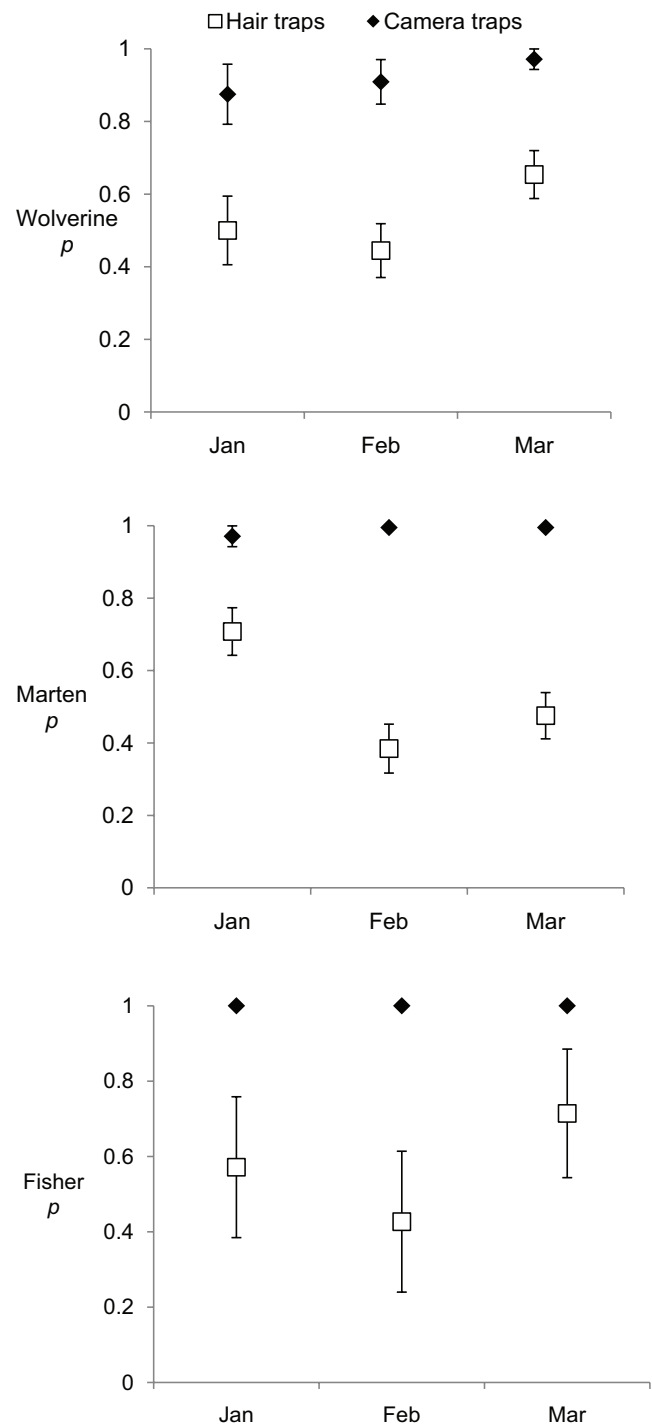


Figure 3. Probability of detection (p) varied through time for wolverines, fisher, and marten in the Rocky Mountains of Alberta, Canada, 2006–2008. All 3 species were more likely to be detected via camera traps than hair traps.

by the presence of fisher (ER = 0.02) or wolverine (ER = 0.02).

DISCUSSION

In our multi-method approach, the conditional probability of occupancy (θ) at hair traps given the animal was present at the sampling site was less than 1; NGT via barbed-wire hair

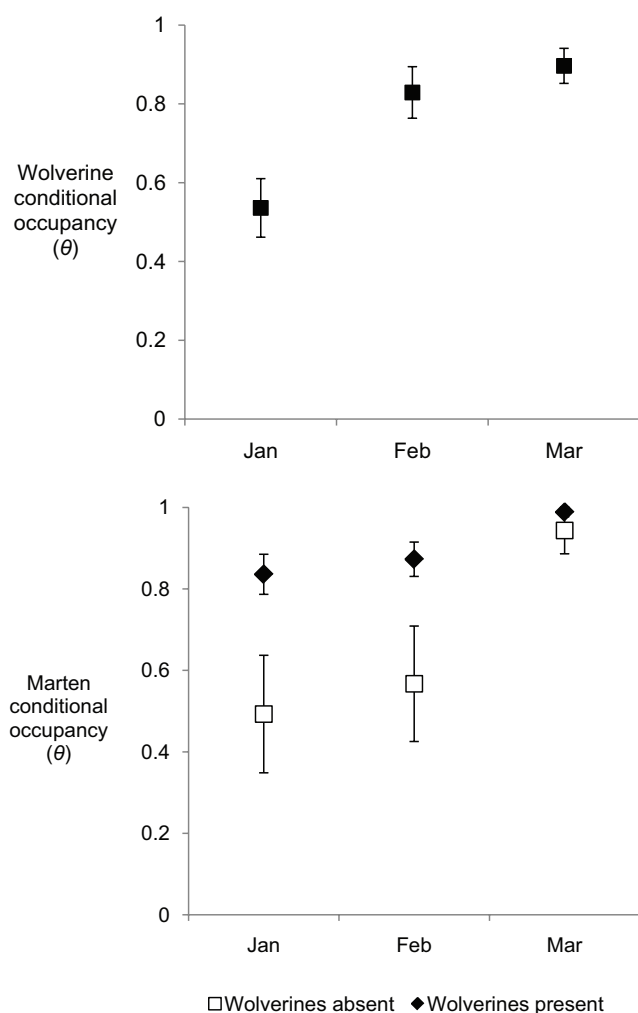


Figure 4. The conditional probability of occupancy at an NGT trap, given presence at a survey site, increased in monthly surveys for marten and wolverine. Marten present at a site were more likely to occupy the hair trap if wolverine were present at a site during the study (\blacklozenge) than if they were absent (\square); this difference diminished through time.

trapping sometimes failed to detect mustelids confirmed present by cameras. Explicitly sampling NGT hair traps with camera traps allows scientists to quantify the degree to which occupancy estimates are biased because of detection error. Other studies have integrated camera- and hair-trapping for single species (e.g., wolverines; Magoun et al. 2011) or multiple species (Long et al. 2007) using qualitative approaches. Few have used multiple methods to improve inferences, although quantitative models are emerging (Sollmann et al. 2013).

The degree of bias in occupancy estimates varied among species. Conducting NGT sampling for only 1 month could underestimate wolverine and marten occupancy as much as half, and fishers by two-thirds ($1 - \theta$), depending on time of year and the presence of heterospecifics. Extending the NGT sampling duration to 3 months increased the likelihood that mustelid occupancy estimated via NGT sampling reflected occupancy estimated from cameras. We propose that this error is not unique to our sampling method but may be widespread among NGT surveys. Imperfect detection from

hair sampling has been reported previously. Nichols et al. (2008) also found that hair traps underestimated occurrence, and Long et al. (2007) found that hair snares significantly under-sampled black bears and fisher compared to cameras and scat detection dogs. Campbell et al. (2008) provided a qualitative comparison of error in noninvasive methods such as NGT and showed that underestimation may be quite common. Imperfect detection poses a problem to robust inference if the degree of error is unknown and is not fully accounted for when estimating population size, distribution, density, and other key ecological parameters. Mark-recapture analysis, for example, aims to estimate and account for this error in abundance estimation, but these estimates are rarely independently validated and may retain an unknown degree of error.

Camera traps can provide some of this validation; cameras were substantially more reliable at detecting mustelids than were NGT surveys. Estimated p from camera trapping differs markedly among mammal species, probabilistic designs, and sampling methods (O'Connell et al. 2006, 2010). Cameras can fail to detect species, if present, for several reasons: cameras may malfunction, movement or snow can obscure identification of the species in the photo, or the species may simply fail to appear at the camera site within a monthly survey. We found that wolverines were detected by cameras, when present with 0.87–0.97 probability depending on survey month. Fisher and marten showed even greater detection probability with cameras. This high detectability (near 1.0) allowed us to reliably incorporate heterospecific presence into occupancy models; otherwise, that covariate would have its own unmodeled error. High detectability is a significant benefit of camera trapping. The dual-method approach of NGT sampling and cameras combines the diverse biological data garnered from the former, with the greater detection probabilities of the latter.

Arguably, detection error is not a problem if the detection probabilities are modeled. With sufficient surveys, PFA becomes reasonably low. After 3 months of surveys, which is a long survey duration compared to many (Kendall and McKelvey 2008), mustelid PFA was reduced to <0.10 in hair trapping and <0.01 in camera trapping. Even when error is low, quantifying it helps to judge precision and is necessary for correcting occupancy estimates. We recommend that hair trapping for NGT studies be of sufficient duration and repetition to increase p , decrease PFA, and reduce detection error. A sampling level that is sufficient to meet study objectives can be informed by pilot data and analysis. Bailey et al. (2007) provide guidelines for sampling trade-offs in occupancy studies based on pilot data.

Our study illustrates that unmodeled variation in conditional occupancy (θ) differs among species and may not be random. Even after repeated surveys and with a small PFA, conditional occupancy at an NGT trap given presence could be as low as half that of true occupancy determined by cameras. More importantly, θ varied with the occurrence of heterospecifics, whose abundance may vary across ecosystems. Interactions among the 3 mustelid species affected their detectability and occupancy estimates. Because

detection error was not distributed randomly among sites, it could quite conceivably lead to spurious conclusions about distribution or abundance, and the ecological processes driving them.

Modeling variability in θ in relation to ecosystem characteristics not only adjusts for this error, but also provides its own ecological insights. Sympatric marten and fisher have high dietary overlap (Zielinski and Duncan 2004) and compete for resources. Interference competition and dominance hierarchy between marten and fisher has received limited attention (see references in Krohn et al. 1997), and wolverines none at all. Our results indicate that fisher present at a site were much less likely to use the hair trap at sites where marten occurred, which may reflect their unwillingness to obtain a bait reward at the cost of perceived risk. In contrast to our expectation, the probability that a marten occupies a hair trap increases if it had been used by a wolverine. We hypothesize that marten are risk-averse and more likely to interact with a trap if wolverines have successfully done so without consequence. Alternatively, perceived competition for resources may signal to marten a greater need for the bait reward. Interference competition, where marten preclude fisher from a carcass, or exploitation competition, where marten consume a carcass before fisher, may explain trap avoidance by fisher at sites with marten. Examination of dominance relationships and spatiotemporal partitioning is possible by analyzing photographs, and may help elucidate these mechanisms. Observed variation in θ is possibly related to other unmeasured differences among sites, such as habitat, although this seems unlikely based on existing data (see Fisher et al. 2012). Excepting this possibility, sympatric heterospecifics appear to negatively or positively affect error in NGT hair-trapping studies in a non-trivial way.

Noninvasive genetic tagging is a useful tool that has greatly expanded our understanding of ecology and conservation biology. Genetic data are irreplaceable for identifying individuals, assessing relatedness, and investigating gene flow through landscape genetics, provided that errors in genetic analysis can be modeled (Taberlet et al. 1999, Mills et al. 2000, Waits and Leberg 2000). We maintain that detection error associated with NGT also deserves analytical diligence. Different hair-trapping methods likely have different probabilities of detection (e.g., Gompper et al. 2006, Long et al. 2007, Nichols et al. 2008). Unmodeled detection error in NGT surveys may underestimate species distributions and population sizes, and yield spurious conclusions about ecological relationships based on these data. Although some studies (Kendall et al. 2009, Sollmann et al. 2013) have used multiple data sources to assess and account for detection error, many do not. Independent validation and multi-method modeling provide an empirical lens through which to evaluate the accuracy of NGT estimates.

MANAGEMENT IMPLICATIONS

Noninvasive genetic tagging has been adopted by many management agencies to survey wildlife populations and

conduct ecological research. Sampling error represents a source of uncertainty that equates to management risk. Management actions based on imprecise or possibly inaccurate information may not provide the desired outcome for wildlife populations. Remote cameras that explicitly sample NGT hair traps allow managers to explicitly quantify and adjust for this error. Moreover, the added information from cameras allows an examination of animal behavior, including interactions with conspecifics and heterospecifics, and even the presence of young, which can provide estimates of reproductive rates. Although cameras are an added expense to NGT, using camera data to subsample hair data by genetically analyzing only those hair samples where cameras confirm the species' presence may more than compensate for the cost by savings in genetic analysis. Quantifying bias in occupancy estimates from NGT studies can provide more robust estimates of population parameters, which may increase our confidence in wildlife management actions based on noninvasive data.

ACKNOWLEDGMENTS

Alberta Innovates—Technology Futures (AITF), Natural Sciences and Engineering Research Council of Canada, MITACS, and University of Victoria provided funding to J.T.F. Data collection was funded by AITF; Government of Alberta, Parks Division; Alberta Environment and Sustainable Resource Development; Alberta Conservation Association; and Foundation for North American Wild Sheep. Thanks to M. Hiltz, L. Nolan, D. Pan, L. Peleshok, S. Newman, L. Roy, C. Twitchell, D. Vujnovic, K. Vujnovic, M. Wheatley, A. Fisher, L. Wilkinson, J. Gould, and many others. L. Chalifour at Galiano Conservancy Association provided graphics. D. MacKenzie at Proteus Wildlife Research Consultants lent helpful advice. C. Harris at Wildlife Genetics International (WGI) performed genetic analyses. D. Paetkau at WGI and D. Linden provided invaluable input on a previous version.

LITERATURE CITED

- Alberta Grizzly Bear Recovery Plan 2008–2013. 2008. Alberta Sustainable Resource Development, Fish and Wildlife Division, Alberta Species at Risk Recovery Plan No. 15. Edmonton, Alberta, Canada.
- Amstrup, S. C., T. L. McDonald, and B. F. J. Manly. 2005. Handbook of capture-recapture analysis. Princeton University Press, Princeton, New Jersey, USA.
- Anderson, D. R. 2008. Model based inferences in the life sciences: a primer on evidence. Springer, London, United Kingdom.
- Bailey, L. L., J. E. Hines, J. D. Nichols, and D. I. MacKenzie. 2007. Sampling design trade-offs in occupancy studies with imperfect detection: examples and software. *Ecological Applications* 17:281–290.
- Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach. Second edition. Springer, London, United Kingdom.
- Campbell, L. A., R. A. Long, and W. J. Zielinski. 2008. Integrating multiple methods to achieve survey objectives. Pages 223–237 in R. A. Long, P. MacKay, W. J. Zielinski, and J. C. Ray, editors. *Noninvasive survey methods for carnivores*. Island Press, Washington, D.C., USA.
- Fisher, J. T., B. Anholt, S. Bradbury, M. Wheatley, and J. P. Volpe. 2012. Spatial segregation of sympatric marten and fishers: the influence of landscapes and species-scapes. *Ecography* 36:240–248.

- Fisher, J. T., B. Anholt, and J. P. Volpe. 2011. Body mass explains characteristic scales of habitat selection in terrestrial mammals. *Ecology and Evolution* 1:517–528.
- Fisher, J. T., S. Bradbury, M. Wheatley, B. Anholt, L. Roy, J. P. Volpe, and L. Nolan. 2013. Wolverines on the Rocky Mountain slopes: natural heterogeneity and landscape alteration as predictors of distribution. *Canadian Journal of Zoology* 91:706–716.
- Fisher, J. T., M. T. Wheatley, and D. MacKenzie. 2014. Spatial patterns of breeding success of grizzly bears from multi-state hierarchical models. *Conservation Biology* 28:in press.
- Gardner, B., J. A. Royle, M. T. Wegan, R. E. Rainbolt, and P. D. Curtis. 2010. Estimating black bear density using DNA data from hair snares. *Journal of Wildlife Management* 74:318–325.
- Gompper, M. E., R. W. Kays, J. C. Ray, S. D. LaPoint, D. A. Bogan, and J. R. Cryan. 2006. A comparison of non-invasive techniques to survey carnivore communities in northeastern North America. *Wildlife Society Bulletin* 34:1142–1151.
- Hedmark, E., and H. Ellegren. 2007. DNA-based monitoring of two newly founded Scandinavian wolverine populations. *Conservation Genetics* 8:843–852.
- Hines, J. E. 2006. PRESENCE4-Software to estimate patch occupancy and related parameters. USGS-PWRC. <<http://www.mbr-pwrc.usgs.gov/software/presence.shtml>>. Accessed 20 May 2014.
- Johnson, W., and S. J. O'Brien. 1997. Phylogenetic reconstruction of the Felidae using 16S rRNA and NADH-5 mitochondrial genes. *Journal of Molecular Evolution* 44:S98–S116.
- Karanth, K. U., J. D. Nichols, N. S. Kumar, and J. E. Hines. 2006. Assessing tiger population dynamics using photographic capture-recapture sampling. *Ecology* 87:2925–2937.
- Kendall, K. C., and K. S. McKelvey. 2008. Hair collection. Pages 141–182 in R. A. Long, P. MacKay, W. J. Zielinski, and J. C. Ray, editors. *Noninvasive survey methods for carnivores*. Island Press, Washington, D.C., USA.
- Kendall, K. C., J. B. Stetz, J. Boulanger, A. C. Macleod, D. Paetkau, and G. C. White. 2009. Demography and genetic structure of a recovering grizzly bear population. *Journal of Wildlife Management* 73:3–17.
- Krohn, W. B., W. J. Zielinski, and R. B. Boone. 1997. Relations among fishers, snow, and martens in California: results from small-scale spatial comparisons. Pages 211–232 in G. Proulx, H. N. Bryant, and P. M. Woodard, editors. *Martes: taxonomy, ecology, techniques, and management*. Provincial Museum of Alberta, Edmonton, Alberta, Canada.
- Lindenmayer, D. B., and G. E. Likens. 2010. The science and application of ecological monitoring. *Biological Conservation* 143:1317–1328.
- Long, R. A., T. M. Donovan, P. MacKay, W. Zielinski, and J. Buzas. 2007. Comparing scat detection dogs, cameras, and hair snares for surveying carnivores. *Journal of Wildlife Management* 71:2018–2025.
- Long, R. A., and W. J. Zielinski. 2008. Designing effective noninvasive carnivore surveys. Pages 8–44 in R. A. Long, P. MacKay, W. J. Zielinski, and J. C. Ray, editors. *Noninvasive survey methods for carnivores*. Island Press, Washington, D.C., USA.
- MacKay, P., W. J. Zielinski, R. A. Long, and J. C. Ray. 2008. Noninvasive research and carnivore conservation. Pages 1–7 in R. A. Long, P. MacKay, W. J. Zielinski, and J. C. Ray, editors. *Noninvasive survey methods for carnivores*. Island Press, Washington, D.C., USA.
- MacKenzie, D. I., J. D. Nichols, G. B. Lachman, S. Droege, J. A. Royle, and C. A. Langtimm. 2002. Estimating site occupancy rates when detection probabilities are less than one. *Ecology* 83:2248–2255.
- MacKenzie, D. I., J. D. Nichols, J. A. Royle, K. H. Pollock, L. L. Bailey, and J. E. Hines. 2006. Occupancy estimation and modeling: inferring patterns and dynamics of species occurrence. Elsevier, New York, New York, USA.
- Magoun, A. J., C. D. Long, M. K. Schwartz, K. L. Pilgrim, R. E. Lowell, and P. Valkenburg. 2011. Integrating motion-detection cameras and hair snares for wolverine identification. *Journal of Wildlife Management* 75:731–739.
- Manel, S., M. Schwartz, G. Luikart, and P. Taberlet. 2003. Landscape genetics: combining landscape ecology and population genetics. *Trends in Ecology and Evolution* 18:189–197.
- McKelvey, K. S., and M. K. Schwartz. 2004a. Genetic errors associated with population estimation using non-invasive molecular tagging: problems and new solutions. *Journal of Wildlife Management* 68:439–448.
- McKelvey, K. S., and M. K. Schwartz. 2004b. Providing reliable and accurate genetic capture–mark–recapture estimates in a cost-effective way. *Journal of Wildlife Management* 68:453–456.
- Mills, L. S., J. J. Citta, K. P. Lair, M. K. Schwartz, and D. A. Tallmon. 2000. Estimating animal abundance using noninvasive data sampling: promise and pitfalls. *Ecological Applications* 19:283–294.
- Mowat, G., D. C. Heard, D. R. Seip, K. G. Poole, G. Stenhouse, and D. W. Paetkau. 2005. Grizzly *Ursus arctos* and black bear *Ursus americanus* densities in the interior mountains of North America. *Wildlife Biology* 11:31–48.
- Mowat, G., and D. Paetkau. 2002. Estimating marten *Martes americana* population size using hair capture and genetic tagging. *Wildlife Biology* 8:201–209.
- Nichols, J. D., L. L. Bailey, A. F. O'Connell, Jr., N. W. Talancy, E. H. C. Grant, A. T. Gilbert, E. M. Annand, T. P. Husband, and J. E. Hines. 2008. Multi-scale occupancy estimation and modelling using multiple detection methods. *Journal of Applied Ecology* 45:1321–1329.
- O'Brien, T. G., M. F. Kinnaird, and H. T. Wibisono. 2010. Estimation of species richness of large vertebrates using camera traps: an example from an Indonesian rainforest. Pages 233–252 in A. F. O'Connell, J. D. Nichols, and K. U. Karanth, editors. *Camera traps in ecology*. Springer, London, United Kingdom.
- O'Connell, A. F., J. D. Nichols, and K. U. Karanth. 2010. *Camera traps in ecology*. Springer, London, United Kingdom.
- O'Connell, A. F., N. W. Talancy, L. L. Bailey, J. R. Sauer, R. Cook, and A. T. Gilbert. 2006. Estimating site occupancy and detection probability parameters for meso- and large mammals in a coastal ecosystem. *Journal of Wildlife Management* 70:1625–1633.
- Paetkau, D. 2004. The optimal number of markers in genetic capture–mark–recapture studies. *Journal of Wildlife Management* 68:449–452.
- Rowcliffe, J. M., J. Field, S. T. Turvey, and C. Carbone. 2008. Estimating animal density using camera traps without the need for individual recognition. *Journal of Applied Ecology* 45:1228–1236.
- Sollmann, R., N. M. Törres, M. M. Furtado, A. T. de Almeida Jácomo, F. Palomares, S. Roques, and L. Silveira. 2013. Combining camera-trapping and noninvasive genetic data in a spatial capture–recapture framework improves density estimates for the jaguar. *Biological Conservation* 167: 242–247.
- Taberlet, P., L. Waits, and G. Luikart. 1999. Noninvasive genetic sampling: look before you leap. *Trends in Ecology and Evolution* 14:323–327.
- Tobler, M. W., S. E. Carrillo-Percastegui, R. L. Pitman, R. Mares, and G. Powell. 2008. An evaluation of camera traps for inventorying large and medium sized terrestrial rainforest mammals. *Animal Conservation* 11:169–178.
- Trolle, M., and M. Kéry. 2003. Estimation of ocelot density in the Pantanal using capture–recapture analysis of camera-trapping data. *Journal of Mammalogy* 84:607–614.
- Vitousek, P. M. 1997. Human domination of Earth's ecosystems. *Science* 277:494–499.
- Waits, J. L., and P. L. Leberg. 2000. Biases associated with population estimation using molecular tagging. *Animal Conservation* 3:191–199.
- Waits, L. P. 2004. Using noninvasive genetic sampling to detect and estimate abundance of rare wildlife species. Pages 211–228 in W. L. Thompson, editor. *Sampling rare or elusive species*. Island Press, Washington, D.C., USA.
- Walther, G.-R., E. Post, P. Convey, A. Menzel, C. Parmesan, T. J. Beebee, J.-M. Fromentin, O. Hoegh-Guldberg, and F. Bairlein. 2002. Ecological responses to recent climate change. *Nature* 416:389–395.
- Williams, B. W., D. R. Etter, D. W. Linden, K. F. Millenbah, S. R. Winterstein, and K. T. Scribner. 2009. Noninvasive hair sampling and genetic tagging of co-distributed fishers and American martens. *Journal of Wildlife Management* 73:26–34.
- Yoccoz, N. G., J. D. Nichols, and T. Boulinier. 2001. Monitoring of biological diversity in space and time. *Trends in Ecology and Evolution* 16:446–453.
- Zielinski, W. J., and N. P. Duncan. 2004. Diets of sympatric populations of American martens (*Martes americana*) and fishers (*Martes pennanti*) in California. *Journal of Mammalogy* 85:470–477.

Associate Editor: Andrew Kroll.