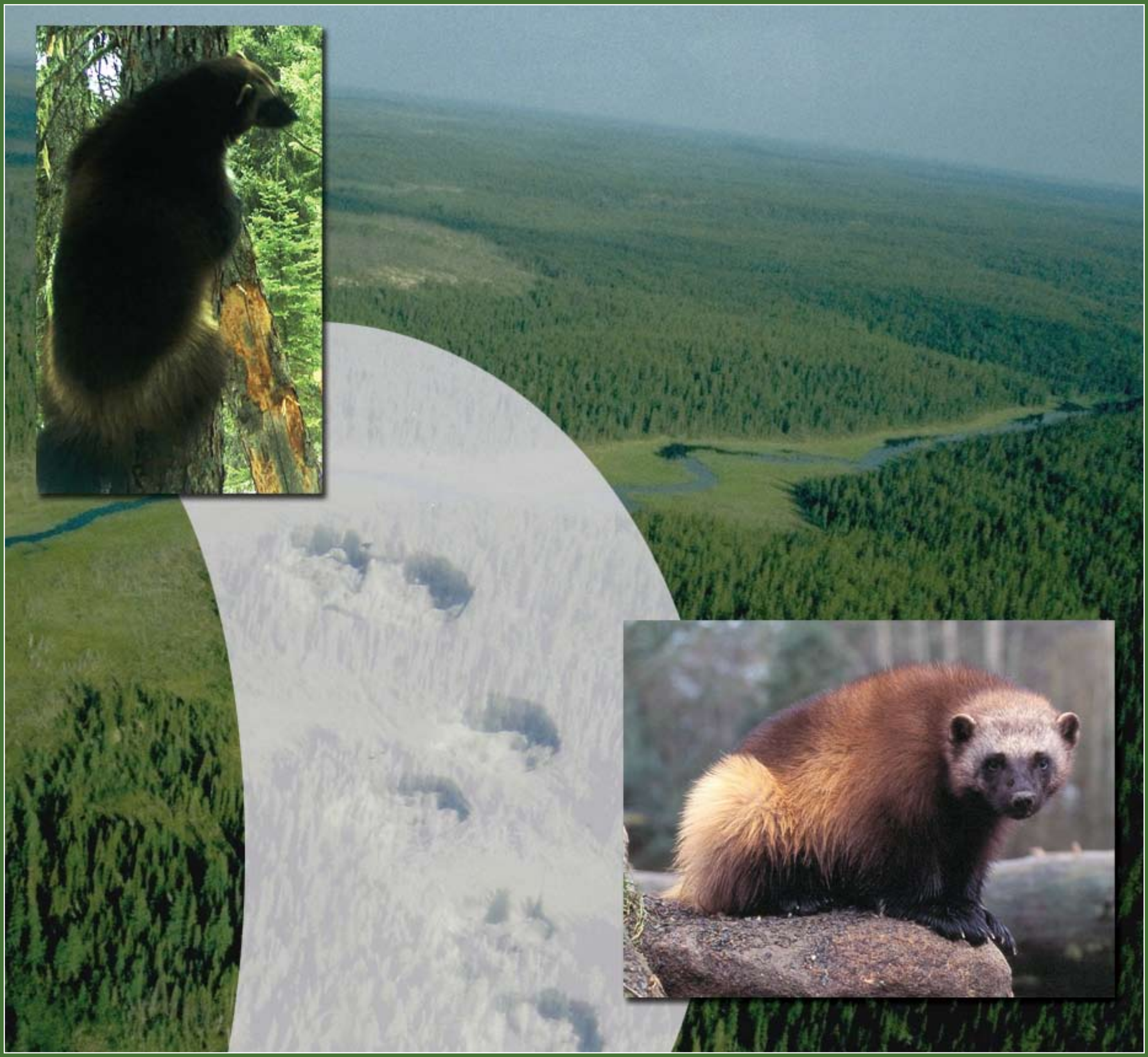

Surveying and Monitoring Wolverines in Ontario and Other Lowland, Boreal Forest Habitats:
Recommendations and Protocols

NWSI Field Guide FG-06

WILDLIFE CONSERVATION SOCIETY CANADA

Ontario

Surveying and Monitoring Wolverines in Ontario and Other Lowland, Boreal Forest Habitats:

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by

Erin L. Koen

Justina C. Ray

Jeff Bowman

F. Neil Dawson

Audrey J. Magoun

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Wildlife Conservation Society Canada
720 Spadina Avenue, Suite 600
Toronto, Ontario M5S 2T9 CANADA
Telephone: (416) 850-9038
www.wcscanada.org

Ministry of Natural Resources
Northwest Science and Information
RR 1, 25th Side Road
Thunder Bay, Ontario P7C 4T9 CANADA
Telephone: (807) 939-2501
http://nwsi.mnr.gov.on.ca

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Executive Summary

In April 2004, the Ministry of Natural Resources listed the wolverine (*Gulo gulo*) as “threatened” on the Species at Risk in Ontario list. This designation was assigned because the species’ range within the province had declined by ≥50%, human presence and resource development activity is increasing in areas where wolverines presently occur, and the low reproductive rates and large home range sizes of wolverines render populations slow to recover from the loss of many individuals. An important step in the implementation of a recovery strategy is the collection of population information through regular surveys and monitoring. Wolverines have large home ranges, low population densities, and occur in remote areas, making them a difficult species to study. Therefore, since 2002 the Ontario Boreal Wolverine Project has tested several methods for detecting wolverines at different spatial scales, including aerial track surveys, interviews with local trappers, hair snares, remote cameras, live-trapping, and radio telemetry.

In part I of this document, we review available techniques to survey and monitor wolverines at both large (>100,000 km^2) and small (<100,000 km^2) spatial scales. We address several survey objectives: estimating distribution, relative abundance, abundance, and monitoring population changes over time. Based on our work on wolverines in Ontario, we have provided recommendations for survey techniques according to scale and objective.

At both large and small spatial scales, we recommend hierarchical spatial modeling (HSM) based on aerial snow track surveys to estimate wolverine distribution and relative abundance. Additionally, this technique can be used to monitor changes in wolverine distribution and relative abundance over time. At small spatial scales, we also recommend ground-based snow track surveys to estimate wolverine relative abundance. For estimating wolverine abundance at small scales, we recommend capture-recapture methods using hair snares.

In part II, we provide protocols and logistical considerations for managers of wolverine populations who would like to implement the recommended techniques: ground-based snow track surveys and hair snares for small-scale surveys, and HSM aerial snow track surveys for both large- and small-scale surveys. We outline budget and effort considerations, and provide a step-by-step protocol for data collection, data management, and analysis.

This document is aimed at those responsible for monitoring wolverine populations as part of recovery efforts or in conducting research on this elusive animal. The recommendations and protocols are intended as survey methods for wolverines and other wide-ranging species in lowland, boreal forest habitats.

Sommaire

En avril 2004, le ministère des Richesses naturelles a ajouté le carcajou (*Gulo gulo*) à la Liste des espèces en péril en Ontario. Cette désignation d'espèce « en péril » a été attribuée en raison de la diminution de ≥50 % de l'aire de distribution de l'espèce à l'intérieur de la province, de l'intensification de la présence humaine et des activités de développement des ressources dans les régions déjà affectées et parce que les faibles taux de reproduction et la grande étendue des domaines vitaux des carcajous font en sorte que les populations se rétablissent lentement à la suite de la perte de nombreux individus. Une étape importante dans la mise en œuvre d'une stratégie de rétablissement est la collecte de données sur les populations à intervalles réguliers par des levers et des observations. Les carcajous occupent un domaine vital étendu, leur densité de population est faible et ils habitent des régions éloignées, ce qui en complique l'étude. En conséquence, depuis 2002, l'Ontario Boreal Wolverine Project met à l'essai diverses méthodes de détection du carcajou à différentes échelles spatiales, dont des levers aériens de pistes, des entrevues de trappeurs locaux, la collecte de poils au collet, l'utilisation d'appareils photos à déclenchement par télécommande, le trappage d'animaux vivants et la radiotélémesure.

Dans la première partie de ce document, nous passons en revue les techniques disponibles pour étudier et observer le carcajou à de grandes (>100 000 km^2) et petites (<100 000 km^2) échelles spatiales. Nous y couvrons plusieurs objectifs d'étude: évaluation de la distribution, de l'abondance relative et de l'abondance, et observation de l'évolution démographique au fil du temps. Sur la base de nos travaux menés sur les carcajous en Ontario, nous recommandons diverses techniques d'étude selon l'échelle et les objectifs.

À grande ou à petite échelle spatiale, nous recommandons la modélisation spatiale hiérarchique (MSH) à partir de levers aériens de pistes dans la neige pour évaluer la distribution des carcajous et leur abondance relative. De plus, cette technique peut être utilisée pour suivre l'évolution de la distribution et l'abondance relative des carcajous au fil du temps. À de petites échelles spatiales, nous recommandons également l'observation au sol de pistes dans la neige pour évaluer l'abondance relative des carcajous. Aux fins d'évaluer l'abondance des carcajous à de petites échelles, nous recommandons le recours à des méthodes de recensement par capture et recapture utilisant la collecte de poils au collet.

Dans la deuxième partie, nous présentons des protocoles et des considérations logistiques aux gestionnaires de populations de carcajous qui aimeraient mettre en œuvre les techniques recommandées: observation au sol de pistes dans la neige et collecte de poils au collet dans le cas d'études à petite échelle; MSH de levers aériens de pistes dans la neige dans le cas d'études à petite ou à grande échelle. Nous tenons compte des budgets et des efforts requis par chacune et présentons un protocole par étape pour la collecte, la gestion et l'analyse de données.

Ce document est à l'intention des personnes chargées d'observer des populations de carcajous dans le cadre d'efforts de rétablissement ou de mener de la recherche sur cet animal fugace. Les recommandations et les protocoles doivent servir de méthodes pour étudier le carcajou et d'autres espèces à distribution étendue habitant les basses terres de la forêt boréale.

1.0 Introduction

1.1 The need to survey and monitor wolverines

Wolverines (*Gulo gulo*) are regarded as one of the most rare and least studied mammalian carnivores in North America (Banci 1994, Ruggiero *et al*. 1994, Weaver *et al*. 1996). Recent work, however, has increased our knowledge of wolverine ecology substantially (Persson 2007, Ruggiero *et al*. 2007), thereby providing us with a renewed opportunity to focus on wolverine management and conservation. Wolverines occur in North America and Eurasia, and their global conservation status has been assessed as vulnerable on the International Union for the Conservation of Nature and Natural Resources (IUCN) Red List (Mustelid Species Group 1996). Historically, wolverines ranged across most of Canada, excluding Newfoundland, Prince Edward Island, and Nova Scotia (Hash 1987, COSEWIC 2003; Figure 1). In the United States, wolverines historically occupied many of the northern states, with western populations occurring as far south as California, Utah, and Colorado (Hash 1987, COSEWIC 2003, Aubry *et al*. 2007). Verifiable data on the historical range is sparse; according to Laliberte and Ripple (2004), wolverines currently occupy roughly 63% of their historical range in North America, but the extent of the historical range is uncertain (Aubry *et al*. 2007, Slough 2007).

In the United States, wolverines are currently present only in Montana, Idaho, Washington, northwest Wyoming, and Alaska (Aubry *et al*. 2007). In Canada, two disjunct populations are recognized (COSEWIC 2003). The eastern population occupies northern Quebec and Labrador, and is currently listed as endangered. There have been no confirmed records of wolverines in Quebec since 1978 (Fortin *et al*. 2005), although there have been unconfirmed sightings (Slough 2007). An aerial snow track survey in 2004 found no evidence of wolverines in Labrador (Schmelzer 2005). A national recovery plan for the eastern wolverine population is now in place (Fortin *et al*. 2005). The western population occurs in Ontario, Manitoba, Saskatchewan, Alberta, and throughout much of British Columbia, Yukon, Northwest Territories, and Nunavut (Slough 2007). The western population of wolverines has been listed as a species of special concern since 1982, with the most recent reassessment conducted in 2003 (COSEWIC 2003).

In Ontario, wolverines historically occurred throughout much of the province (Hall and Kelson 1959, Banfield 1974). However, their range has receded northward since the latter half of the 1800s (Van Zyll de Jong 1975, Dawson 2000). The disappearance of wolverines in southern Ontario occurred around the same time as human settlement, logging, and railroad construction increased in the same area, though it is impossible to confirm the direct cause of the historical wolverine population decline. Currently, wolverines are primarily confined to northwestern Ontario, outside of roaded and managed areas of the province (Figure 2). The Ontario wolverine population has a relatively low density (Van Zyll de Jong 1975, Dawson 2000, COSEWIC 2003, Slough 2007, Ontario Wolverine Recovery Team, in prep.) and was listed as threatened on the Species at Risk in Ontario list in April 2004. A recovery strategy for wolverines in Ontario is currently in progress.

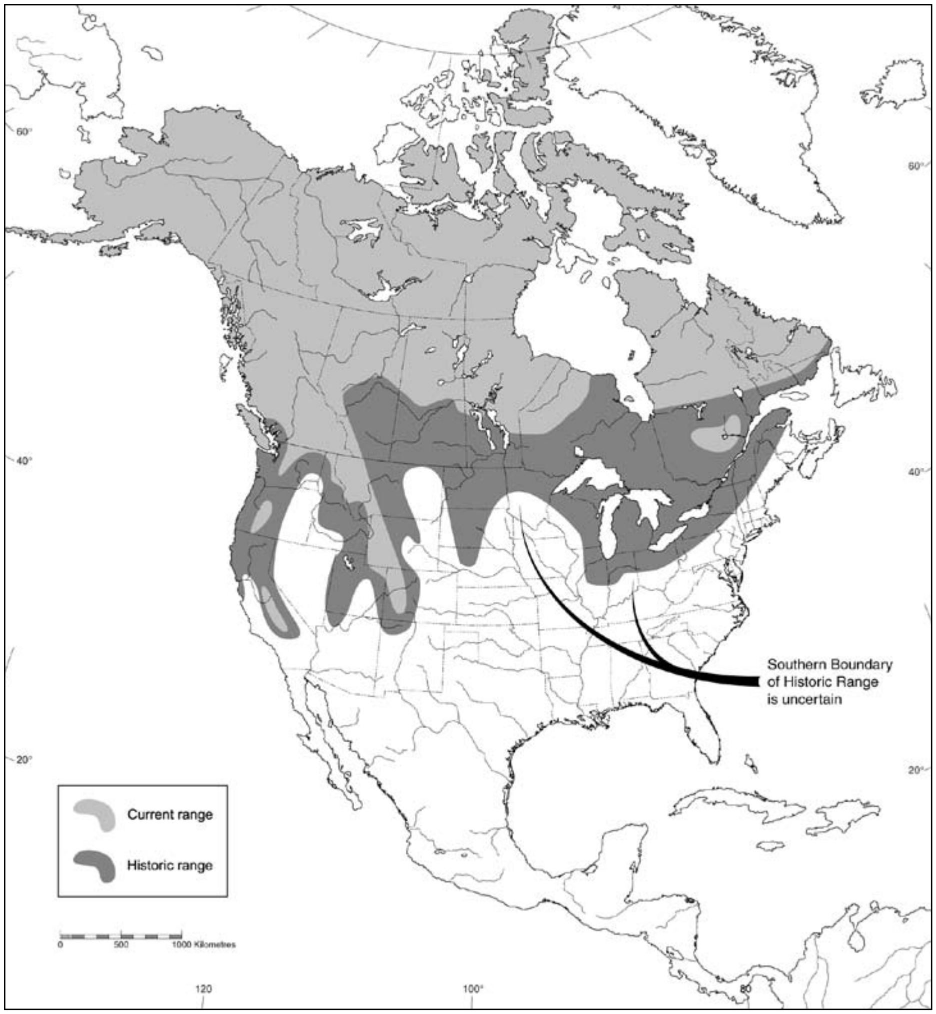
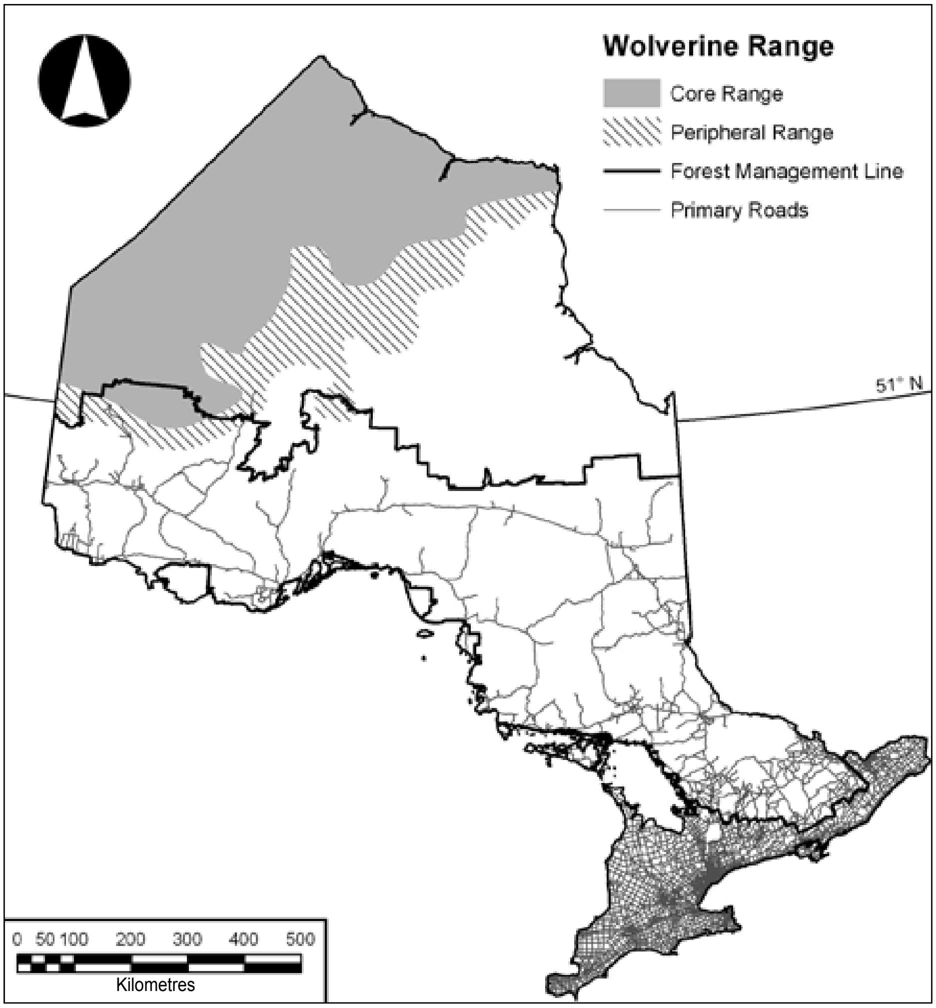


Figure 1. Estimated wolverine (*Gulo gulo*) distribution in North America (reproduced from COSEWIC [2003]).

Wolverines have life-history characteristics that contribute to their low intrinsic ability to recover and repopulate areas where they have been extirpated, including low population density, large home ranges, and low productivity (Weaver *et al*. 1996; Banci & Proulx 1999). These factors render wolverine populations particularly vulnerable to human-caused mortality, which is likely additive to natural mortality (Krebs *et al*. 2004). Indeed, Krebs *et al*. (2004) suggested that wolverine harvest is not sustainable without immigration from neighbouring, unharvested populations. Detailed discussions of biological and environmental factors that might contribute to wolverine decline and limit natural recovery are found in Dawson (2000) and Fortin *et al*. (2005).

There is little definitive empirical evidence linking wolverine population decline to any direct or indirect cause, due to the limited amount of study in areas subject to active resource development, a condition that characterizes much of present-day wolverine range. Given the combined lack of knowledge regarding a mechanism for population decline and the relatively low resilience of wolverine populations (Weaver *et al*. 1996), more research is essential, particularly as human development activities encroach into wolverine population strongholds. An important first step is the collection of baseline population information through population surveys and monitoring.



Map prepared by Wildlife Conservation Society Canada. Data Source: Ontario Boreal Wolverine Project, a collaboration between the Ontario Ministry of Natural Resources, Wildlife Conservation Society Canada and The Wolverine Foundation, Inc.

Figure 2. Current wolverine range in Ontario, Canada.

1.2 Ontario boreal wolverine project and wolverine recovery in Ontario

In view of the probable low wolverine population density in Ontario relative to populations farther west (Van Zyll de Jong 1975, COSEWIC 2003, Slough 2007), the planned northward shift of human development and uncertain future of suitable habitat, and the paucity of baseline ecological data on wolverines in Ontario, we began a large-scale, multi-faceted project to address issues associated with surveying and monitoring wolverines in Ontario. Since 2002, we have tested various methods for detecting wolverines at different scales, including aerial track surveys, interviews with local trappers, hair snares, remote cameras, live-trapping, and radio telemetry. These data represent the first step in developing a rigorous monitoring program for wolverines from which to base recovery actions and monitor their success. Such a monitoring program is not only applicable to wolverines in Ontario, but also to other wide-ranging species in similar lowland, boreal forest habitats (e.g. woodland caribou [*Rangifer tarandus caribou*]).

1.3 Challenges of surveying and monitoring wolverines

Surveying and monitoring wolverines pose a challenge for wildlife managers because wolverines are rare, elusive, and occur over large, remote areas. Common methods for estimating population abundance, such as capture-recapture, prove difficult at any spatial scale because of the need for large samples to produce reliable results, which is a logistically difficult enterprise for species that occur at low population densities. Furthermore, wolverines have large home ranges: up to 2,034 km^2 for adult males in Ontario (over a nine-month period; Dawson *et al*. submitted) and an average of 754 km^2 for adult females in the Greater Yellowstone area (Inman *et al*. 2003). Thus, surveys must cover a large area in order to include a sufficient number of animals to produce statistically sound results. Moreover, because of the challenge of detecting wolverines, considerable effort is required to ensure a high probability of detecting the species. These difficulties are compounded by the fact that wolverines tend to occupy remote areas, making most surveys logistically difficult to implement. Almost half of Ontario above the managed forest boundary (ca. 450,000 km^2), for example, is characterized by these conditions.

1.4 Document goals

We intend for this manual to be used by wildlife managers and conservationists faced with the task of surveying and monitoring wolverines in lowland, boreal forests. We provide a critical review of available techniques, and assessment of the viability of these techniques for the study of wolverines in Ontario and similar lowland, boreal forest habitats. This assessment is based on our study on wolverines in Ontario, so that managers can benefit from the results of our recent work. Finally, we provide recommendations and protocols of techniques for surveying and monitoring wolverines so that managers can implement these techniques in the context of their own wolverine monitoring programs. This theoretical and practical foundation should prove useful for wolverine management and recovery in Ontario and other lowland boreal forests, and potentially for managers of other rare and elusive mammals in Ontario and elsewhere. It is not an in-depth review of wolverine biology; comprehensive reviews can be found in Hash (1987), Banci (1994), Dawson (2000), COSEWIC (2003), and Fortin *et al*. (2005).

There are two parts to this manual: reviews and recommendations, and protocols. Part I is structured around the spatial scale of the study objective: large or small scale. Within each of these divisions, we have reviewed, assessed, and discussed the techniques appropriate for addressing population questions at each scale. We provide an assessment of the usefulness of each technique and recommendations for wolverine surveys based on results from the Ontario Boreal Wolverine Project. In part II, we provide protocols and logistical considerations for managers of wolverine populations who would like to implement the recommended techniques. We outline budget and effort considerations, and provide a step-by-step protocol for data collection, data management, and analysis.

Part I:

Review and Recommendations

2.0 Defining Study Objectives

The first step in the design of an effective survey is to have a clear definition of the study objectives; only then can one select the appropriate survey design (Yoccoz *et al*. 2001, MacKenzie and Royle 2005). For the purposes of this document, we define four survey objectives (Long and Zielinski 2008): distribution, relative abundance, abundance and population density, and monitoring over time. The following discussions focus primarily on collecting and analyzing presence-absence and capture-recapture data from surveys of mammalian forest carnivores in general, and wolverines in particular.

2.1 Distribution

A common objective for wildlife managers is to detect whether a species is present in an area or to document the extent of a species' distribution. In the latter case, the study area is often too large to survey in its entirety, so presence-absence surveys are conducted for a sample of units and the results are extrapolated to the whole study area (Box 1). The unit of measure is the proportion of the sample units occupied, or the probability of a particular sample unit being occupied (Long and Zielinski 2008).

Box 1. Terminology.

For the purposes of this document, we have used the following terminology:

- the *study area* (Figure 3a) is an arbitrarily defined area chosen by the investigators, or defined by the animal population in question (discussed in Box 3).
- a *sample unit* (Figure 3b) refers to a plot within the study area that may be surveyed once or repeatedly.
- a *sample station* (Figure 3c) refers to one or several stations within a sample unit where animals are sampled or detected using devices such as a hair snare or remote camera.
- a *season* refers to a period of time during which several repeat surveys are conducted. For example, a study covers two seasons (or years, in this example) if several repeat surveys are conducted in a sample unit in April, and several more repeat surveys are conducted in the same sample unit the following April.

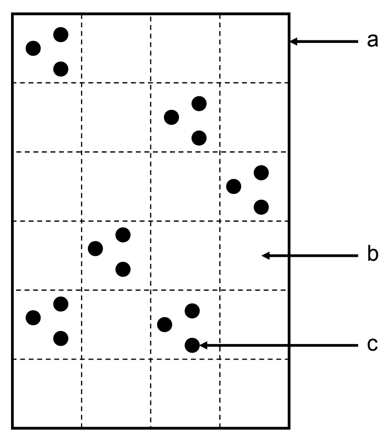


Figure 3. An example of the distinction between a) survey area, b) sample unit, and c) sample stations, for the purposes of this document.

Many of the survey methods that we will review in sections 4.0 and 6.0 are based on presence-absence data (i.e. tracks, hair snares, remote cameras). Whether the presence-absence data are used to estimate distribution, relative abundance, or used in monitoring, the inherent issues are the same: how does one know when a species is truly absent, and how does one allocate limited resources to maximize precision of the occupancy estimate and power to detect changes in population size over time?

Presence of a species can often be confirmed at a location, but absence is difficult or impossible to confirm with certainty. Non-detection of a species at a location could mean that the species was truly not present, or the species was present but the survey failed to detect it. The probability of detecting a species that is in fact present can be estimated by repeatedly surveying the sample unit in a relatively short time frame, as the probability of concluding a false-absence decreases with the number of repeat surveys (MacKenzie 2005). Because failure to account for false-absences underestimates occupancy, MacKenzie *et al*. (2002) developed a model to estimate the proportion of sample units occupied that incorporates the probability of detecting the species. We refer the reader to MacKenzie *et al*. (2002) and MacKenzie *et al*. (2006) for further detail.

2.2 Relative abundance

Measures of relative abundance generally use the rate of detection of animal sign as an index of true abundance. Sign such as tracks, hair, scat, or photographs can be used. Similarly, the number of animals harvested (Wood and Odum 1964, Gompper and Hackett 2005), or the proportion of sample units occupied (MacKenzie and Nichols 2004), have been used as correlates of abundance. The assumption is that there is a positive, linear relationship between rate of detection and abundance: if there are more animals in a population, they will leave more sign, be harvested more often, and occupy more area. Several studies have compared independent estimates of abundance and relative abundance; the general consensus is that indices of abundance do not translate into abundance, but can be useful for identifying trends in population size over time (Travaini *et al*. 1996, Sargeant *et al*. 1998, Sadlier *et al*. 2004, Lynch *et al*. 2006, Ray and Zielinski 2008). Additionally, several studies have simultaneously compared different indices of population abundance, and found that some techniques are more efficient than others, depending on the species of interest (Choate *et al*. 2006, Gompper *et al*. 2006, Barea-Azcon *et al*. 2007). Indeed, Conn *et al*. (2004) do not recommend count statistics as indices of population size unless the probability of detection is the same between populations being compared, or simulations are conducted to evaluate the effect of differences in detection probability on population estimates.

2.3 Abundance and density

Direct estimates of the number of individuals in a population (abundance) or the number of individuals per unit area (density) are considered preferable to estimates of relative abundance because they do not rely upon assumptions about the relationship between the index value and the actual population size (Long and Zielinski 2008). Capture-recapture methods are most common for obtaining abundance estimates (Long and Zielinski 2008).

The most basic capture-recapture technique, the Lincoln-Petersen model, involves “capturing” and marking a segment of the population, and releasing them back into the population. A second “capturing” event records the number of marked and unmarked individuals. The general assumption is that the ratio of marked to unmarked individuals in the second capturing event is equal to the ratio of individuals marked in the first capture event to the (unknown) size of the population. There have been many extensions to this basic model, and those that apply specifically to non-invasive genetic sampling are reviewed in Lukacs and Burnham (2005). Capture-recapture methods require individual identifications of animals, either by pelage, external markers (such as ear tags), or DNA. Surveys to collect such information can be invasive (live-trapping and tagging), non-invasive (using DNA from hair or scat, or photographs from camera traps), or a combination of both. Indeed, "capturing" does not necessarily mean physical capture; animals can be "captured" by leaving hair or scat, or by being photographed. For population estimates to be accurate and precise, many repeat surveys are necessary (for high capture probabilities), and a relatively large number of animals needs to be captured more than once (Otis *et al*. 1978). Bartmann *et al*. (1987) recommended marking >45% of the population when using Lincoln-Petersen models. Because considerable effort is required to capture and recapture a large number of animals with effort distributed evenly across the study area, these methods are better suited for studies at small spatial scales (section 6.0). We review assumptions for capture-recapture models in Box 2.

Some work has focused on devising methods to estimate population abundance that do not rely upon identification of individuals. For example, Royle and Nichols (2003) proposed a technique that takes advantage of the relationship between abundance and detectability at repeatedly sampled sites to estimate abundance, although this method has not yet been used extensively (Long and Zielinski 2008). Zhou and Griffiths (2007) also proposed a method to estimate abundance from presence-absence data that appears to work well for aggregated populations. Becker (1991) and Becker *et al*. (1998) proposed methods to estimate abundance without identifying individuals (section 6.1.1), although their methods still require that snow tracks from one individual be distinguished from those of others.

Population density estimates can be obtained from abundance estimates when the size of the area that bounds the population in question, or the effective sampling area, is known. If baits or lures are used for attracting animals to the sample unit, it is generally not possible to estimate the size of the study area, as animals could move great distances if the bait is detected from far away. Common approaches for estimating the effective sampling area are reviewed in Box 3.

2.4 Monitoring over time

Population monitoring is an attempt to detect changes in population abundance over time. Regression analyses are often used to estimate the slope of population size estimates over time (Harris 1986, Gerrodette 1987, Joseph *et al*. 2006); if the slope is significantly different than zero, the null hypothesis of no change in population size over time can be rejected. Population size estimates can be obtained from direct abundance (e.g. capture-recapture) or relative abundance estimates (e.g. count, presence-absence, harvest data, probability of occurrence, etc.). Range extent or distribution can also be used as a metric for monitoring purposes.

A priori power analysis is essential to the design of successful monitoring programs, as the study must be sensitive enough to detect real, biologically significant changes in distribution, relative abundance, or abundance (Zielinski and Stauffer 1996, Gibbs *et al*. 1999, Legg and Nagy 2006). Statistical power, or the probability of detecting a change that has, indeed, occurred, is related to a variety of factors that are explained in turn below: variability in the index, both between and within sample units; the number of seasons or time periods that are being compared; the effect size, or magnitude of population change; and type I error rate (α).

Box 2. A review of closed-population capture-recapture models.

Closed-population capture-recapture models assume that the population is geographically (no immigration or emigration) and demographically (no births or deaths) closed between sampling events (Otis *et al*. 1978). Therefore, the study area should be large enough and repeat surveys should be completed in a relatively short period of time in order to reduce the chance of violating these assumptions. Closed population capture-recapture models also assume that all individuals are available for capture, and all have the same capture and detection probabilities within each sampling occasion (Otis *et al*. 1978). This latter assumption is generally difficult to meet because individuals in some demographic groups may be more likely to be "trapped" (Larrucea *et al*. 2007), the effectiveness of baits can vary over time, placement of traps can affect accessibility (i.e. traps placed in the center of a territory are less likely to be visited by multiple individuals than traps set near the periphery [Larrucea *et al*. 2007]), and detectability for one individual may vary between sampling occasions (i.e. the individual may become more or less difficult to recapture). Finally, capture-recapture methods assume that individuals are uniquely and permanently marked and all previously "captured" individuals can be distinguished from unmarked individuals (Otis *et al*. 1978).

Capture-recapture methods generally consist of an initial marking period, followed by several sampling sessions. The primary data to be analyzed include the capture history for each individual (a series of ones and zeros indicating whether the individual was detected or not detected, respectively, for each sampling session), and the number of "new captures" during each recapture occasion as compared to the number of "recaptures".

Program CAPTURE (Table 1; Otis *et al*. 1978) has been used extensively to estimate the population abundance of closed populations from remote camera surveys (e.g. Karanth 1995, Karanth and Nichols 1998, Henschel and Ray 2003, Silver *et al*. 2004, Jackson *et al*. 2006), hair snare surveys (Mowat and Strobeck 2000, Waits and Leberg 2000, Boulanger *et al*. 2004), and scat surveys (Eggert *et al*. 2003, Flagstad *et al*. 2004). A summary of capture histories is inputted into the software, which produces an abundance estimate, standard error and 95% confidence intervals on the estimate, and capture probabilities. The program tests for population closure, though there are other programs (CLOSURE; Table 1) that will also test this assumption. Program CAPTURE evaluates the fit of several models, such as models that account for differences in capture probabilities between individuals, differences in capture probabilities between sampling occasions for the same individual, and interactions between these variables. Program MARK (Table 1) has also been used for abundance estimates from capture-recapture data (Boulanger and McLellan 2001, Wasser *et al*. 2004, Bellemain *et al*. 2005, Mulders *et al*. 2007).

Violation of the closure assumption. Capture-recapture studies are often designed with capture stations located within sample units so that individuals have a relatively equal probability of encountering and being captured by a trap. Sample unit size is usually no smaller than the smallest home range to further homogenize capture probabilities between individuals (Kendall and McKelvey 2008). When resources are limited (i.e. a finite number of trap stations can be set and maintained), investigators must choose between a small study area (with small grid cells) and a large study area (with large grid cells). The former scenario will have higher capture probabilities and more precise population estimates (Boulanger and McLellan 2001, Boulanger *et al*. 2002, 2004). However, the probability of violating the assumption of geographic closure is also higher with a smaller study area (Boulanger and McLellan 2001, Boulanger *et al*. 2002, 2004). Kendall (1999) assessed the robustness of closed-population capture-recapture models to violations of this assumption and found that population estimates were biased unless movement in and out of the study area was completely random.

Box 3. Estimating sampling area for population density estimates.

For population density estimates, investigators must have an estimate of the area sampled (effective study area). Here, we review several common methods for estimating effective study area size. Karanth and Nichols (1998) estimated the size of the area that they surveyed using remote cameras with the mean maximum distance moved (MMDM) technique. This technique was also used by Karanth *et al*. (2004) and Jackson *et al*. (2006). The area surveyed was equal to a polygon around the outermost trap locations plus a boundary strip around the perimeter of the trap-grid, since animals residing outside of the grid are also available to be trapped. The MMDM was estimated from animals that were trapped more than once at different traps; the width of the boundary strip was one-half of the MMDM between recaptures across individuals.

Silver *et al*. (2004) used a similar method to estimate effective study area; they generated a circular buffer around each trap, with a radius equal to one-half of the MMDM between camera trap recaptures across individuals. They merged the area of all of the buffers as an estimate of the area sampled. Sweitzer *et al*. (2000)'s method for estimating effective study area size was similar to Silver *et al*. (2004)'s method except that they used a range of buffer radii estimated from the range of home range sizes obtained from radio-collared individuals.

Soisalo and Cavalcanti (2006) compared four estimates of effective study area size for jaguar (*Panthera onca*) population density estimates. Buffer strips were estimated using one-half of the MMDM estimated from camera traps, full MMDM estimated from camera traps, actual MMDM estimated from a sample of radio-collared individuals, and home ranges estimated from radio-collared individuals. They found that estimates using one-half of the MMDM from camera traps underestimated the MMDM by jaguars and led to overestimates of population density. They recommended using radio-telemetry data to calculate suitable buffer strip widths for jaguar density estimates.

Variability in the index of abundance (or direct estimate of abundance) *between* sample units is a function of heterogeneity in the population distribution (Link *et al*. 1994), and can be reduced by increasing the number of sample units surveyed each year. Alternatively, variability within sample units is a function of imperfect detection and short-term temporal variation (Link *et al*. 1994), and can be reduced by increasing the number of repeat surveys within a season. Both types of variability will contribute to the power of a monitoring study; however, there is a trade-off between them when resources are limited. Fortunately, several simulation studies have addressed this issue and concluded unanimously that in a population monitoring context, power to detect a population decline is increased when the number of sample units is increased more so than the number of repeat surveys per sample unit (Gerrodette 1987, Kendall *et al*. 1992, Beier and Cunningham 1996, Zielinski and Stauffer 1996, Strayer 1999, Field *et al*. 2005, Mackenzie 2005, Joseph *et al*. 2006). At the same time, however, investigators must keep in mind that more than one survey per sample unit should be completed in order to estimate the probability of detection (section 2.1): MacKenzie (2005) recommended greater than three repeat surveys per season when monitoring over multiple seasons. Rhodes *et al*. (2006), using simulation, found that the power to detect declines in occupancy over time was maximized when survey efforts were directed toward high-quality habitats. However, this recommendation is contingent upon the knowledge of what high-quality habitat is and where it is located.

The length of the monitoring period also affects the power of the analysis: increasing the number of seasons or time periods will increase the power to detect population change (Harris 1986, Gibbs *et al*. 1998, Mackenzie 2005, Bailey *et al*. 2007). This does, however, come at a cost, as increasing the number of seasons necessarily means decreasing the number of sample units surveyed and number of repeat surveys when resources are limited (MacKenzie 2005, Bailey *et al*. 2007). Bailey *et al*. (2007) have designed

Estimate	Software	Description	Reference
SUPE	SUPEPOP	Estimates population size from aerial survey data collected using Becker *et al*. (1998)'s protocol.	Becker *et al*. (1998)[1]
HSM	openBUGS	Executes the Markov Chain Monte Carlo routine to estimate probability of occurrence.	Magoun *et al*. (2007a)[2] Banerjee *et al*. (2004) Royle *et al*. (2007)
Occupancy	PRESENCE	Estimates the probability that a site is occupied given uncertain detection.	MacKenzie *et al*. (2003)[3]
	GENPRES	Explores trade-offs of spatial and temporal allocation of sampling effort.	Bailey *et al*. (2007)[4]
Monitoring	MONITOR	Estimates the power of monitoring programs.	Gibbs *et al*. (1998)[5]
	G*Power	Estimates statistical power.	Faul *et al*. (2007)[6]
Abundance (capture-recapture)	MARK	Provides parameter estimation for capture-recapture data.	White and Burnham (1999)[4]
	CAPTURE	Estimates capture probability and population size for a closed population.	Otis *et al*. (1978)[4] Rextad and Burnham (1991)
	CLOSURE	Tests for population closure.	Stanley and Burnham (1999)[7]
	CAPWIRE	Estimates abundance from a single sample-session with capture heterogeneity.	Miller *et al*. (2005)[8]

[1] ftp://ftpr3.adfg.state.ak.us/MISC/PROGRAMS/SUPEPOP/
[2] www.mathstat.helsinki.fi/openbugs/
[3] http://www.proteus.co.nz
[4] http://www.mbr-pwrc.usgs.gov/software/
[5] http://www.mbr-pwrc.usgs.gov/software/monitor.html
[6] http://www.psycho.uni-duesseldorf.de/abteilungen/aap/gpower3/
[7] www.mesc.usgs.gov/products/software/clostest/clostest.asp
[8] www.cnr.uidaho.edu/lecg/

Table 1. Available software for estimating distribution, relative abundance, and abundance of wildlife populations.

software (program GENPRES; Table 1) that weighs the costs and benefits of altering the number of sample units, number of repeat surveys, and length of the monitoring period (number of seasons).

The effect size, or magnitude of population change, is a further consideration with respect to statistical power: large changes in abundance are easier to detect than small changes (Kendall *et al*. 1992, Zielinski and Stauffer 1996, Strayer 1999). If it is necessary to detect small changes in population abundance, it will generally require more survey effort. As such, it is critical that *a priori* power analyses are conducted to make sure that adequate effort is expended on the survey from the beginning. It is entirely possible, for example, that given index variation, it will not be feasible to detect small effects (Field *et al*. 2007), in which case establishing a monitoring program with such a goal would be futile. Strayer (1999) found that presence-absence data has low power to detect declines of <20–50%, especially when the species was rare, distribution was patchy, or population declines were uniform over the study area rather than distinct local extinction events. Similarly, Zielinski and Stauffer (1996) used simulations to show that the power to detect population index change was lower for species that occupied a small proportion of sampling units.

Finally, power to detect population change is affected by the value chosen for α. The type I error rate (α) is the probability of rejecting the null hypothesis when it is true (i.e. there is no change in the population size, yet a change is detected). Likewise, the type II error rate (β) is the probability of failing to reject the null hypothesis when it is false (i.e. there is a change in population size, but it is not detected). Typically, the type I error rate is arbitrarily set at 0.05 (i.e. the test has a 5% chance of detecting a change that does not exist) and the type II error rate is set at 0.20 (i.e. the test has an 80% chance of detecting a change that is, in fact, present). The power of a test can be increased by increasing the type I error rate (i.e. increasing the chance of rejecting the null hypothesis of no population change). Field *et al*. (2005, 2007) argue that, in a population monitoring context, α and β should be equal. The consequences of making a type II error are much greater than a type I error: in the former, the result is potential extinction, whereas in the latter, the result is generally unnecessary additional management (Beier and Cunningham 1996, Zielinski and Stauffer 1996, Legg and Nagy 2006). Beier and Cunningham (1996) and Strayer (1999) suggest increasing α to 0.1–0.25 for the purposes of population monitoring.

As an alternative to regression analyses, MacKenzie *et al*. (2003) extended the general model for estimating the proportion of sites occupied described in MacKenzie *et al*. (2002) to population monitoring. Given detection histories for sample units, this method estimates site occupancy, colonization, and local extinction probabilities. The model assumes that there is constancy across sites at any one time for the following three parameters: the probability that a site is occupied, the probability that the species is detected there, and the probability of colonization or local extinction. However, some of these assumptions can be relaxed if appropriate covariates, such as habitat differences, are incorporated into the model (MacKenzie *et al*. 2003). Software (program PRESENCE; Table 1) is available for this type of data analysis.

3.0 Defining Scale

Defining both the temporal and spatial scale of the study and understanding the potential trade-offs inherent in the multiple available options are important in the design of population surveys and monitoring programs (MacKenzie and Royle 2005). When it comes to designing surveying and monitoring programs for wolverines, there are several key questions that wildlife managers ask, outlined below.

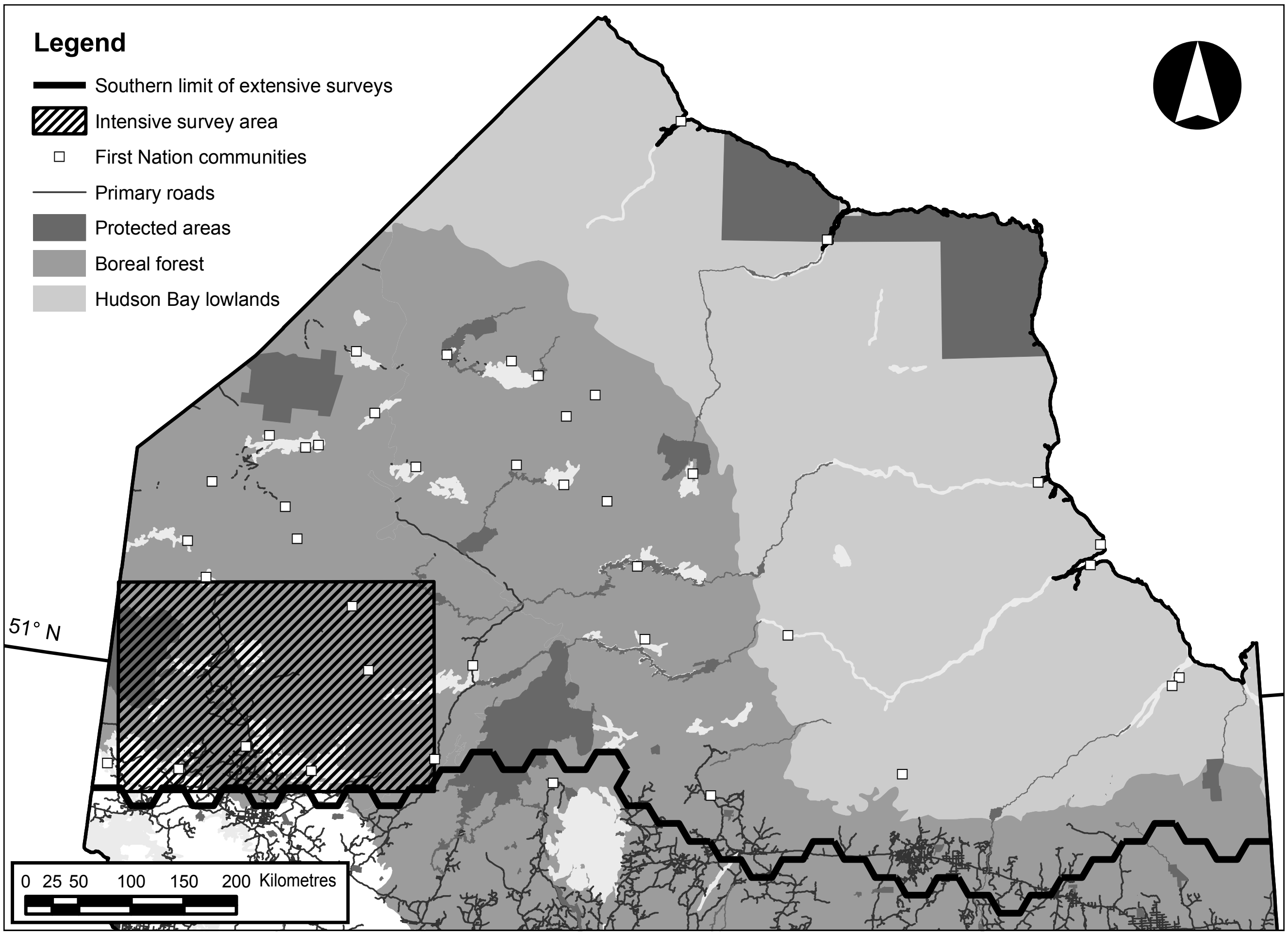
3.1 Spatial scale

3.1.1 How large should the study area be?

The spatial scale over which a study is conducted is an important consideration that is fundamentally based on both the objectives of the study and the biology of the target species. If the overall distribution of a species is of interest, a study that spans large spatial scales is often warranted. When resources (such as time and money) are limited, there is a trade-off between spatial scale and resolution; surveys over large areas necessarily will have fewer sample units. Alternatively, if the distribution of a species spans a relatively small area, or distribution at the edge of its supposed distribution is of interest, a more intensive, higher resolution study over a smaller area may be appropriate.

Population estimates obtained from capture-recapture models are more accurate with large populations (Otis *et al*. 1978). Thus, the study area must be large enough to contain many individuals, and the size will ultimately depend on the biology of the species of interest, particularly their home range size. Animals with relatively large home ranges (hundreds to thousands of km^2) will require study areas of several thousand square kilometres, whereas for species with small home ranges on the order of 10–100 km^2, study areas that are considerably smaller in size will be sufficient. In the case of wolverines, a relatively large area with suitable habitat is required to support a viable, self-sustaining population. For example, Sæther *et al*. (2005) estimated that 46 sexually mature (at least three years old) female wolverines in a population are necessary to avoid the risk of extinction. Thus, Magoun *et al*. (2005) estimated that, given that female wolverine home ranges vary between 100–400 km^2, at least 20,000 km^2 of suitable habitat would be required to support a viable, self-sustaining wolverine population. This illustrates the scale necessary for monitoring wolverine populations.

For the purposes of this document, we will define large-scale studies as those that occur across the entire wolverine range in Ontario (>100,000 km^2; Figure 4) and small-scale studies on the order necessary to monitor a viable, self-sustaining wolverine population (<100,000 km^2; Figure 4).



Map prepared by Wildlife Conservation Society Canada. Data Source: Ontario Boreal Wolverine Project, a collaboration between the Ontario Ministry of Natural Resources, Wildlife Conservation Society Canada and The Wolverine Foundation, Inc.

Figure 4. Large (extensive; north of the solid black line) and small (intensive) scale study areas in northern Ontario used by the Ontario Boreal Wolverine Project, 2003–2005.

3.1.2 How large should the sample units be?

The size of the sample unit can influence estimates of occupancy (MacKenzie 2005): If the sample unit is smaller than the home range size of a species, then in any one sample unit, the probability of that species being physically present during the time of the survey is random (given uniform habitat distribution), whereas if the sample units are similar in size to the home range, the species is likely always present in the sample unit (MacKenzie 2005). The former scenario violates the assumption that a site is closed to changes in occupancy (Box 2), which could lead to biased estimates when the unit is surveyed multiple times.

In a capture-recapture context, sample units are usually evenly distributed in grids across the study area (Otis *et al*. 1978). Grid cell size is usually no smaller than the smallest individual home range size to ensure that each individual has an equal probability of being captured (Kendall and McKelvey 2008). Small grid cells may result in a higher probability of recapture relative to larger grid cells, but the probability of violating the assumption of population closure will also be higher (Box 2; Boulanger and McLellan 2001, Boulanger *et al*. 2002, 2004).

3.1.3 How many sample units should be surveyed?

The size of the sample unit will dictate the number of units in a study area. In addition, as mentioned in section 3.2.2, there is a trade-off between the number of sample units and number of repeat surveys, which will influence design decisions when resources are limited (Bailey *et al*. 2007). To maximize precision of occupancy estimates, the number of sample units should increase (with a concomitant decrease in the number of repeat surveys) as the probability of detection increases or the probability of occupancy decreases (Field *et al*. 2005, MacKenzie and Royle 2005).

As mentioned in section 2.1, not all sample units in the study area are necessarily surveyed. See Box 4 for a review of sampling schemes.

Box 4. A review of sampling schemes.

It is common for investigators to divide their study area into sample units, and survey only a portion of these units. There are several ways to choose which sample units to survey. The following review is based on Hayek and Buzas (1997). The most appropriate sampling schemes for wolverine surveys are discussed for the three recommended techniques (aerial track surveys in section 9.0; ground-based track surveys in section 10.0, and hair snare surveys in section 11.0) in Part II of this document.

Simple random sampling. This is the ideal sampling scheme for obtaining unbiased means and variances, but is often not practical for field studies as some of the sample units might not be accessible (for example, owing to a lack of roads).

Systematic sampling. Here, sample units are selected based on ordered intervals. This can be advantageous because sample units can be evenly distributed about the entire study area. However, the confidence intervals of the mean are wider for systematic samples relative to randomly selected samples, and variance cannot be calculated unless replicate systematic sampling is conducted. If the target population is homogeneously or randomly distributed across the area of interest, or the sample size is small relative to the total possible samples, then simple random sample formulas can be used on systematically collected data to produce relatively unbiased estimates.

Stratified sampling. For this technique, the study area is stratified according to some variable (for example, elevation or habitat composition) thought to cause variation in an attribute of the target population (for example, density). The study area is divided based on these variables, and sampling (random or systematic) is performed independently within each stratum. Thus, between-stratum variation is eliminated, resulting in more precise estimates. Systematic samples are sometimes referred to as being spatially stratified. This design allows for assessing variation in some attribute over space.

3.2 Temporal scale

3.2.1 How long should sampling sessions be?

When resources are limited, there is a trade-off between the number of sampling stations and the length of time the stations are deployed. If the species occurs at a low density, short sampling sessions may fail to detect a species that is present, assuming that the probability of detection increases with survey length (Gompper *et al*. 2006). Latency to first detection (LTD), defined as the time until initial detection of the target species at a sampling station, can be used to determine the length of sampling session that maximizes the probability of detection (Zielinski and Stauffer 1996, Foresman and Pearson 1998, Gompper *et al*. 2006). This value will vary with the species in question and the survey method used to detect it (Gompper *et* *al*. 2006). Thus, no clear recommendations can be given, except that, ideally, investigators should run a pilot study to determine the LTD for their study, and ensure that sampling sessions are at least as long as the observed LTD.

3.2.2 How many times should the survey of a particular sample unit be repeated?

Repeatedly surveying a sample unit will increase the likelihood of detecting a species that is actually present (MacKenzie 2005). When resources are limited, there is a trade-off between the number of repeat surveys and the number of sample units (Bailey *et al*. 2007). Field *et al*. (2005) and MacKenzie and Royle (2005) found that two to three surveys per sample unit maximized precision of the occupancy estimate when the probability of detection was >0.5, and for rare species (low occupancy), it was best to conduct fewer repeat surveys at more sample units. If the probability of detection is low, however, more repeat surveys are required (Field *et al*. 2005, MacKenzie and Royle 2005). Bailey *et al*. (2007) described computer software (program GENPRES; Table 1) that estimates the optimal allocation of sampling effort (number of repeat surveys and number of sample units) to maximize precision and minimize bias for occupancy surveys.

MacKenzie and Royle (2005) stressed that the timing of repeat surveys should be such that detection heterogeneity (different detection probabilities over space or time) is minimized; otherwise, occupancy will be underestimated (MacKenzie and Royle 2005). An additional way to deal with heterogeneity in detection probability is to factor in covariates thought to influence this parameter in the model (MacKenzie *et al*. 2002).

When repeat surveys are used to minimize false absences, it is assumed that the sites are closed over the sampling period; that no unoccupied sites become occupied, and no occupied sites become abandoned (MacKenzie *et al*. 2002). Thus, the length of the sampling session should be such that this assumption is met, and will ultimately depend on the nature of the species of interest (e.g. how far individuals range relative to the size of the sample unit, how fast individuals move between units, generation time of the population, and seasonality of the environment).

In a capture-recapture context (Box 2), precision of the population estimate increases with the number of repeat surveys and the probability of recapture (Otis *et al*. 1978, Kendall and McKelvey 2008). The probability of recapture can be increased by decreasing grid cell size (thus increasing the density of sample stations in the study area), though there is a trade-off between this and the probability of violating the assumption of population closure (Box 2; Boulanger and McLellan 2001, Boulanger *et al*. 2002, 2004).

3.2.3 Over how many seasons should a population be monitored?

A consideration for monitoring trends in population abundance is distinguishing real changes in abundance from noise (i.e. natural fluctuations in abundance due to annual cycles, seasonal variation, and irregular fluctuations [Gibbs *et al*. 1998; Legg and Nagy 2006]). If the temporal scale at which monitoring occurs is not long enough to distinguish between these and any real trends that may be occurring, investigators are at risk of misinterpreting the status of the population at any given point (Bailey *et al*. 2007). Furthermore, the power to detect a trend increases with the number of seasons over which the population is monitored (Harris 1986, Gibbs *et al*. 1998, Mackenzie 2005, Bailey *et al*. 2007; see section 2.4). Thus, *a priori* power analyses should be conducted in order to determine the number of seasons necessary to detect a biologically significant magnitude of population change.

4.0 Large Spatial Scale Review of Methods

The following section provides a review of available methods for surveying and monitoring wolverine populations across a study area of >100,000 km^2 in size, or the scale of current wolverine distribution in Ontario (300,000 km^2), followed by recommendations for methodology as a function of survey objective.

4.1 Snow track surveys

4.1.1 Aerial

Most aerial survey methods have been designed for smaller spatial scales (section 6.1.1). The transect intercept probability sampling (TIPS; Becker 1991) and sample unit probability estimator (SUPE; Becker *et al*. 1998) methods, used to survey wolverine populations in relatively small areas of 2,000–32,000 km^2 (Becker 1991, Golden *et al*. 2007b), require that all tracks are followed forward and backward, and for SUPE, that sample units are searched exhaustively. Furthermore, Golden *et al*. (2007b) recommended that investigators exhaustively search 45–70% of the sample units. Thus, these methods are not logistically or economically feasible for studies spanning regions as large as northern Ontario. In response to this need, Magoun *et al*. (2007a) developed a hierarchical spatial modeling (HSM) technique to estimate the distribution (or probability of occurrence) and relative abundance of wolverines that can be applied to large areas (>100,000 km^2) with some modification. The technique that Magoun *et al*. (2007a) used for northern Ontario was designed to require one or several passes through hexagon-shaped sample units, with investigators simply recording detection or non-detection of tracks.

The HSM analysis (Magoun *et al*. 2007a) was based on methods presented by Sargeant *et al*. (2005), where the probability of track occurrence is estimated for each sample unit based on Markov Chain Monte Carlo (MCMC) simulation. The probability of detecting tracks, based on data obtained from repeat surveys, is incorporated into the model. For large study areas, however, it will no longer be logistically or economically feasible to use multiple discrete visits as the basis for determining detection probability as done by Magoun *et al*. (2007a). In such cases, Magoun *et al*. (in prep.) instead considered portions of 1,000-km^2 hexagons as separate transects. Another option might be to fly additional transects through those hexagons with no tracks detected in the first pass, for up to three passes (Gardner *et al*. in prep.). The design of surveys to estimate probability of occurrence must address heterogeneity in detection probability (MacKenzie *et al*. 2002, MacKenzie 2005). Factors other than track abundance most likely to affect this metric include the distribution of forest openings, thickness of canopy cover, and time since last snowfall or windstorm. When multiple survey teams are used, it is assumed that detection probabilities are not influenced by differences in the ability of teams to detect tracks. This is discussed more thoroughly in section 9.0. The use of HSM aerial surveys depends on the availability of experienced observers and pilots, adequate snow conditions, and aircraft that are highly maneuverable and can fly relatively low and therefore have a low risk of missing tracks. Sample units must be appropriately scaled, and a large proportion of the sample units must be surveyed in areas with both high and low wolverine abundance (Ray *et al*. in prep.). To date, this technique has been used at a large spatial scale by Magoun *et al*. (in prep.) to estimate the occurrence probability of wolverines in a 521,000-km^2 area in northern Ontario, divided into 521 sampling units of 1,000-km^2 hexagons. The design was also used in Labrador (Schmelzer 2005), although no wolverines were detected.

4.1.2 Ground

Lindén *et al*. (1996) described the "wildlife triangle" technique to estimate the relative abundance of many wildlife species in Finland, based on counts of tracks that intercept transects. There are over 1,500 triangle-shaped transects across Finland, with 4-km-long sides, for a total transect length of 12 km per triangle. Wildlife triangle density is one triangle/200 km^2 in southern Finland and one triangle/300 km^2 in northern Finland (Lindén *et al*. 1996). Triangles are searched twice annually; in summer and winter. In winter, over 6,000 volunteers from hunting clubs in Finland search transects twice for animal tracks in the snow. On the first day, all tracks are marked with a line in the snow, and on the following day, all fresh, unmarked tracks are counted. Alternatively, triangles can be searched once, one or two days after a snowfall that covers old tracks (Lindén *et al*. 1996). Volunteers count and record all fresh tracks that cross the transect, regardless of whether the same individual crossed the transect multiple times (Högmander and Penttinen 1996, Lindén *et al*. 1996). If one assumes that track density (track counts per unit area) is proportional to animal density (Pellikka *et al*. 2005), survey data can provide an estimate of relative population abundance.

In Finland, the placement of wildlife triangles is permanent and triangles are surveyed every few years, such that estimates of relative abundance (number of tracks that cross the transect/10-km transect/24 hours) can be compared throughout the country and changes in relative abundance over time can be monitored (Helle *et al*. 1996). Furthermore, these estimates can be translated into estimates of absolute abundance by dividing the average track density by the average distance traveled by an individual in one day (Danilov *et al*. 1996, Högmander and Penttinen 1996).

Bayne *et al*. (2005) have adapted Finnish wildlife triangles (Lindén *et al*. 1996) to estimate and monitor the relative abundance of forest birds and mammals in the province of Alberta (>660,000 km^2) over many years. They divided the province into a grid, with triangle-shaped transects permanently placed at the center of each grid cell, for a total of 1,656 transects separated by 20 km. The protocol used by Bayne *et al*. (2005) differs from the Finnish wildlife triangles (Lindén *et al*. 1996) in that teams of two people completed each triangle, the perimeter of the equilateral triangle was 9 km (rather than 12 km), transects were surveyed only once (between three and 10 days after the last snowfall), and the number of days that tracks had accumulated (days since last snowfall) was estimated and accounted for in the analysis (Bayne *et al*. 2005). Bayne *et al*. (2005) suggested that the proposed survey intensity (each transect surveyed once every five years for 15 years) would provide adequate power to detect changes in population abundance over time for most species. However, species with few occurrences, such as wolverines, would need to be surveyed more intensively (i.e. more transects; Bayne *et al*. 2005). Bayne *et al*. (2006) compared surveys using 9-km-long, triangle-shaped transects surveyed by foot to 10-km-long, linear transects along trails and seismic lines surveyed by snowmobile, and recommended using snowmobiles to survey transects for wolverine tracks in Alberta; they found more individual tracks via snowmobile, presumably because more area could be covered. For their survey via snowmobile, Bayne *et al*. (2006) estimated the presence or absence of tracks crossing the transect for every 250-m segment of the transect, rather than the number of tracks that crossed the transect. The ground-based snow track surveys of Lindén *et al*. (1996) and Bayne *et al*. (2005) were designed to estimate the abundance of numerous species simultaneously. Although both studies detected wolverines, in Alberta Bayne *et al*. (2005, 2006) reported low detection rates.

4.2 Interviews

Under the Species at Risk Act (SARA), the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) is appointed to assess and classify the status of wildlife species in Canada; this assessment is based on both science and local knowledge. Knowledge derived from indigenous or local residents has been described as "the knowledge acquired from extensive observation of an area or species" (Huntington 2000) and "knowledge that helps monitor, interpret, and respond to dynamic changes in ecosystems" (Berkes *et al*. 2000). Methods for data collection vary, but generally, past or current knowledge is collected from local people with semi-directive interviews, questionnaires, workshops, and/or collaborative field work (see Huntington 2000).

Information from local or indigenous residents, such as trappers or other users of the land, can provide valuable information on the natural history and demography of wildlife populations. This can be particularly valuable for wildlife populations in remote areas, where baseline ecological data is otherwise difficult to obtain (Gilchrist *et al*. 2005). Estimates of relative abundance, historical distribution, trapping effort, and spatially explicit locations for unreported harvests and animal sightings can be obtained with local knowledge. However, Gilchrist *et al*. (2005) and Usher (2000) contend that local knowledge should undergo testing and validation, just as scientific data should. Moller *et al*. (2004) advocate that local knowledge and science should complement each other in population modeling: scientific data is often over short time periods and large areas, whereas local knowledge data is over long periods of time but in small, local areas. Together, they can provide a more complete picture of population dynamics across both temporal and spatial scales (Moller *et al*. 2004, Gilchrist *et al*. 2005).

Local knowledge has been used for gaining insight into wolverine populations in Canada. Cardinal (2004) collected indigenous knowledge in the Yukon, Northwest Territories, and Nunavut, elucidating population size and trends, movements, habitat selection, and food habits. Ray (2004) collected indigenous knowledge in northern Ontario and gained information on historical distribution, relative abundance, unreported harvest, and cultural attitudes towards wolverines. Ray (2004) cautioned that the ability to collect robust information on wolverine ecology will rely on the extent to which trappers and hunters target wolverines, and hence pay attention to wolverine habits and ecology. Ray (2004) obtained 153 temporally and spatially explicit wolverine locations in northern Ontario since 1990 from interviews with First Nations trappers and hunters.

4.3 Opportunistic observations

Opportunistic observations provide presence-only location data for the species of interest. The source of the locations is often from incidental or unreported harvest, roadkill, or sightings. Most commonly, such locations and other relevant information are submitted by members of the public following wolverine observation events, rather than being solicited (such as during an interview; see section 4.2). Those assembling this information usually impose a filter on them, by verifying the information to the extent possible, and ensuring thorough documentation of any data associated with the observation.

Locations obtained opportunistically, provided that they are reliable, can supplement other data when generating distribution maps, albeit with several important caveats. Foremost, locations must be spatially and temporally explicit, with the resolution depending on the objectives of the study. For example, if the study goal is to estimate habitat use, observations should be exact, whereas if the goal is presence-absence in a series of sample units, observations need only be precise at the scale of a given sample unit. Opportunistic observations tend to be clustered in areas inhabited by people, which limits their use for geographically unbiased distribution maps. Finally, opportunistic observations give presence-only data; one cannot assume absence of a species in areas where there are no observations.

Aubry *et al*. (2007) used verified and documented wolverine occurrence records to map the historic and current distributions of wolverines in the contiguous United States. Opportunistic observations can give insight into the presence of species outside of intensively studied areas. Miller (1972), for example, documented the presence of wolverines in Gatineau Park, Quebec in 1972. Dawson (2000) used a combination of wolverine sightings and harvest records to describe the historic distribution and range contraction of wolverines in Ontario.

4.4 Harvest records

There is a fairly complete record of fur harvests in North America in the 20th century, and patchy records dating as far back as the 1600s (Obbard *et al*. 1987). Thus, harvest records present a potentially rich source of information as an index of trends in furbearer population size over time, assuming that the number of animals harvested is proportional to population abundance. Theoretically, in years when population density is low, few animals are harvested because it becomes increasingly more difficult to harvest animals as their numbers decrease. Smith *et al*. (1984) found that harvest records were not a reliable index of population abundance for certain furbearer species in South Carolina. Conversely, Gompper and Hackett (2005) matched harvest records with catch-per-unit effort data and found that population declines indexed in harvest records were, in fact, real population declines and not an artifact of harvest effort. Similarly, Bowman *et al*. (2007) found that declining mink harvests appeared related to actual population declines across Canada.

Assumed population trends based on fur harvest records can be confounded by trapper effort, which is an uncontrolled variable in harvest records in Ontario and elsewhere. Increases in the harvest could be a function of increasing population density of the target species, or an increase in trapper effort (Weistein 1977, Winterhalder 1980). Trapper effort can be influenced by the population density of other species (Weistein 1977, Winterhalder 1980) and pelt prices (Siemer *et al*. 1994), though this has been debated (Elton and Nicholson 1942, Daigle *et al*. 1998). The number of licensed trappers has decreased in the northeastern United States (Siemer *et al*. 1994, Daigle *et al*. 1998) and Ontario (Novak 1987) in recent years.

Bulmer (1974, 1975) used harvest data to assess wolverine population fluctuations in relation to populations of other furbearers in Canada. Similarly, Slough (2007) summarized current wolverine harvest data across Canada. Analysis of harvest records and incidental observations has, until recently, been the primary method for monitoring wolverine populations in Ontario. There are several limitations to this index, however, in addition to the inability to control for trapper effort. Foremost, there has been a “zero-quota” (no harvest allowed) for wolverines on non-aboriginal trapping licences in Ontario since 2001–2002. Thus, harvest records cannot be a useful index for population trends if few animals are harvested. Second, harvest records do not account for unreported harvest (such as animals taken for personal use), the extent of which is unknown. Ray (2004) and Cardinal (2004) used local knowledge in Ontario, and Yukon, Northwest Territories, and Nunavut, respectively, to quantify unrecorded harvests in northern communities. Ray (2004) found that >90% of the legal harvest in northern Ontario was reported in the fur auction (i.e. on harvest records), illustrating that harvest records appear to accurately depict the true legal harvest in this province. Conversely, Cardinal (2004) found that fur harvest records do not accurately reflect true harvest. This discrepancy is likely because of regional markets for wolverine fur in northwestern Canada. Finally, harvests tend to be centered on human settlements, resulting in harvest records that are geographically biased (Golden *et al*. 2007a).

4.5 Effective population size

Genetic material obtained from non-invasive sampling (e.g. hair snares) and harvested samples can be used to index actual population size. Theoretically, genetic diversity decreases with decreasing effective population size (Ne; Soulé 1976, Frankham 1996). N_e is defined as the size of an ideal population experiencing the same rate of genetic change as the natural population of interest (Schwartz *et al*. 1998). There are two common measures of N_e: variance N_e (N_{eV}) and inbreeding N_e (N_{eI}). Leberg (2005) defined the former as the size of an ideal population experiencing genetic drift at the same rate as the actual population, and the latter as the size of an ideal population losing heterozygosity owing to increased relatedness, at the same rate as the actual population. In this ideal population, all individuals have an equal chance of being the parents of any progeny making up the next generation (Leberg 2005), and it is assumed that selection, mutation, population subdivision, and migration do not affect gene frequencies (Schwartz *et al*. 1998, Leberg 2005). Soulé (1976) showed that for several species ranging from marine invertebrates to mammals, the relationship between heterozygosity and population size was positive, with a correlation coefficient of 0.7. Indeed, Johnson *et al*. (2004) documented a significant decline in greater prairie chicken (*Tympanuchus cupido*) genetic variability concomitant with a decrease in populations size; Miller and Waits (2003) found decreased allelic diversity in small populations of grizzly bears (*Ursus arctos*); Hauser *et al*. (2002) found significant declines in the genetic diversity of New Zealand snapper (*Pagrus auratus*) since exploitation of the population began; and Spencer *et al*. (2000) created an experimental bottleneck in western mosquitofish (*Gambusia affinis*) populations to show that allelic diversity and temporal variance in allele frequency was decreased in small populations.

N_e, estimated from genetic samples (Box 5; Table 2) can be used as an index of actual population size (N) as there is a fairly consistent relationship between N_e and N across species: Frankham (1995) reviewed published papers and estimated the average N_e/N ratio for 192 estimates of 102 species to be 0.1–0.11 (when fluctuation in population size, variation in family size, and unequal sex ratios were controlled for). In some instances however, estimates of N_e were greater than estimates of N (Aspi *et al*. 2006). Hauser *et al*. (2002) found that the relationship between N_e and N was constant over time in New Zealand snapper populations.

One general assumption of this technique is that the population is not subdivided (Schwartz *et al*. 1998, Leberg 2005), meaning that genetically differentiated subpopulations should not be combined into one sample. For wolverines in Ontario, this assumption may be difficult to meet. Wilson *et al*. (2000) used allozyme and mtDNA data to assess the genetic variability of wolverine populations in the Northwest Territories and found that populations of wolverines <350 km apart were genetically different. Kyle and Strobeck (2001), however, found that wolverines in Ontario and Manitoba may cluster into a genetically homogeneous group, which suggests that estimates of N_e could be made over this extent. At this point, it seems apparent that more research is required into the extent to which assumptions of this method would be violated for wolverines.

Table 2. A comparison of the disadvantages of several methods to estimate N_{eV}.

Disadvantage	Linkage disequilibrium	Heterozygote excess	Temporal change
Requires more than one sample			✔
Requires a large sample of individuals	>90[1]	30–60[2]	30–45[1,3]
Requires analysis of a large number of polymorphic loci	>6[1]	10–20[2]	>5–10[1]
Requires a polygamous, random-mating system		✔	
No overlapping generations	✔	✔	✔

[1] Schwartz *et al*. (1998)
[2] Luikart and Cornuet (1999)
[3] in successive generations
[4] Jorde and Ryman (1995)'s modification can be used for samples with overlapping generations

Box 5. Genetic diversity and effective population size.

Effective population size (N_e) can be estimated from both demographic (e.g. variance in reproductive success) and genetic data (Schwartz *et al*. 1998). Demographic parameters tend to overestimate N_e (Schwartz *et al*. 1998) and are difficult to obtain for rare carnivores, thus we will focus on N_e estimates derived from genetic data. We note here that, as stated by Schwartz *et al*. (1999), a comprehensive review of all methods to estimate N_e would be a large undertaking and is beyond the scope of this document. Rather, here we introduce readers to several common techniques used for the conservation of natural populations.

Genetic-based estimates of N_e require genetic markers, primarily microsatellites (e.g. Miller and Waits 2003). Leberg (2005) noted that the use of microsatellites generally resulted in more power than allozymes for detecting differences in N_e. Similarly, Spencer *et al*. (2000) found that microsatellites provided more power to distinguish different sizes of bottlenecks than allozymes, and Funk *et al*. (1999) found that estimates of N_e obtained from allozyme data had large confidence intervals.

Leberg (2005) grouped N_e estimators into three categories: moment estimators, which compare observed estimates of N_e to what is expected based on theory, maximum likelihood estimators (Wang 2001, Laval *et al*. 2003), and Bayesian estimators (Beaumont 1999, Laval *et al*. 2003). Leberg (2005) pointed out that comparisons of bias and precision between the three groups of estimators tend to be contradictory, and suggests using more than one method. Other methods, such as the coalescent approach to estimate N_eI, are discussed by Leberg (2005).

Schwartz *et al*. (1998) reviewed three moment estimators for estimating N_{eV} from allelic frequency data: linkage (or gametic) disequilibrium, heterozygote excess, and temporal change in allelic frequency. These approaches assume that the population is randomly sampled and is not subdivided, there is no migration, the population size is stable, the sample does not contain overlapping generations, the genetic markers are not under selection or linked to markers under selection, and there is no mutation (Leberg 2005).

Linkage disequilibrium. This technique is based on the non-random association of alleles at different loci. In large populations, there should be no correlation between alleles at independent loci. In small populations, genetic drift will cause the divergence of observed and expected frequencies of combinations of alleles from different loci (Bartley *et al*. 1992, Schwartz *et al*. 1998, Leberg 2005).

Heterozygote excess. In small populations there will be differences in genotype frequencies between sexes by chance, and offspring will tend to be more heterozygous than expected based on overall frequencies in the population (Schwartz *et al*. 1998, Luikart and Cornuet 1999, Leberg 2005). Luikart and Cornuet (1998) describe the heterozygote excess technique as follows: During a bottleneck, alleles are lost faster than heterozygosity. Therefore, observed heterozygosity at each locus in a bottlenecked population will be higher than expected heterozygosity in samples of similar size with the same number of alleles. Expected heterozygosity can be estimated with the stepwise mutation model (Ohta and Kimura 1973) or the infinite alleles model (Kimura and Crow 1964; also described below). Luikart and Cornuet (1998) noted that the bottleneck-induced heterozygote excess is only detectable for approximately 0.2–4.0 N_e generations.

Both linkage disequilibrium and heterozygote excess methods estimate the effective number of breeding adults, but this can be nearly equivalent to N_{eV} if generations from which the samples are taken do not overlap (Schwartz *et al*. 1998). Both methods are advantageous in that they require only one sample (as opposed to temporal change methods described below). However, the former method requires large samples that may not be attainable from small populations, and the latter method is only useful for species with mating systems that have a random union of gametes (all male gametes have an equal chance of combining with all female gametes), which limits its use for populations with social structure (Schwartz *et al*. 1998, Luikart and Cornuet 1999, Leberg 2005). Because of these disadvantages, methods that employ temporal changes in allele frequencies (Funk *et al*. 1999, Hauser *et al*. 2002, Aspi *et al*. 2006) are more common (Schwartz *et al*. 1998).

Temporal change. This method is based on changes in allele frequencies between two samples taken at two points in time. It is based on the fact that allele frequencies change more rapidly in small populations than in large ones because of genetic drift. Precision of the N_{eV} estimate can be increased by increasing the number of generations between samples as this allows more drift to occur (Waples 1989), and by increasing the number of independent loci sampled, the number of individuals sampled, and the number of alleles examined (Waples 1989, Leberg 2005). Jorde and Ryman (1995)'s modification to the temporal method relaxed the assumption of no overlapping generations. Spencer *et al*. (2000) found that temporal variance in allele frequency was more sensitive to genetic changes resulting from an experimental bottleneck than estimates obtained from heterozygosity excess or direct counts of heterozygosity.

The techniques described above estimate N_{eV}. N_{eI} can be estimated from microsatellite or allozyme data using the stepwise mutation model (Ohta and Kimura 1973) or the infinite alleles model (Kimura and Crow 1964), respectively (Leberg 2005; e.g. Rooney *et al*. 1999, Harley *et al*. 2005). These methods require estimates of mean heterozygosity and mutation rate per locus. Only one sampling period is necessary, but unless mutation rates for the population are known, users must make an assumption about mutation rate (Rooney *et al*. [1999] used 2.05 x10^{-4}, the average estimated mutation rate for mouse, human, and pig).

5.0 Large-scale Wolverine Surveys: Recommendations

In the following section we have assessed methods described in section 4.0 in terms of their feasibility for surveying and monitoring wolverines in Ontario and other lowland, boreal forests at large spatial scales. We have then made recommendations based on various study objectives.

5.1 Distribution

Estimates of species distribution are often derived from the proportion of sample units occupied or the probability of a unit being occupied. For wolverines in Ontario and other lowland, boreal forests, these estimates are logistically difficult to obtain at large spatial scales because wolverines occur at relatively low densities over large, remote areas on the order of 300,000 km^2. Thus, the ideal technique for estimating wolverine distribution in Ontario will necessarily be both cost and time effective, and unbiased across the landscape.

Interviews (section 4.2) and opportunistic observations (section 4.3) used to obtain spatially explicit presence data can be useful for supplementing wolverine range maps at large spatial scales. However, because these locations tend to be clustered around human settlements, they tell us little about distribution outside of these areas (i.e. we cannot assume wolverines are absent in areas where there have been no recorded locations). Similarly, though harvest records (section 4.4) have been used in the past to map wolverine distribution, the wolverine harvest is now closed for non-aboriginal trappers in the southern end of the distribution in Ontario. Additionally, harvest records provide presence-only data at the scale of traplines (but a pelt is not always sold by the harvester, and thus can be associated with a different trapline from which it was actually captured), and can be geographically biased towards areas used by humans. Thus, locations obtained from opportunistic observations, interviews, or harvest records alone do not provide an accurate or complete estimate of wolverine distribution in Ontario, although they can provide useful supplementary information.

Ground-based wildlife transects (section 4.1.2), as described by Bayne *et al*. (2005, 2006), could be used to detect wolverines at large spatial scales in Ontario and other lowland, boreal forests. However, given the logistical constraints of conducting surveys in large, remote areas, and the alternative of aerial surveys that are much more cost and time effective at large spatial scales, we do not recommend ground-based snow track surveys for estimating the distribution of wolverines at this scale.

We recommend HSM aerial snow track surveys (section 4.1.1) for estimating wolverine distribution and probability of occurrence at large spatial scales. It is a flexible technique in that flight paths can be chosen such that flight time over areas where tracks are more visible is maximized, and sample units need not be exhaustively surveyed, minimizing flight time per sample unit. The probability of detecting tracks in sample units that were either not surveyed or no tracks were detected can be estimated and incorporated into the model that estimates probability of occurrence. The HSM method is a relatively efficient, geographically unbiased technique for estimating wolverine distribution in Ontario and other lowland, boreal forests at large spatial scales.

5.2 Relative abundance

Estimates of relative abundance generally use the rate of detection of animal sign, the proportion of sample units occupied, or the number of animals harvested or captured, as a correlate of actual abundance. However, because wolverine range in Ontario is relatively large, few methods to estimate relative abundance at the scale of >100,000 km^2 are available (compared to small scale relative abundance estimators in section 6.0).

Harvest records can be used as an index of relative abundance under the assumption that there is a known and predictable relationship between the number of wolverines harvested and the true abundance of wolverines. However, as mentioned in section 4.4, the utility of this information is affected by the fact that wolverine harvest is now closed for non-aboriginal trappers in Ontario, harvest information can only be collected at the trapline scale, and harvests are geographically biased in that they tend to be centered on human settlements or roads. At a large spatial scale, this index will fail to detect changes in wolverine abundance in areas where wolverines are not trapped. There is some indication that wolverine harvest frequency is not a reflection of relative abundance: in a comparison of harvest rate versus probability of occurrence (obtained from aerial surveys) in northern Ontario, Magoun *et al*. (in prep.) found differing patterns of relative abundance across the range. Additionally, Magoun *et al*. (in prep.) noted that one particular area where wolverines have been subject to relatively high harvest rate reflected the open habitat conditions of the taiga/tundra where wolverines could be easily intercepted by snowmobile and hence subject to relatively high rates of opportunistic harvest.

Ground-based wildlife transects (section 4.1.2), as described by Bayne *et al*. (2005, 2006) for Alberta (>600,000 km^2), could conceivably be used to estimate the relative abundance of wolverines at large spatial scales in Ontario (500,000 km^2) and other lowland, boreal forests. Although Bayne *et al*. (2005) recommended ground-based snow track surveys over remote camera and hair snare surveys for rare species in Alberta, such as wolverines, they noted that several transects per grid cell might be required to obtain sufficient probability of detection for such species. This stipulation, coupled with logistical constraints of conducting surveys in large, remote areas, decreases the feasibility of using ground-based surveys that target wolverines in northern Ontario at large scales. This is especially true given that aerial snow track surveys are available and have been shown to be time and cost efficient, and effective for estimating wolverine relative abundance at large scales (section 4.1.1).

N_e estimated from allelic heterozygosity (section 4.5) could be used as an index of wolverine population abundance at large spatial scales using samples obtained opportunistically from harvested animals, hair snares, and live-trapped animals. However, estimates of N_e obtained from temporal changes in allele frequencies requires samples from two time periods separated by more than two generations (Box 5). Estimates of N_e using the infinite alleles and stepwise mutation models (Rooney *et al*. 1999, Hartley *et al*. 2005) could be compared to estimates from populations in other lowland, boreal forests as an index of actual population abundance. The greatest current drawback of genetic methods is uncertainty related to assumptions of these methods for estimating N_e. Before we can recommend this method, more work needs to be carried out evaluating these assumptions.

Once again, we recommend the HSM aerial snow track survey (section 4.1.1) for estimating the relative abundance of wolverines at large spatial scales in Ontario and other lowland, boreal forests. This technique assumes that track detectability is uniform across the study area at the scale of sample units used in large-scale surveys (1000 km^2), and track abundance is positively correlated with wolverine abundance. Provided that these assumptions are met, this technique can be used to efficiently estimate differences in relative abundance (i.e. probability of occurrence) between different parts of the study area. Although we do not know the true relationship between the probability of track occurrence and wolverine abundance in Ontario, occupancy has been shown to correlate with abundance for a number of species; Gaston *et al*. (2000) reviewed the growing body of literature examining this positive correlation. Generally, as the local abundance of a species increases, the area over which it occurs tends also to increase, and conversely, as a species' abundance decreases, so does its range (Gaston *et al*. 2000).

5.3 Abundance and density

Estimates of population abundance and density generally require the identification of individuals by external tags, pelage, or DNA, and several techniques for estimating abundance from snow tracks at small scales have been used (see section 6.1). At a large spatial scale, capturing and/or detecting enough animals to produce accurate and precise estimates is not feasible, therefore common capture-recapture techniques (see section 6.0) are generally not recommended at this scale. Estimates of population abundance can also be obtained with aerial surveys described by Becker (1991) and Becker *et al*. (1998), but are best suited for estimates at the smaller spatial scale (section 6.1.1). At this time, therefore, we believe that obtaining population density estimates at the scale of wolverine distribution in Ontario is not a realistic survey goal.

5.4 Monitoring populations over time

Estimates of distribution, relative abundance, and abundance all lend themselves well to monitoring trends in such parameters over time. The ideal technique for estimating trends in population size indices at large spatial scales is cost and time efficient and unbiased across the landscape. For monitoring wolverine populations in Ontario and other lowland, boreal forests at large spatial scales, there are few options.

Opportunistic observations (section 4.3) and locations obtained from local knowledge (section 4.2) tend to be geographically biased and sporadic: alone they are not reliable sources of occupancy over many seasons. Likewise, harvest effort is not distributed evenly across the landscape, thus harvest records are not an ideal estimator of relative abundance at a large spatial scale. Nevertheless, locations obtained from these methods can be useful to produce a rough estimate of historical distribution to which current distributions can be qualitatively compared. For example, by focused questioning of local knowledge holders, one can collect qualitative information on trends over time, and information from localized areas could be amalgamated into a larger regional picture.

Ground-based snow track surveys (section 4.1.2) are not logistically feasible at large spatial scales in northern Ontario. Furthermore, as these surveys have not been tested for wolverines in northern Ontario, we do not know how many transects per 1,000-km^2 sample unit would be necessary to obtain sufficient statistical power to detect biologically significant changes in wolverine distribution over time.

Changes in abundance inferred through changes in N_e (section 4.5) over several seasons could be used as an index to monitor changes in wolverine population abundance at large spatial scales (Schwartz *et al*. 2007). However, the success of this technique depends on the ability of investigators to obtain genetic samples from wolverines across the range of wolverines in Ontario and the extent to which the assumptions are met. Thus we cannot recommend this technique to monitor wolverine populations at large spatial scales in Ontario at this time.

To monitor wolverine populations at large spatial scales in Ontario and other lowland, boreal forests, we recommend estimating probability of occurrence with HSM aerial snow track surveys (section 4.1.1) over many seasons. As a monitoring tool, investigators can estimate the change in probability of occurrence over time. Specifically, by mapping core (areas with detection probability ≥ 0.8) and peripheral range, investigators can monitor wolverine range expansion or contraction over time.

Box 6. Summary of recommendations for large-scale wolverine surveys.

There are few techniques available to efficiently survey and monitor wolverine populations at large spatial scales. Interviews (section 4.2), opportunistic observations (section 4.3), and harvest records (section 4.4) can be used to supplement distribution maps, as an index of relative abundance, or to generate a qualitative historical picture of trends over time, but tend to be geographically biased and limited in that investigators cannot infer species absence where there are no observations. Thus, we do not recommend these techniques for surveying and monitoring wolverines in Ontario. We also do not recommend ground-based snow track surveys (section 4.1.2) to index and monitor wolverine populations at large spatial scales for logistical reasons: the vastness and remoteness of northern Ontario precludes the use of any ground-based method at this scale, especially relative to the potential efficiency of aerial-based methods. Further, we consider that more research is required to validate assumptions related to using genetic methods to estimate N_e for wolverines in Ontario (section 4.5). Therefore, we recommend hierarchical spatial modeling based on aerial snow track surveys (section 4.1.1) as an efficient technique to estimate wolverine distribution and relative abundance in northern Ontario and other lowland, boreal forests, at large spatial scales (>100,000 km^2). In addition to providing distribution and relative abundance estimates, this technique can be used to monitor wolverine distribution and relative abundance over time (section 5.4).

6.0 Small Spatial Scale: Review of Methods

The following section provides a review of available methods for surveying and monitoring wolverines across a study area of <100,000 km^2 in size, followed by recommendations for survey techniques as a function of survey objectives.

6.1 Snow track surveys

6.1.1 Aerial

Herein, we describe three survey designs for estimating population distribution or abundance from aerial snow track surveys at small spatial scales: transect intercept probability sampling (TIPS; Becker 1991), sample unit probability estimator (SUPE; Becker *et al*. 1998), and hierarchical spatial modeling (HSM; Magoun *et al*. 2007a). These designs share several assumptions (Table 3).

Table 3. Assumptions of three designs for estimating population abundance (transect intercept probability sampling [TIPS; Becker 1991]; sample unit probability estimator [SUPE; Becker *et al*. 1998]) and distribution (hierarchical spatial modeling [HSM; Magoun *et al*. 2007a]) from aerial snow track surveys at small spatial scales.

Assumption	TIPS	SUPE	HSM
All animals of interest move and leave tracks during the course of the survey	✔	✔	✔
All tracks of the target species are observed and recognized (tracks are not missed)	✔	✔	
Tracks are continuous	✔	✔	
Animal movements are independent of the sampling process	✔	✔	✔
Pre- and post-snowstorm tracks can be distinguished	✔	✔	
Tracks can be followed forward to the animal and backward to the location at the end of the snowstorm	✔	✔	
Number of animals in the group can be distinguished	✔	✔	
When survey lasts >1 day, individuals are not missed or counted twice	✔	✔	
Probability of detecting tracks is proportional to abundance			✔

TIPS. Becker (1991) proposed TIPS to estimate the abundance of terrestrial furbearers. This method depends on encountering tracks along a defined transect. A repeated, systematic sample design is used whereby the study area is divided into strata, and a transect is randomly chosen from within each stratum. This process is repeated, so that multiple transects within each stratum provide an opportunity to estimate variance (Becker 1991). When a target species' fresh track in the snow is identified, it is followed forward until the animal is sighted and backward to its origin. The distance between these two locations parallel to the x-axis, and the length of the x-axis, are used to estimate the probability of encountering a track, which is then used to estimate abundance (see Becker 1991 for a detailed description of data analysis). Variations on this design allow the user to stagger or stack x-axes in large study areas, survey over multiple days, or use TIPS in conjunction with data from a sample of radio-collared animals in order to relax the assumption that all tracks that intercept a transect can be followed (Becker *et al*. 2004).

TIPS has been used to estimate wolverine abundance in a 1,871-km^2 study area in Alaska (Becker 1991) and a 2,700-km^2 study area also in Alaska (Becker and Gardner 1992). Becker *et al*. (2004) noted that the coefficient of variation (CV) when using TIPS is high (13–74% in published studies), and that the CV tends to decrease as transect density increases. Thus, Becker *et al*. (2004) suggest a transect density of 255 km/1,000 km^2 to obtain a CV of 10%. Becker *et al*. (2004) also suggest that transects be 20–35 km long, and that wolverine surveys begin within 12 to 24 hours after a snowfall.

SUPE. The TIPS method is limited in areas with dense canopy cover or when the study area is so large that it will take several days to complete the survey (Table 4). Becker *et al*. (1998) addressed shortcomings of the TIPS method with the development of the SUPE method to estimate population abundance. Rather than sampling transects, investigators sample stratified, randomly selected quadrats, or sample units. Although this technique shares the same basic assumptions as TIPS (Table 3), SUPE is designed for surveys in larger areas that take more than one day to complete, is more flexible in route selection when conditions are not optimal (i.e. in dense canopy cover or mountainous terrain), and can employ prior knowledge (from harvest patterns, distribution of prey, etc.) of areas likely to contain tracks of the target species.

The study area is divided into sample units that usually range from 10–41 km^2 (Becker *et al*. 2004). The sample units are grouped into strata based on how likely they are to contain tracks of the species of interest, and the sample units to be searched are chosen randomly from within each strata. Searching proportionally more sample units in the high-likelihood strata should improve the precision of the abundance estimate (Becker *et al*. 1998). Sample units are searched exhaustively, and when a track is encountered, it is followed forward until the animal is observed and backward to its location at the end of the last snowfall. Investigators record the number of track groups and all sample units that the tracks go through. Search intensity will depend on conditions such as the amount of tree cover, but should be sufficient to ensure that the assumptions have been met (Table 3; Becker *et al*. 1998). Becker *et al*. (1998) described the procedure for estimating population size and variance from these data. For each stratum, abundance estimates are based on the number of track groups encountered, the number of sample units the track group passes through, and the number of sample units in each stratum that were surveyed. Software is available for data analyses (program SUPEPOP; Table 1).

Patterson *et al*. (2004) used the SUPE method to estimate wolf density in Algonquin Park, Ontario (3,425 km^2). Several studies have also used the SUPE method to estimate wolverine abundance at small spatial scales. Becker *et al*. (1998) estimated wolverine abundance in a 31,373-km^2 area in Alaska over 10 days, and Golden *et al*. (2007b) estimated wolverine abundance in a 4,340-km^2 forested, mountainous area in Alaska, and a 3,375-km^2 relatively flat area in Yukon. Golden *et al*. (2007b) recommended sampling 65–70% of the sample units in the high-likelihood stratum, and 45–50% of the sample units in the low-likelihood stratum to obtain reasonable CVs. Becker *et al*. (2004) suggested that when using the SUPE method for estimating wolverine abundance, surveys should commence 12–24 hours after a snowfall, and should be completed within two to three days.

HSM. Magoun *et al*. (2007a) designed an aerial survey for efficiently estimating and mapping the probability of occurrence of wolverine tracks in larger (>10,000 km^2), forested areas. Their technique does not require that tracks be followed forward and backward; rather, only the presence or absence of tracks is recorded. Tracks from the same individual detected in multiple sample units will not bias the data, so there need not be an upper limit on the number of days past fresh snowfall to prevent the detection of tracks from one individual multiple times. Sample units are surveyed by flying through the center of each sample unit at bearings chosen to increase the likelihood of detecting tracks (i.e. flying over open areas rather than dense vegetation). Sample units are surveyed repeatedly to derive estimates of detection probability, though all sample units need not be surveyed. Data analysis is similar to that of Sargeant *et al*. (2005). MCMC methods are used to model probability of occurrence and core areas of occupation (Magoun *et al*. 2007a). The analysis is described in detail in Magoun *et al*. (2007a) and section 9.0, and software is available (see Appendix 3).

To date, this technique has been used once, by Magoun *et al*. (2007a), to estimate the distribution and probability of occurrence of wolverines in a 60,000 km^2 study area in northwestern Ontario. Magoun *et al*. (2007a) searched 98% of the 100-km^2 sample units in their study area, and recommended that ≥70% of sample units should have strong evidence of presence or absence for accurate estimates of occurrence. They estimated that this technique required six times less flying time than would TIPS or SUPE for their study area (also, see Table 4). Although HSM does not generate estimates of abundance, it provides a baseline spatial distribution to which future surveys can be compared in a monitoring program (Magoun *et al*. 2007a). The use of this technique depends on the availability of experienced pilots and observers, aircraft that are highly maneuverable and can fly relatively low, and adequate snow conditions.

6.1.2 Ground

Like most aerial survey techniques, ground-based surveys were originally designed to be deployed over small (<100,000 km^2) study areas. For example, Sargeant *et al*. (2005) used a ground-based track survey in western Kansas; an area of approximately 70,000 km^2. They searched for swift fox tracks in naturally occurring substrates, such as dirt roads and used MCMC simulation to estimate the probability of track occurrence for sample units that were unsearched or no tracks were detected. Although Sargeant *et al*. (2005)'s method could theoretically be applied to wolverines, the remoteness of most typical wolverine range precludes its use for surveying wolverines. However, as discussed above, this method has been adapted for aerial surveys for wolverines over various spatial scales.

Counts of animal tracks in snow can be used as an index of population abundance; theoretically, areas with high population densities will have more tracks than areas with low population densities. Thompson *et al*. (1989) counted the number of mammal tracks encountered for every kilometre of transect searched, and Beauvais and Buskirk (1999) corrected this estimate for the number of days since last snowfall. Wildlife triangles (Lindén *et al*. 1996), discussed in detail in section 4.1.2, are similar to the methods of Thompson *et al*. (1989) and Beauvais and Buskirk (1999), except that transects are in the shape of triangles rather than parallel transects. Trackers count all fresh tracks that cross the transect/10 km/24 hours, and this is used as an index of track density (Högmander and Penttinen 1996). If one assumes that track density is proportional to animal density (Pellikka *et al*. 2005), then survey data can provide an estimate of relative population abundance, which can be compared across regions or over time (Helle *et al*. 1996).

a)

Pros	TIPS	SUPE	HSM
Allows surveys to last more than one day	✔	✔	✔
Incomplete surveys are acceptable		✔	✔
Gives probability/estimate of occurrence for unsampled units	✔	✔	✔
Estimates abundance	✔	✔	
Software available for analysis		✔	✔
Allows flexibility in route selection when conditions are not optimal		✔	✔
Precision can be increased by stratifying units based on prior knowledge of occupancy		✔	
Robust to autocorrelated data			✔
Can be used to efficiently survey areas >10,000 km^2 in size			✔

b)

Cons	TIPS	SUPE	HSM
Requires experienced pilots and trackers	✔	✔	✔
Requires aircraft that are highly maneuverable and can fly relatively low	✔	✔	✔
Difficult to meet the assumption that all tracks are seen	✔		
Need to follow tracks forwards and backwards	✔	✔	
Sample units need to be searched exhaustively		✔	
Relatively time consuming	✔	✔	
Highly dependent on snow cover and track condition	✔	✔	

Table 4. Pros (a) and cons (b) of three designs for estimating population abundance (transect intercept probability sampling [TIPS; Becker 1991]; sample unit probability estimator [SUPE; Becker *et al*. 1998]) and distribution (hierarchical spatial modeling [HSM; Magoun *et al*. 2007a]) from aerial snow track surveys.

Although it can be adapted for large study areas (see discussion in section 4.1.2), the Finnish wildlife triangle method (Lindén *et al*. 1996, Bayne *et al*. 2005) becomes more feasible at smaller spatial scales because effort can be allocated towards a higher transect density, which will increase the probability of track detection. Several studies have used ground-based methods to survey wolverines: Halfpenny *et al*. (1995) recommended surveying 10-km^2 sample units by snowshoe or ski to detect wolverine tracks. Bayne *et al*. (2006) compared surveys conducted on foot to those conducted via snowmobile, and encountered more wolverine tracks per transect with the latter. Ulizio *et al*. (2006) collected hair and scat samples for DNA analysis while following wolverine tracks in the snow and found that this was an efficient method to verify track identification and identify individuals for capture-recapture studies if sample sizes are large. For example, Ulizio *et al*. (2006) estimated that they detected 80% of the individual wolverines in the population using snow tracking.

6.2 Remote cameras

Remote cameras have increased in popularity along with blossoming technology for surveys of carnivores such as snow leopards (*Uncia uncia*; Jackson *et al*. 2006), leopards (*Panthera pardus*; Henschel and Ray 2003), tigers (*Panthera tigris*; Karanth and Nichols 1998), grizzly bears (*Ursus arctos*; Mace *et al*. 1994), bobcats (*Lynx rufus*; Heilbrun *et al*. 2006, Long *et al*. 2007), coyotes (*Canis latrans*; Larrucea *et al*. 2007, Long *et al*. 2007), martens (*Martes americana*; Foresman and Pearson 1998), fishers (*Martes pennanti*; Foresman and Pearson 1998, Long *et al*. 2007), Canada lynx (*Lynx canadensis*; Foresman and Pearson 1998), and wolverines (Foresman and Pearson 1998, Fisher 2004, 2005, Magoun *et al*. 2007b). Generally, cameras are set up in a grid or along transects throughout the study area, and movement of an animal in front of the camera triggers the camera to take a time-stamped photograph (Box 7). Camera stations are evenly distributed across the study area so that each individual has at least some opportunity to be "captured". Within grid cells, cameras can be set up in locations that will maximize the probability of detection, such as along high-travel routes (Jackson *et al*. 2006, Larrucea *et al*. 2007), in areas where sign, such as tracks or scat, of the target species has been previously observed (Karanth and Nichols 1998), or where the target species can be lured to the station with bait or scent lures (Mace *et al*. 1994). After the initial equipment investment, remote cameras show promise as a cost-effective, non-invasive method to collect abundant data with minimal effort (Table 5). As equipment per sample unit can be expensive and cameras need to be set up and monitored from the ground, this method is best suited for studies at small spatial scales.

Box 7. A review of remote camera equipment.

There are two main components to a remote camera set-up: the camera and the trigger. Kays and Slauson (2008) have summarized the pros and cons of several camera and trigger devices, which we review here.

Still cameras can use 35mm film or digital technology. There is a clear advantage to using digital cameras, in that the number of exposures is much greater than film and it is less likely that the camera will run out of exposures before the survey period is over. Disadvantages of digital cameras include greater battery consumption and a delay between trigger and photo capture compared to film cameras (Jackson *et al*. 2005). However, advances in digital technology are quickly offsetting these disadvantages.

There are several options available for triggers to trip the camera when the target animal is in view of the camera. Pressure pads can be used so that the photograph is taken when the animal steps on the pad (e.g. Moruzzi *et al*. 2002). Alternatively, advances in infrared technology have led to two commonly used trigger devises: active (AIR) and passive (PIR) infrared detectors. Active infrared detectors emit a beam of infrared light, and the camera is triggered when an object breaks the beam by moving in between the source and sensor of the beam. These types of detectors are advantageous in that they are more sensitive and can be set to a wider range of target areas compared to PIR detectors. However, they are also prone to photographing non-target species, and can be set off by wind or rain. Passive infrared detectors use the differential temperature between moving objects in front of the sensor and the surrounding environment to trigger the camera. They are advantageous in that they only require one sensor component housed with the camera, which facilitates set-up, and they are generally less expensive and weigh less than AIR devices (Henschel and Ray 2003). However, they can be triggered when ambient temperatures approach mammalian body temperature or by direct sunlight.

Remote cameras can be an effective method for estimating species distribution because species identification with photographs is rarely ambiguous. Spatial and temporal trade-offs discussed in section 3.0 apply to remote camera surveys. The effectiveness of camera surveys compared to other survey methods to detect carnivores varies depending on which species is of interest (Gompper *et al*. 2006). For example, O'Connell *et al*. (2006) found that cameras had a higher detection probability than track plates and hair snares for most mammalian species in their study, while Bull *et al*. (1992) found that track plates were more likely than cameras to detect martens (Martes americana).

Carbone *et al*. (2001) showed that photographic rate (number of camera days/photograph) could be used to accurately index tiger population density. This finding is particularly useful when individuals of the target species cannot be distinguished. However, Carbone *et al*. (2001)'s methods and conclusions have been met with skepticism (Jenelle *et al*. 2002). Jenelle *et al*. (2002) stressed that if individuals cannot be identified in photographs, camera surveys should be restricted to estimates of presence-absence, and not used for estimates of relative abundance.

One of the benefits of using cameras to survey carnivore populations is the potential to use a capture-recapture framework to estimate population abundance and density if individuals can be distinguished. Some studies have used differences in pelage between individuals (Karanth and Nichols 1998, Jackson *et al*. 2006, Larrucea *et al*. 2007), or external markers, such as ear-tags, applied by the investigators (Sweitzer *et al*. 2000, Martorello *et al*. 2001) to identify individual animals. Camera surveys must be repeated within a season and capture histories for each individual are recorded. Capture histories are used for abundance estimates with capture-recapture models (Box 2), and these abundance estimates can further be used to estimate population density (Box 3). Increasing the number of capture sessions will increase the probability of capture and result in more precise population estimates (Otis *et al*. 1978).

Remote camera surveys have been tested as a method to detect wolverines (Fisher 2004, 2005, Lofroth and Krebs 2007, Magoun *et al*. 2007b). Fisher (2004, 2005) used remote cameras in Alberta and photographed one and zero wolverines in 1,026 (Fisher 2004) and 697 trap nights (Fisher 2005), respectively. Lofroth and Krebs (2007) set up remote cameras in randomly selected 100-km^2 grids in British Columbia and photographed 14 individual wolverines 30 times in one study area, and 22 individuals 47 times in another study area. Magoun *et al*. (2007b) used remote cameras to detect wolverines in Alaska and were able to identify individuals by unique pelage patterns on the neck and chest. Magoun *et al*. (2007b) recommended remote cameras over hair snares for capture-recapture studies of wolverines in Alaska. In a 2,000-km^2 portion of northern Ontario, members of the Ontario Boreal Wolverine Project (unpublished) used remote cameras to photograph and identify individual wolverines by pelage patterns. One or two camera stations were set up within each of 20 100-km^2 hexagons, in locations likely to be visited by wolverines. Camera station set-up was integral to taking photographs that allowed investigators to unequivocally identify individual wolverines.

6.3 Hair snares

Hair snares are a relatively non-invasive method to survey carnivore populations (Table 5). Hair snare devices are designed to take advantage of instinctive behaviours of the target species to entice them to make physical contact with the hair snare and leave a hair sample behind (Kendall and McKelvey 2008). For example, lynx (McDaniel *et al*. 2000), bobcats

(Harrison 2006), and gray foxes (*Urocyon cinereoargenteus*; Downey *et al*. 2007) can be sampled with scented rub pads, martens (Foran *et al*. 1997) and foxes (*Vulpes macrotis mutica* and *Vulpes velox*; Bremner-Harrison *et al*. 2006) with baited cubbies, bears (*Ursus arctos* and *Ursus americana*; Woods *et al*. 1999) and badgers (*Meles meles*; Frantz *et al*. 2004) with baited corrals, and wolverines (Mulders *et al*. 2007) with baited posts wrapped in barbed wire.

Hair snare surveys can be used to detect presence-absence of a species in a sample unit, similar to remote camera surveys (section 6.2). Hair snare surveys to detect presence-absence have similar design constraints as well; limited resources must be allocated to maximize the probability of detecting a species (sections 2.1 and 3.0). However, hair snare surveys have an additional limitation: species identification can be ambiguous. It is possible to use hair morphology to identify species (Belant 2003, Lynch *et al*. 2006), but often DNA analysis is required (Box 8).

To use hair snare surveys in a capture-recapture framework to estimate population abundance requires that individuals are distinguishable. This can be accomplished with microsatellite analyses (Box 8). The general assumptions of capture-recapture apply here as well (Box 2). Repeat surveys are conducted in a short time period to produce capture histories for each individual. Software, such as programs MARK or CAPTURE (Table 1), is available to estimate abundance from the capture histories. Population density estimates can be calculated from abundance estimates if the size of the study area can be defined (Box 3).

Since DNA analysis is relatively expensive, genotyping every strand of hair from every barb may not be economically feasible. Generally, all hair strands on one barb are considered one sample (Kendall and McKelvey 2008). To minimize the number of samples that are genotyped, investigators can subsample from barbs at each hair snare station since hair on adjacent barbs is likely to be from the same individual (Kendall and McKelvey 2008). Boulanger *et al*. (2006) found that increasing the number of barbs per station increased the number of hair samples but did not change the population abundance estimate or precision of the estimate. If >1 animal has left hair on one barb, pooling the hair from that barb to increase the size of the DNA sample can lead to genotyping errors that combine the two genotypes and create a spurious individual (see Box 8; Alpers *et al*. 2003, Schwartz and Monfort 2008). Consequently, several investigators have designed hair snare traps that prevent multiple individuals from leaving a sample (Belant 2003, Bremner-Harrison *et al*. 2006). Alternatively, investigators can reduce the probability of mixed samples by removing hair more frequently or providing single-serving bait (Kendall and McKelvey 2008).

The success of DNA amplification from hair follicles is related to the quality of the sample (Roon *et al*. 2003). Ultraviolet light and moisture can degrade DNA. Roon *et al*. (2003) found that amplification success was higher when mtDNA was extracted two weeks after collection compared to two months, and when nDNA was extracted <6 months after collection. Furthermore, storing hair at -20°C resulted in higher amplification success than using a silica desiccant at room temperature (Roon *et al*. 2003).

Hair snares have been used to detect and identify individual wolverines (Mowat *et al*. 2003, Fisher 2004, 2005). Mowat *et al*. (2003) tested three hair snare designs for wolverines in Alberta: baited barbed-wire corral traps, baited boxes with barbed wire across the entrance, and scented rub pads. They found that there was low visitation to all traps by wolverines, but had greatest success with baited corral traps (Mowat *et al*. 2003). Fisher (2004, 2005) found that corral traps were not effective for snagging hair from wolverines in Alberta, and had much greater success with barbed wire

Box 8. A review of genetic sampling for identification of species and individuals.

There are two main types of DNA found in animal cells: mitochondrial (mtDNA) and nuclear (nDNA) DNA. MtDNA is maternally inherited, each cell has hundreds to thousands of copies, and it is often less variable within species than nDNA, but it is variable between species (Schwartz and Monfort 2008). Thus, mtDNA is often used for species identification. nDNA, on the other hand, is inherited from both parents, is usually present in two copies in the nucleus of each cell, and is variable both within and between species (Schwartz and Monfort 2008). Thus, nDNA is used for individual identification and gender determination. Microsatellite analysis is a tool commonly used to identify individuals from nDNA. Microsatellites are regions of the DNA sequence with repeating units of two to five base pairs. The number of times the nucleotide sequence is repeated usually varies between individuals. DNA can be obtained from many sources (see Waits 2004) but for surveys of rare carnivores, DNA is commonly obtained from hair follicles or sloughed intestinal epithelial cells found in faeces (Waits 2004). The DNA sample is amplified to yield millions of copies of the DNA region of interest using polymerase chain reaction.

Population abundance can be underestimated if investigators fail to distinguish between individuals. Several microsatellite loci are often required for individual identification; the number depends on the number of individuals likely to be sampled and the genetic variation of the species or population (Waits and Paetkau 2005). For example, Kyle and Strobeck (2001) used 12 microsatellite loci to identify North American wolverines. If too few microsatellite loci are used, if loci have low variability, or the population consists of highly related individuals, investigators will tend to assign the same identity to distinct individuals (shadow effect). In a capture-recapture framework, this will cause investigators to overestimate the number of recaptures and thus underestimate population abundance (Mills *et al*. 2000, Waits and Leberg 2000).

Genetic samples obtained from hair and faeces are often of low quantity and quality, and thus are prone to genotyping errors (Goossens *et al*. 1998). Failing to detect two alleles when they are present (false homozygotes, or allelic dropout) and less commonly, detecting alleles that are not actually present (false alleles) can ultimately lead to biased estimates of population size (Waits 2004). False allele errors can usually be detected and thus false homozygotes are more problematic (Waits 2004). The end result of false homozygote genotyping errors is that different genotypes are assigned to different samples from the same individual, inflating the number of individuals. In a capture-recapture study, this would underestimate the number of recaptures and cause investigators to overestimate population abundance (Waits and Leberg 2000, Creel *et al*. 2003). Several researchers have described error-checking protocols to detect genotyping errors (Taberlet *et al*. 1996, Paetkau 2003, McKelvey and Schwartz 2004a).

There is a trade-off between two of the types of errors described (shadow effect and allelic dropout). Increasing the number of microsatellite loci that are analyzed increases the probability of identity (or decreases the shadow effect), but also increases genotyping errors (Waits and Leberg 2000, Creel *et al*. 2003). Thus, Waits and Leberg (2000) suggested genotyping only those loci that significantly contribute to increasing the probability of identity.

wrapped around a tree or post. Of 54 and 312 hair samples collected, 32 (Fisher 2004) and 239 (Fisher 2005) samples, respectively, were adequate for identifying species with mtDNA analysis. They identified five individual wolverines at seven sites (Fisher 2005). Magoun *et al.* (unpublished data) tested hair snare designs on captive wolverines and found the most effective design was a series of alligator clips mounted on a horizontal bar and attached to the end of a runpole above which a bait was suspended. They used this design successfully to collect hair from wild wolverines in Alaska at three sites where wolverines were visiting baited camera trapping sites; they are conducting tests of the design over a larger area in 2009 (Magoun *et al.* 2008). Magoun *et al*. (2007b) recommended setting hair snare traps in March, because prior to that, wolverine hair was difficult to remove and tended to break so that the hair samples did not include follicles. Mulders *et al*. (2007) used 284 baited posts wrapped in barbed wire to snag hair from wolverines in a 2,556-km^2 study area in the tundra of Northwest Territories. They identified 29 male and 24 female wolverines, and estimated probability of detection at >0.5 for both sexes. Baited hair snares were used in northern Ontario for detecting and identifying individual wolverines (Ontario Boreal Wolverine Project, unpublished). Investigators used barbed wire wrapped around trees between the end of February and middle of April. Sites were pre-baited and three to four snares were set up within 100-km^2 hexagons. Several wolverines were detected, but there was evidence that wolverines could take the bait without leaving hair on the barbs, suggesting that hair snares have to be carefully set.

6.4 Scat surveys

Scat surveys are a non-invasive method for collecting genetic data to estimate population parameters since this technique does not require the animal to be lured to the sampling device (Table 5). Surveys are often completed by walking transects through the study area (Harrison *et al*. 2004, Wasser *et al*. 2004, Harrison 2006, Mackay *et al*. 2008), or less commonly, by a non-systematic attempt to survey the entire study area (e.g. Eggert *et al*. 2003) or by obtaining samples opportunistically (Hedmark *et al*. 2004, Bellemain *et al*. 2005). Some studies depend on humans to encounter scat (e.g. Palomares *et al*. 2002, Eggert *et al*. 2003, Harrison *et al*. 2004, Bellmain *et al*. 2005), but recently, studies have employed scat detection dogs to seek out scat with great success (Wasser *et al*. 2004, Harrison 2006, Long *et al*. 2007, MacKay *et al*. 2008). Scat surveys for some forest carnivore species using scat detection dogs have higher detection probabilities than other non-invasive methods such as hair snare and track surveys (Harrison *et al*. 2002, Harrison 2006, Long *et al*. 2007) and are more efficient at detecting scats than humans alone (MacKay *et al*. 2008).

Some investigators have depended on morphological characteristics of scat to identify the species that deposited it (Lynch *et al*. 2006). However, scat morphology alone might not lead to unequivocal species identification as some attributes, such as scat diameter, might overlap between coexisting species (Farrell *et al*. 2000, Reed *et al*. 2004). Davidson *et al*. (2002) found that expert naturalists in the Untied Kingdom were unable to accurately distinguish pine marten (*Martes martes*) scat from fox (*Vulpes vulpes*) scat. Thus, DNA-based field studies are becoming more popular (Lukacs and Burnham 2005).

Scat surveys have been used to estimate species distribution (Palomares *et al*. 2002). Scat surveys have also been used for estimates of relative abundance; Harrison *et al*. (2004) used the number of transects with scat in New Mexico as an index of swift fox (*Vulpes velox*) abundance state wide. It is assumed that there is a positive correlation between the rate or probability of scat detection and abundance of scat, and, in turn, a positive correlation with abundance of scat and population abundance. As it is often unknown whether this assumption has been violated for scat surveys, and because individuals can potentially be distinguished using microsatellite analyses with scat samples (Box 8), abundance estimates are often preferred over relative abundance estimates (MacKay *et al*. 2008).

Scat surveys intended to estimate population abundance depend on the ability to use DNA from sloughed intestinal epithelial cells found in scat to identify individuals. Amplification success tends to be lower for DNA found in scat relative to hair (Waits 2004). Hedmark *et al*. (2004) and Flagstad *et* *al*. (2004) reported amplification success of 65% and 77%, respectively, for nDNA from wolverine scat. Amplification success for DNA from scat can be affected by time between defecation and amplification (Piggott 2004, Murphy *et al*. 2007), preservation method (Wasser *et al*. 1997, Frantzen *et al*. 1998), and field conditions, such as temperature and moisture (Murphy *et al*. 2007). Recommendations for minimizing the effects of these variables vary between species and studies, but generally, cool and dry conditions between defecation and scat collection will result in higher amplification success (Farrell *et al*. 2000, Murphy *et al*. 2007), therefore studies should be planned for drier, cooler seasons.

There are two common methods for estimating population abundance from genotype data obtained from scat: capture-recapture (Box 2), and rarefaction curves (Box 9). Eggert *et al*. (2003) and Bellemain *et al*. (2005) compared population abundance estimates using both methods to independent abundance estimates and came to similar conclusions. Estimates obtained with rarefaction curves with the Kohn *et al*. (1999) equation tended to be biased high with large confidence intervals, and the Eggert *et al*. (2003) equation estimates tended to be biased low. Bellemain *et al*. (2005) recommended capture-recapture methods over rarefaction curves for abundance estimation because they found these estimates to be close to those obtained independently from radio telemetry. In particular, Bellemain *et al*. (2005) recommended closed-population capture-recapture models found in program MARK over basic Lincoln-Petersen models (section 2.3) because Lincoln-Peterson models (as well as rarefaction curve models [Petit and Valière 2006]) cannot account for unequal detection probabilities. However, Miller *et al*. (2005) stressed that no method works best for all sampling situations. The same assumptions apply for capture-recapture studies based on scat surveys as for capture-recapture methods in general (Box 2). Specifically for scat surveys, it is assumed that all individuals have an equal probability of detection; differences in defecation rates and differences in the likelihood of investigators finding scats between sexes, age classes, and animals of differing reproductive and social status, can bias population estimates.

Flagstad *et al*. (2004) used a scat survey to estimate population abundance of wolverines in Norway, and Hedmark *et al*. (2004) and Ulizio *et al*. (2006) assessed the efficacy of using wolverine scat to genotype individual wolverines. Squires *et al*. (2006, 2007) used the Lincoln-Petersen capture-recapture estimator to estimate wolverine population density in Montana. They used a radio-marked sample of wolverines as the original marked sample, and “recaptured” wolverines by walking along wolverine tracks in the snow collecting hair and scat samples for DNA analysis (Ulizio *et al*. 2006).

6.5 Live-trapping

Live-trapping is the most invasive of the survey methods reviewed here, as it involves physical capture of the animal in baited traps that are checked daily for captures. One of the benefits of live-trapping is that animal identification is nearly certain, whereas hair or scat samples can suffer from genotyping errors and the inability to amplify some samples (Box 8). Furthermore, it gives investigators an opportunity to uniquely mark each individual with ear tags or a radio collar for future identification, which is essential for capture-recapture estimates when individuals cannot be identified by unique pelage patterns. Catch-per-unit-effort (CPUE) data can be used as an index of population abundance, assuming that there is a predictable correlation between number of animals trapped per unit effort and true abundance. Squires *et al*. (2006) reported one wolverine capture in 39–149 trap nights in Montana, Lofroth and Krebs (2007) reported one wolverine

Box 9. Estimating abundance from a single sampling session.

There are drawbacks to scat surveys not present in other small-scale, non-invasive sampling methods. Foremost, the investigator does not know how long ago the sample was deposited; therefore, sampling occasions are poorly defined (Lukacs and Burnham 2005). Secondly, the investigator is likely to find multiple samples from the same individual, and classical capture-recapture models cannot take advantage of this information (Petit and Valière 2006). Thus, rarefaction curves are commonly used to estimate population abundance from scat surveys (Kohn *et al*. 1999, Eggert *et al*. 2003, Wilson *et al*. 2003, Bellemain *et al*. 2005, Lukacs and Burnham 2005). To produce a rarefaction curve, genotyped samples are randomly drawn from the total set of samples, and the number of unique individuals in each random sample is plotted against the number of samples (Figure 5). The curve will reach an asymptote when few unique individuals are drawn relative to individuals previously drawn. The population estimate is the number of individuals in the sample at the asymptote. Several equations have been suggested to estimate the value at the asymptote (Kohn *et al*. 1999, Eggert *et al*. 2003, Petit and Valière 2006). The order that the samples are drawn affects the estimate, so the process is repeated multiple times, with the samples randomly permuted with each repeat. The mean is plotted, and approximate confidence intervals can be estimated using the standard deviation of the mean (Petit and Valière 2006).

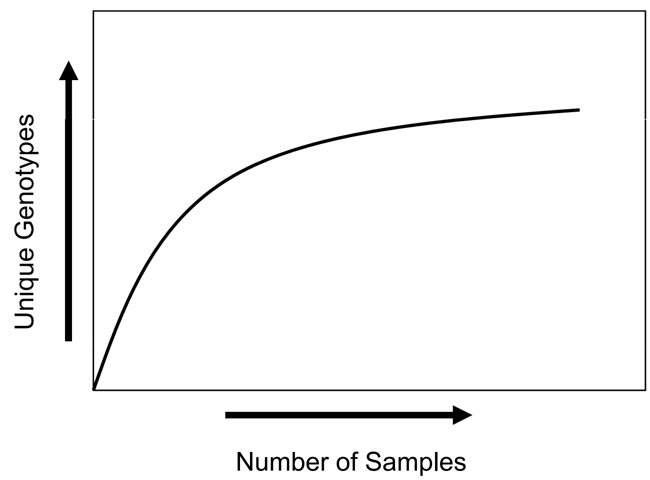


Figure 5. An example of a rarefaction curve.

Rarefaction curves have limitations, in that it is assumed that detection probabilities are constant over space and time (Petit and Valière 2006). Specifically, differences in defecation rates and differences in the likelihood of investigators finding scat between sexes, age classes, and animals of differing reproductive and social status, can bias population estimates. Capture-recapture models in program MARK and program CAPTURE attempt to accommodate capture heterogeneity, and Miller *et al*. (2005) have developed software (CAPWIRE; Table 1) to estimate abundance from studies with a single sample session and capture heterogeneity (but see Bromaghin *et al*. 2007 and Miller *et al*. 2007). When there is no sampling heterogeneity, population estimates obtained from capture-recapture and rarefaction curves are comparable (Petit and Valière 2006).

capture in 48 trap nights in British Columbia, and Dawson *et al*. (submitted) reported 0.83 wolverine captures per 100 trap nights in northern Ontario.

Live-trapping data can also be used to estimate population abundance with capture-recapture models (Finley *et al*. 2005, Lofroth and Krebs 2007; Box 2). Animals trapped in the initial capture session are marked, and the ratio of marked to unmarked animals in subsequent capture sessions is used to estimate population size. Assumptions outlined in Box 2 apply to this method of collecting capture-recapture data. Lofroth and Krebs (2007) used wolverine live-trapping data in British Columbia to estimate population abundance using an open-model estimator, marking 50% of the population in one of their study areas (Krebs and Lewis 2000).

Live-trapping also gives investigators an opportunity to radio collar captured animals. If the target species is territorial, estimates of population abundance and density can be obtained using radio telemetry. Home ranges of resident adults are mapped and, assuming that home ranges of adjacent individuals do not overlap, it is assumed that areas uninhabited by radio-marked animals support an unmarked animal of average home range size, provided that the area contains suitable habitat (Fuller *et al*. 2001, Koen *et al*. 2007). Fuller *et al*. (2001) marked 53–55% of the population to estimate population density of fishers (*Martes pennanti*). Banci and Harestad (1990) used this technique to estimate the population density of wolverines in Yukon. Although this method is expensive and labour-intensive, it can produce accurate estimates of population density (Garant and Crete 1997, Fuller *et al*. 2001).

6.6 Interviews

Similar to large spatial scales (section 4.2), at small scales local knowledge can provide estimates of relative abundance, historical distribution, trapping effort, and spatially explicit locations of harvest and sightings. Locally, this information can be used to monitor qualitative trends in relative abundance and distribution over time if effort is comparable across time. However, at scales larger than local communities, these data become geographically biased, as effort tends to be centered on human settlements and roads. Furthermore, these data indicate presence-only, and an absence of data cannot be interpreted as absence of the species.

6.7 Observations

Opportunistic observations provide presence-only location data which can supplement other data when estimating local species occurrence. The source of these locations can be from incidental or unreported harvest, roadkill, or sightings. Similar to a large spatial scale (section 4.3), opportunistic observations at a small scale must be spatially and temporally explicit. These locations tend to be geographically biased towards areas inhabited by humans. If investigators are interested in using opportunistic observations in these areas, it must be clear that one cannot assume a species is absent where there are no locations.

6.8 Effective population size

Similar to large spatial scales, effective population size (N_e; section 4.5) can be estimated from allelic heterozygosity as an index of actual population size at small spatial scales. As with large-scale survey applications, we consider that assumptions associated with using genetic measures to assess effective population size must still be validated, and therefore we cannot recommend this approach at this time.

Table 5. Pros (a) and cons (b) of several carnivore survey techniques at small spatial scales

a)

Pros	Cameras	Hair snares	Scat	Live-trap
Data have time signature	✔			✔
Species ID is unambiguous without DNA analysis	✔			✔
Individual ID is sometimes unambiguous (without DNA analysis)				✔
Non-invasive	✔	✔	✔	

b)

Cons	Cameras	Hair snares	Scat	Live-trap
Animals are lured from far away (survey area is unknown)	✔	✔		✔
Set-up equipment is expensive	✔			✔
Species and individual ID is expensive		✔	✔	
Equipment can fail to detect species	✔	✔	✔	✔
Electronic equipment can fail	✔			
Animals may become trap-shy or trap-happy	✔	✔		✔
Labour-intensive	✔	✔	✔	✔

7.0 Small-scale Wolverine Surveys: Recommendations

In the following section we have assessed methods described in section 6.0 in terms of their feasibility for surveying and monitoring wolverines in Ontario and other lowland, boreal forests at small spatial scales. We have then made recommendations based on various study objectives.

7.1 Distribution

Estimates of distribution are often in the form of the proportion of sample units occupied, or the probability of a unit being occupied. The variable of interest is presence or absence of the species, and repeated surveys provide estimates of the probability of detection when the species is in fact, present. For wolverines in Ontario and other lowland, boreal forests at a small spatial scale (<100,000 km^2), all methods described in section 6.0 can potentially be used for presence-absence surveys. Occupancy estimates will be more accurate and precise when detection probabilities are high (MacKenzie and Royle 2005). Thus, the ideal method for estimating occupancy for wolverines is one with a high detection probability given a realistic budget and the logistical constraints of surveying wolverines in remote areas.

Similar to large spatial scales (section 4.2), at small scales local knowledge (section 6.6) can provide estimates of relative abundance, historical distribution, trapping effort, and spatially-explicit locations of harvest and sightings. However, interviews (section 6.6) and opportunistic observations (section 6.7) are not reliable sources of presence data in that investigators cannot count on obtaining data, and presence-only data tell investigators nothing about absence of the species in an area. Furthermore, these data are geographically biased, as effort tends to be centered on human settlements. Camera (section 6.2), hair snare (section 6.3), and scat surveys (section 6.4) are labour-intensive, and live-trapping (section 6.5) is even more so because traps must be checked daily. Thus, we do not recommend these techniques if detection (presence or absence) of wolverines is the study objective, especially when snow track surveys are available, which are efficient and much less labour-intensive.

If detecting presence or absence of wolverines is the only objective, we recommend HSM aerial snow track surveys (section 6.1.1) because large areas of rough and inaccessible terrain can be surveyed efficiently and effectively. These surveys require the availability of experienced pilots and observers, and aircraft that are highly maneuverable and can fly relatively low.

7.2 Relative abundance

Estimates of relative abundance often use the rate of detection of animal sign, such as tracks, scat, hair, number of live-captures, or number of photographs at a camera trap, as an index of actual abundance. These methods assume that the number of stations with detections, or the number of detections per station, is higher when abundance is larger. However, indices of abundance require that there is a known and predictable relationship between the index and actual abundance. For wolverines in Ontario, we do not know the true relationship between estimates of relative abundance (rate of sign detection) and true abundance. Moreover, since these techniques (cameras, hair snares, live-captures) are labour-intensive and have the potential to be used in a capture-recapture framework to provide estimates of true abundance, we do not recommend their use be limited to estimating the relative abundance of wolverines in Ontario and other lowland, boreal forests (but see section 7.3).

Interviews (section 6.6) and opportunistic observations (section 6.7) could be used to estimate relative abundance, where number of sightings (or some other metric) is assumed to correlate with population abundance. However, only at very small spatial scales (within human communities) where there is constant effort, would these indices be useful, since observations tend to be geographically biased. Since investigators cannot infer absence of a species in areas where there are no observations, and cannot depend on obtaining an abundance of data, we do not recommend using interviews or opportunistic observations as indices of wolverine abundance at small spatial scales.

Once assumptions have been satisfactorily validated, N_e (section 6.8) could be estimated for Ontario wolverines and compared to estimates for different parts of the province as an index of population abundance. Samples could be obtained from hair snares, harvested animals, or live-trapped animals.

Again, we recommend HSM aerial snow track surveys for estimating wolverine relative abundance (section 6.1.1). This technique assumes that wolverine track detection is correlated with track abundance, and ultimately, with wolverine abundance (see Gaston *et al*. 2000). If track detection probabilities are uniform across the study area, then HSM aerial snow track surveys can be used to compare differences in relative wolverine abundance (i.e. probability of occurrence) in different parts of the study area. Aerial snow track surveys require the availability of experienced observers and pilots, and aircraft that are highly maneuverable and can fly relatively low. If these two requirements are not available, we suggest using ground-based snow track surveys to estimate wolverine relative abundance at small spatial scales.

7.3 Abundance and density

There are two main techniques for estimating wolverine abundance at small spatial scales: aerial track surveys proposed by Becker (1991; TIPS) and Becker *et al*. (1998; SUPE), and capture-recapture. We do not recommend TIPS or SUPE aerial track surveys for estimating abundance of wolverines. If experienced pilots, aircraft, and suitable snow cover are available for these surveys, we recommend HSM aerial snow track surveys to estimate distribution instead, as this technique requires much less air time and has fewer assumptions (Table 3). Although these techniques estimate different parameters (TIPS and SUPE estimate abundance, and HSM estimates probability of occurrence), we argue that from a standpoint of wolverine recovery in Ontario, a snapshot estimate of abundance is not as useful as monitoring abundance over time, in which case monitoring probability of occurrence over time using HSM surveys will be similarly useful as monitoring abundance, and more efficient and cost-effective.

Capture-recapture estimates using live-trapping is invasive, traps are expensive, and it is labour-intensive, as traps need to be checked daily. For these reasons, we do not recommend live-trapping for estimating wolverine abundance in a capture-recapture framework. However, if sufficient resources are available and additional information on wolverine ecology is sought (such as habitat use), then live-trapping for the purpose of radio collaring wolverines may be necessary. In this case, investigators can use mapped territories to estimate wolverine population abundance. However, we do not recommend this method if the only goal is to estimate abundance.

Capture-recapture methods using remote cameras, or DNA from hair snares or scat could be used to estimate wolverine abundance. Precise estimates of abundance with capture-recapture methods increases with higher probability of capture and more repeat surveys. Thus, the ideal method for estimating wolverine abundance in Ontario and other lowland, boreal forests should provide both precise and accurate estimates with minimal cost and labour. We recommend hair snare surveys over remote cameras because the set-up cost for cameras can be prohibitive as multiple camera stations will need to be set-up in each sample unit in order to capture and recapture enough animals to obtain reasonable estimate precision. Proper camera station set-up is an imperative; even if several photos of an individual wolverine are taken, the subject must be positioned such that investigators can identify the individual pelage patterns with certainty, and even then, identification is subjective. Furthermore, cameras can malfunction and batteries may die quickly in cold weather. Hair snares, on the other hand, are relatively inexpensive to set up and are not encumbered by electronics. Mulders *et al*. (2007) used hair snares to survey wolverines in Northwest Territories and reported capture probabilities of >0.5. The cost of DNA analysis of hairs can be expensive, but the number of samples that are analysed can be reduced by excluding those hairs that, by morphology, are not wolverine hairs, and further, by subsampling (section 11.0). We have not tested scat collection surveys for wolverine abundance estimates in northern Ontario. Others (Flagstad *et al*. 2004, Squires *et al*. 2006, 2007) have used scat surveys to estimate wolverine abundance. Wolverine scats are notoriously difficult to locate, putting the utility of this method in question. On the other hand, scat detection dogs provide an efficient method for locating scats (Mackay *et al*. 2008). Although scat detection dogs have been successfully used to detect a variety of carnivore species, wolverines are not among these. If dogs are trained to locate wolverine scat in the future, this method could contribute to abundance estimates at small spatial scales in northern Ontario.

We note here that several methods of detection can be combined into one survey to estimate abundance, such as in Ulizio *et al*. (2006), who used DNA from scat and hair to identify individual wolverines. However, if investigators wish to combine remote cameras with DNA-based methods, investigators must have a way to link pelage patterns and DNA signatures to the same individual. A method for linking photographs to hair samples is currently being tested in Alaska (Magoun *et al.* 2008), and this method may prove to be useful for estimating density in small-scale surveys. Unless wolverines are being live-trapped for other purposes (e.g. radio telemetry), we do not recommend this as a method of detection.

7.4 Monitoring populations over time

Estimates of distribution, relative abundance, and abundance collected over several seasons can be used to monitor changes in the parameter over time, if one assumes that changes in the index are positively and predictably correlated with changes in actual population size. At small spatial scales in northern Ontario and other lowland, boreal forests, ideal estimators will be efficient, cost-effective, and will provide precise estimates that are sensitive to changes in actual population size over time.

Interviews with local people (section 6.6) and opportunistic observations (section 6.7) provide presence-only data in areas surrounding human settlements. We do not recommend these methods as a sole means for monitoring wolverine populations because data are sporadic and tell us nothing about wolverine distribution or abundance in areas where there are no locations reported.

We do not recommend live-trapping (section 6.5) to estimate changes in abundance over time, whether estimates are obtained from capture-recapture or from territory mapping. This method is much too expensive to be used over several seasons to estimate changes in abundance, especially when there are more efficient and less-expensive methods available. We also do not recommend remote cameras (section 6.2) to monitor changes in abundance until further testing of this technique indicates that it can provide precise estimates that are sensitive to changes in true abundance and is cost effective even at small spatial scales. We have not yet tested scat surveys (section 6.4) in northern Ontario, and since we do not know if these abundance estimates would be precise enough to be sensitive to changes in true wolverine abundance over time, we cannot recommend this method for population monitoring at this time. Once assumptions have been satisfactorily validated, N_e (section 6.8) as an estimate of relative abundance could be used to monitor wolverine populations at small spatial scales in Ontario.

We recommend hair snare (section 6.3), aerial HSM (section 6.1.1), or ground-based snow track surveys (section 6.1.2) to monitor wolverine populations over time at small spatial scales in northern Ontario and other lowland, boreal forests. Each of these methods estimates a different population parameter; hair snare surveys estimate abundance, aerial HSM surveys estimate distribution (probability of occurrence), and ground-based snow track surveys estimate relative abundance. Yet, each method estimated over several seasons can be used to index changes in wolverine abundance over time. These methods are the best alternatives in that they are most likely to provide estimates that are sensitive to biologically significant changes in population size.

Box 10. Summary of recommendations for small-scale wolverine surveys.

There are several techniques available to index, estimate, and monitor wolverine populations at small spatial scales (<100,000 km^2) in Ontario and other lowland, boreal forests. For estimating wolverine distribution we recommend HSM aerial snow track surveys (section 6.1.1), as this is the most efficient method for obtaining presence or absence data. For estimating wolverine relative abundance, again we recommend HSM aerial snow track surveys. However, this method requires the availability of experienced observers and pilots and aircraft that are highly maneuverable and can fly relatively low. If these are not available, then we recommend ground-based snow track surveys (section 6.1.2) to estimate wolverine relative abundance. For estimating wolverine abundance at small spatial scales, we recommend capture-recapture methods using hair snare surveys (section 6.3) as this is the most cost-effective of the available methods at the scale necessary to survey wolverines. For monitoring wolverine population change over time, we recommend HSM aerial snow track surveys to estimate changes in distribution. If limited resources preclude the use of this method, we then recommend hair snare or ground-based snow track surveys to monitor changes in wolverine population size over time at small spatial scales in Ontario and other lowland, boreal forests.

8.0 Recommendations for Future Work

Further investigation should be focused on testing the protocol for ground-based snow track surveys for wolverines in Ontario at small spatial scales (described in section 10.0). Further refinement and testing of aerial survey methodology will also be highly beneficial, particularly with respect to testing different aircraft for conducting the surveys proposed here. In addition, techniques that estimate effective population size with genetic data to index and monitor wolverine population fluctuations in Ontario and similar regions should be explored and tested. Monitoring protocols that account for the multiple considerations we have outlined in this document should be deployed in areas of concern for wolverines. Adjustments and additions to our protocols may emerge following further application of the methods, and these would only strengthen wolverine monitoring and research in lowland boreal forests.

Part II:
Protocols and Logistics

9.0 Aerial Snow Track Surveys

Monitoring programs targeted at wolverines will generally be of interest for tracking changes in wolverine populations following habitat change, or monitoring the general status of wolverine populations where recovery efforts have taken place. Discerning changes in wolverine populations is a significant challenge because individuals are sparsely distributed, difficult to detect, and occupy often inaccessible areas. Because of their potentially enormous dispersal distances (Gardner *et al*. 1986, Inman *et al*. 2004) it is not enough to survey wolverines in areas where disturbance has occurred; rather, monitoring programs must consider changes in wolverine populations beyond the boundaries of disturbed forests to fully understand widespread changes in distribution and relative abundance. Most wolverine monitoring efforts will require a large-scale perspective (>100,000 km^2) which presents practical and logistical challenges in most of the wolverine range. The vastness and inaccessibility of northern boreal forests in particular require the use of aircraft. However, published aerial survey methodologies aimed at estimating wolverine population density (Becker 1991, Becker *et al*. 1998, Becker *et al*. 2004) are not suitable for large-scale inventories in boreal forests due to the constraints of applying the technique over large, inaccessible areas (section 4.1.1).

Recent advances in the statistical analysis of presence-absence data that can deal with imperfect detection (Vojta 2005, MacKenzie *et al*. 2006) characteristic of low-density, elusive species offer a means of monitoring wildlife over large areas using track survey data (Sargeant *et al*. 2005). Here, we present a recently published method for determining the extent of wolverine distribution and probability of occurrence by means of aerial surveys of tracks in snow and hierarchical spatial modeling. This method is based on techniques and information gathered through research efforts of the Ontario Boreal Wolverine Project since 2005 and was described in an article published in *Journal of Wildlife Management* (see Magoun *et al*. 2007a).

9.1 Survey design and effort

Researchers interested in surveying wolverines in Ontario or other regions with similar habitat must consider the assumptions inherent in this technique (Box 11). We intend for this portion of the manual (section 9.0) to be useful for both large- and small-scale studies, and we make distinctions in the protocol when it differs between scales. Success of this survey method for delineating wolverine distribution and relative abundance in Ontario depends upon several factors, which include the need to conduct the survey in forested habitats that have sufficient open areas in which to detect tracks, the use of an appropriately scaled study area and sample unit, the distribution of sample units in areas with both high and low wolverine abundance, the survey of a large proportion of sample units, and the use of experienced or well-trained wolverine trackers.

- All animals of interest move and leave tracks during the course of the survey
- Animal movements are independent of the sampling process
- Track occurrence is proportional to wolverine abundance
- Wolverine tracks are identified without error

Box 11. Assumptions of aerial surveys of tracks in snow using hierarchical spatial modeling.

9.1.1 Detection probability

Because there are numerous factors that could affect the survey team's ability to detect wolverine tracks in sample units across large study areas (e.g. local weather, snow conditions, differences in canopy cover, variance in observer skill, etc.), any survey approach must deal with the reality of imperfect detection of tracks (i.e. that one cannot know for certain that tracks are absent in a particular sample unit). These factors can be minimized *a priori* through the design, or *a posteriori* through analytical techniques. First, sample units must be surveyed multiple times, regardless of whether tracks were detected there previously, to obtain estimates of detection probability (section 3.0). Second, the design of survey routes through sample units should attempt to break any correlation with sources of variation in detection probability (MacKenzie and Royle 2005). Differences in the probability of detecting tracks should be primarily due to differences in wolverine track abundance rather than to differences in the ability of investigators to see tracks that are, in fact, present. In this way, differences in the probability of detecting tracks is more likely due to differences in wolverine track abundance than differences in the ability of investigators to see tracks that are, in fact, present. Investigators should spread survey effort spatially and temporally across the study area to avoid sampling units with more abundant wolverine tracks during only the best or only the worst survey conditions. Finally, if variation in detection probability cannot be minimized through survey route selection, these variables can be included as covariates in the model (section 9.5).

9.1.2 Study area and sample unit considerations

Because wolverines occur at relatively low densities, have large home ranges, and may occur in disjunct populations, large areas must be searched to obtain a sufficiently large sample of units with detected tracks. The study area should be divided into a tessellation of hexagons, which serve as sample units (see Figure 6). This allows for up to six neighbouring units of equal distance from the sampled unit and with boundaries of equal length (these neighbours are used during data analysis [section 9.5] to estimate probability of occurrence). We recommend two sizes of hexagons: 100 km^2 for "small" study areas (<100,000 km^2) and 1,000 km^2 for "large" study areas (>100,000 km^2). The distinction between these sizes is somewhat arbitrary; available resources (money and manpower), logistics, and degree of precision desired given survey objectives must also be considered. The "small" hexagon size is based on what we considered to be the minimum home range size of resident female wolverines in Ontario, while the "large" hexagons more closely approximates the average home range size of a resident male wolverine (Dawson *et al*. submitted).

Our experience indicates that sample units with wolverine tracks in Ontario have a clumped pattern across the landscape (Figure 6). If this is typical of wolverine populations throughout their range, the number and distribution of sample units with detected tracks needs to be sufficient to detect such regional patterns.

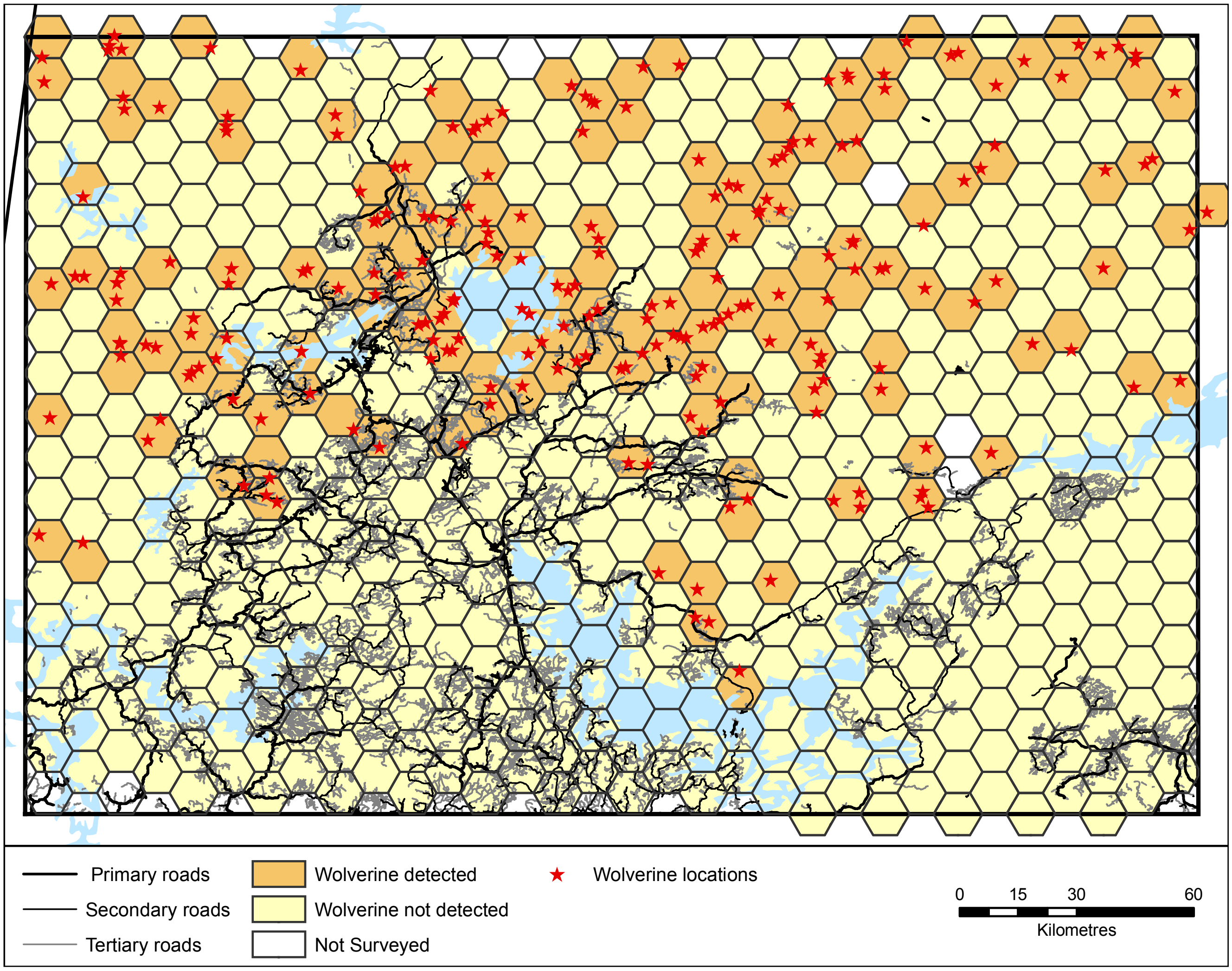


Figure 6. Location of wolverine track detections based on aerial surveys conducted in a 60,000-km^2 study area in northwestern Ontario. This area corresponds to the small (intensive) study area depicted in Figure 4.

In areas where wolverines are expected to occur at very low densities, in widely dispersed core habitats, or in small study areas, smaller sample units could be used to increase the number of units with detected tracks. However, as sample unit size decreases, the number of sample units increases for the same area, requiring more flying time to sample the same proportion of units. Even if flying time is not an issue, it might be more beneficial to increase the number of repeat surveys per sample unit than to decrease sample unit size when study areas are large (Field *et al*. 2005, Mackenzie and Royle 2005). Estimates of occurrence are scale-dependent, with larger units likely to have higher occurrence probabilities than smaller units given the same occurrence distribution (MacKenzie *et al*. 2006). However, larger survey units provide less detail (lower resolution) on spatial characteristics of wolverine occurrence. See section 3.0 for more detail on survey design considerations.

When large hexagon sizes are deployed in large study areas (>100,000 km^2), it is generally not economically feasible to use multiple discrete visits (repeat surveys) to estimate detection probability. In such cases, an option is to divide the flight path through a sample unit into two parts; the first half of the flight, from the side of the unit to the center, would then be considered a separate transect from the second half of the flight (Ray *et al*. in prep.).

When monitoring changes in wolverine occurrence over time using aerial track surveys, researchers should focus their efforts on larger rather than smaller study areas; monitoring population trends in an area much smaller than 60,000 km^2 (used by Magoun *et al*. [2007a]) renders the results too reliant on the fate of individual wolverines rather than the population at large. The same study area should be surveyed over several years, and since the estimated probability of occurrence for each hexagon is influenced by the number of neighbouring hexagons that are surveyed (Ray *et al*. in prep.), effort should be made to survey the same hexagons each year. Additionally, survey effort should be fairly equal across years, such that differences in track occurrence probability between years is due to actual changes in track abundance, rather than differences in track detection probability between years.

If investigators are interested in detecting changes in wolverine occurrence over time in relation to changes in land use, surveys should include areas where resource development is not occurring in addition to the area in question. This will serve to distinguish between baseline population change in the region in areas with little human influence, and population change in areas affected by human land use.

9.1.3 Flight paths

The general design of the survey is for aircraft to follow a flight path that enters one side of a sample unit, passes through the center, exits another side (though not necessarily the opposite side), and then enters the next unit on a heading toward the center of that unit. The design of this aerial survey differs from that of a more traditional design in that straight-line transects are not necessary; the flight path through a sample unit can be sinuous to circumvent large stands of closed canopy forest that block view of the ground. The length, shape, and direction of flight routes depend on weather, day length, location of airstrips with aviation fuel, and number of times the sample units has been surveyed previously.

An idealized survey design for a study area of approximately 30,000 km^2 in size is shown in Figure 7 (with 100-km^2 hexagons as sample units), while an actual rendering of this design is shown in Figure 8. Each day's route is selected from a basic design that includes every other vertical column of survey units (N/S headings) and every third diagonal (for both the NW/SE and the NE/SW headings). Each route should extend across the study area either vertically or diagonally (or both when possible), and routes selected for sampling should provide for even coverage of the study area. Deviations from the idealized design are a result of the desire to end the day as close to the home base as possible, and the limit on the distance (number of hexagons) that can be sampled on a single day given light conditions and distance from base. In this example (Figure 8), routes have been chosen to cover an average of 40 100-km^2 sample units per day for a total of approximately 444 km per day straight-line distance (each hexagon has a diameter of approximately 11 km). We estimate that the survey depicted here can be completed in 12–14 routes, or six to seven days total with two planes. In most cases, however, one can fly more in a day, covering as many as 60 hexagons. This will depend on such factors as the amount of headwind, the length of the day, survey conditions, amount of animal sign, and skill of the survey team. When most of the basic design is included, most units will have five of six neighbours surveyed. As such, it is not necessary for all hexagons to be sampled. It is, however, important to strive for a range of repeat surveys among sampling units (see section 3.2.2). Repeat surveys can be conducted any time, but we suggest that if two surveys through the same sample unit are conducted on the

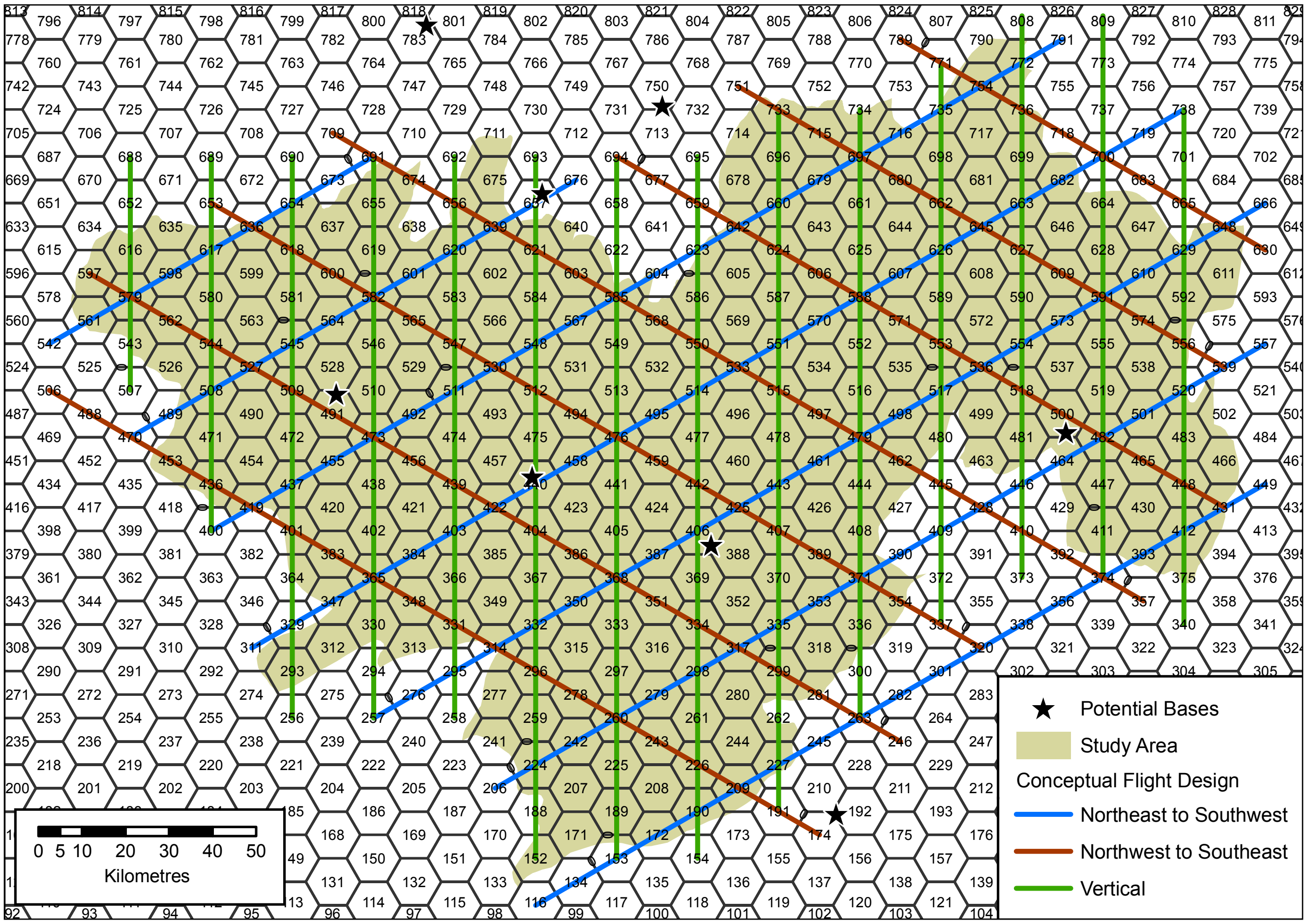


Figure 7. Idealized survey design in a 30,000-km^2 study area with 100-km^2 hexagons, consisting of vertical and diagonal transects.

same day, that a different heading through the hexagon be taken. If identical survey routes are repeated, each survey should be separated by enough time that they can be considered independent, with the probability of detecting the species in one survey not dependent on whether or not it was detected in a previous survey (Mackenzie *et al*. 2006). Our suggestion for this window would be 24 hours; no time separation is necessary if two independent crews are repeating the survey.

It is important that repeat surveys are conducted on sample units regardless of whether tracks were detected there previously or not. Depending on the availability of additional resources, one option is to plot additional routes following the completion of a first stage of surveys to increase sampling to the desired level, either by adding some units that were not sampled in the first stage or increasing the number of repeated surveys for some previously sampled units.

When using large-sized sampling units (1,000 km^2), repeat surveys may not be practical, in which case it is more important to attain fairly even coverage of the area (Figure 9). In practice, modifications to the design due to logistics and changing conditions will yield a mixture of values for the number of repeated surveys, which may range between one and even greater than six, and can be accommodated by the model during data analysis. Note that in Figure 9, more search effort was expended in the eastern part of the study area due to the likelihood of low wolverine densities as judged by harvest records. Holes in effort in the southeastern and northwestern parts of the study areas were due to logistical constraints, but holes such as these should ordinarily be avoided.

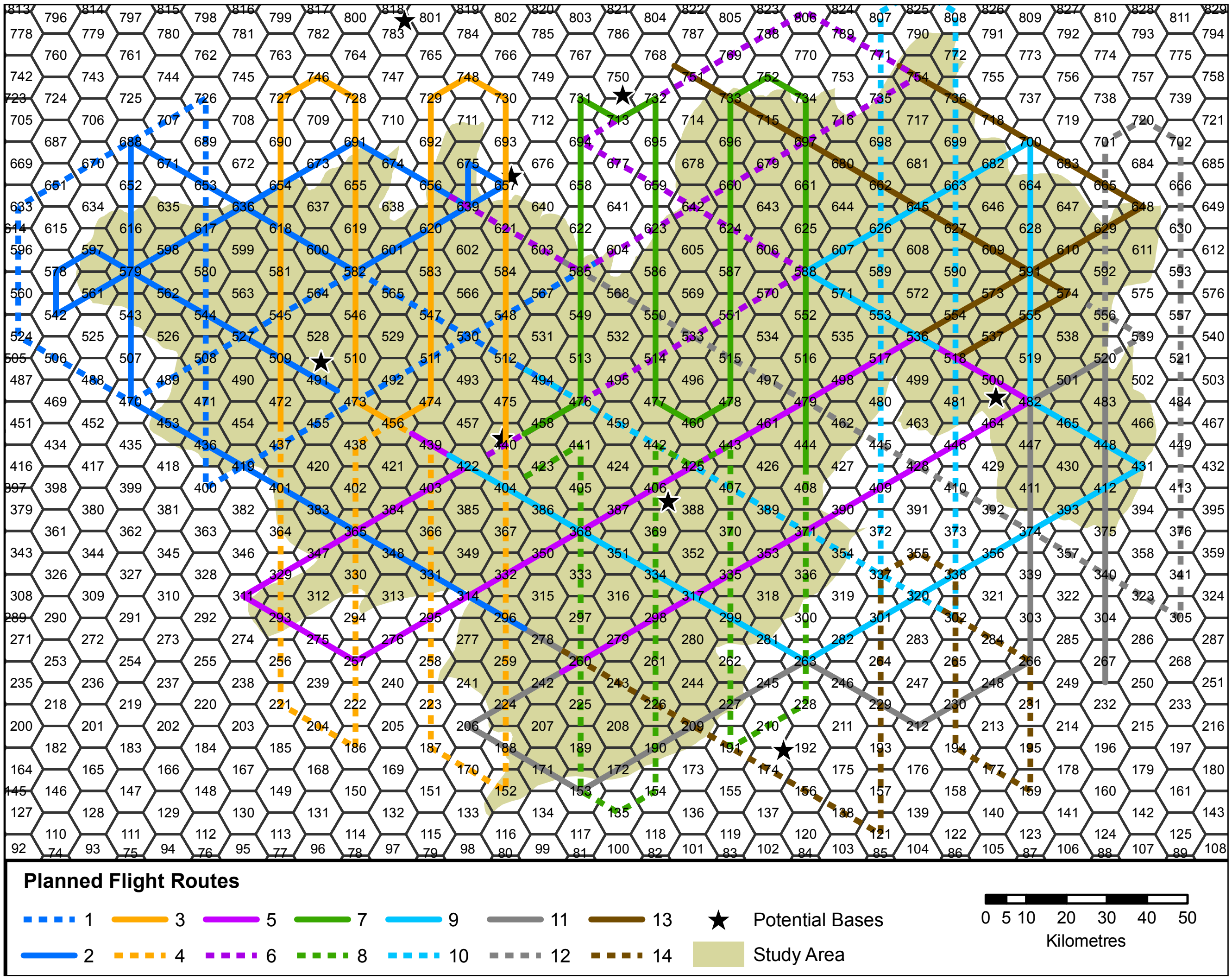


Figure 8. First draft survey design, with candidate remote community home bases. Each day is represented by one colour, with dotted and continuous lines representing pilots one and two, respectively. Note that routes will likely face adjustments depending on weather and other logistical vagaries. Sample units are 100 km^2.

Prior to beginning the surveys, researchers should assign the hexagons unique numbers, plot flight routes through the centers of numbered sample units, and determine the coordinates of the centers using GIS. More often than not, such routes will need to be adjusted during the course of the survey (e.g. if days were cut short because of weather), but for planning purposes it is essential to plot a course in advance to approximate the time required for the complete survey and to inform fuel needs.

The distance across a survey unit is approximately 11 km for 100-km^2 hexagons and 34 km for 1,000-km^2 hexagons but pilots should use a sinuous flight path to maneuver the airplane over open areas along the route rather than maintain a direct line to the center of units. This should minimize time over dense conifer stands where track detection is the most challenging. In our experience, deviations from the designed flight path rarely exceed one kilometre and are usually much less because of the number and distribution of available forest openings in northern Ontario. It is customary to deviate to inspect features such as streams, beaver houses, lake and forested edges, and to avoid closed forest cover and traversing large lakes whenever possible.

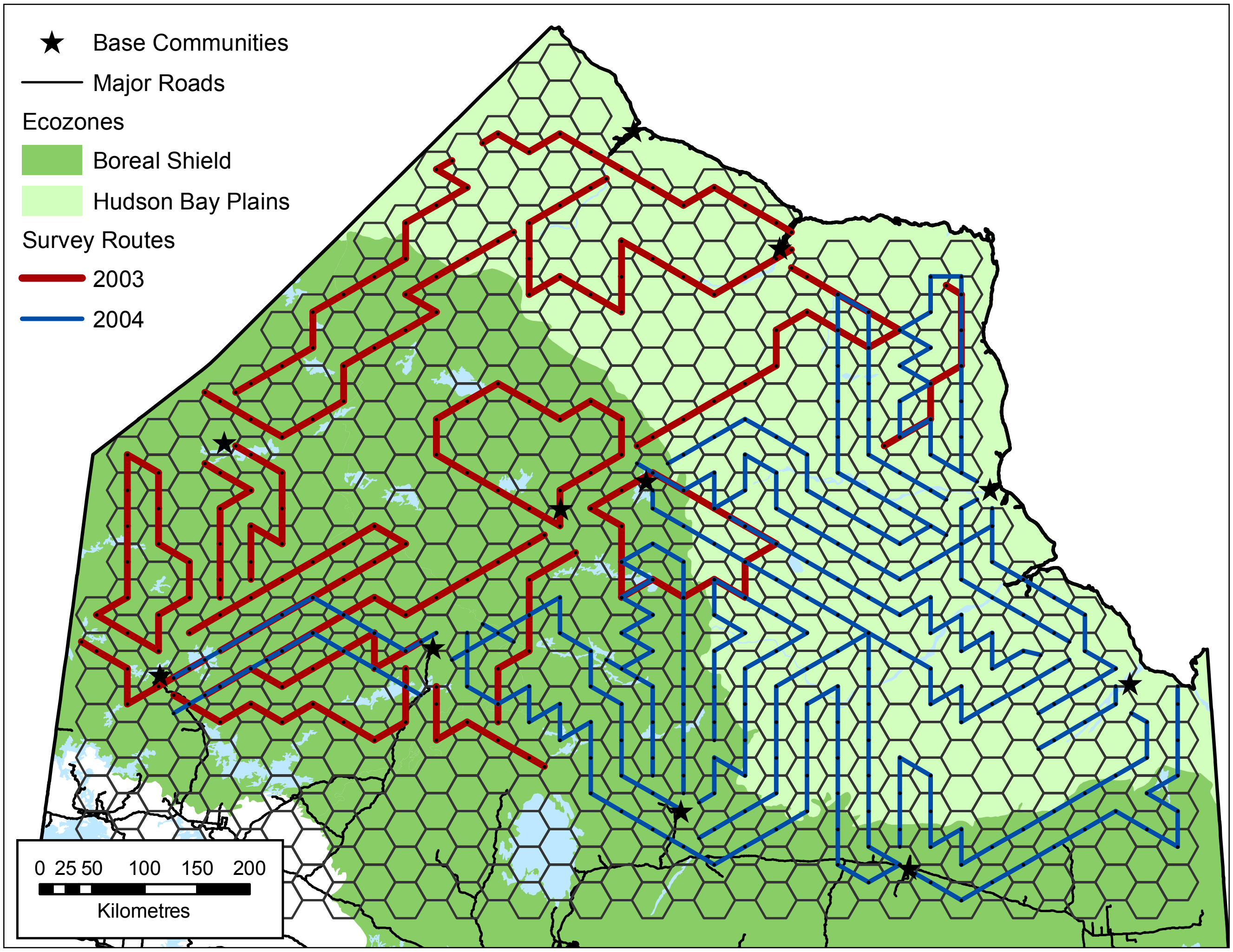


Figure 9. Survey routes in northern Ontario, deployed in the Ontario Boreal Wolverine Project surveys in 2003 and 2004. Sample units are 1,000 km^2.

Open areas or forests (not characterized by closed canopies) where wolverine tracks are detectable are generally well-distributed across boreal forests; in northern Ontario, they comprise >50% of the area. In addition to the numerous open bogs, fens, sparsely covered forest types, and recent cuts and burns, additional areas where wolverine tracks can be most readily spotted include numerous small forest openings, small streams, and forestry roads, as well as dense deciduous forest stands that are leafless in winter.

9.2 Logistics

The logistics associated with planning and executing aerial surveys present a considerable challenge, similar to all survey methods conducted both in winter and in inaccessible terrain. In addition to locating the study area and designing the survey (as described above), pilots need to be engaged well in advance of the survey (six to 12 months), and fuel availability and accommodations must be arranged. More planning is necessary for surveys undertaken in remote areas. The following provides a general checklist:

Survey bases. Once the study area boundaries have been determined, the survey must be designed in relation to the location of municipalities with airports or remote First Nations communities (which generally have airstrips) that can serve as bases during the survey. A community or airport can serve as an adequate base if fuel is available or can be transported, aircraft can be plugged in, lodging is available, and permission is attained from Chief and Council if it is a First Nation community, as well as the responsible authorities for the airport. None of these conditions should be taken for granted and should be confirmed well in advance of the survey.

Aircraft. Although other fixed-wing aircraft can be used, we recommend PA-18 Super Cubs, under the recognition that such airplanes are of limited availability (but see Box 12). This two-seat, tandem aircraft is ideal because it is highly maneuverable and particularly suited for low-level surveys that require tight turns and circling over a point. Moreover, both the pilot and observer can observe the ground from both sides of the aircraft. This aircraft has proven suitability for wolverine track surveys in similar terrain and vegetation types (Becker 1991, Becker *et al.* 1998, Becker *et al.* 2004, Magoun *et al.* 2007a).

Fuel. Adequate fuel for the survey duration must be made available prior to the survey. In areas with relatively large human populations, most airports have 100 low-lead (LL) fuel (or AVGas) available. If bases are in remote communities to which fuel is transported, it will come in 205 L barrels (or drums). To calculate the number of drums required, assume that the aircraft consumes fuel at a rate of 35 L/hr (when using PA-18 Supercubs), and flights last six to seven hours per day. One survey route will likely consist of 40–60 100-km^2 hexagons (or 24–36 1,000-km^2 hexagons). Additional arrangements must be made for transport of fuel to remote communities if necessary. The return of empty fuel barrels must also be arranged. In remote airports, barrels will need to be stored outside. It is best to wait and transport fuel at the relative last minute so that snow does not accumulate on top of the barrels which become impossible to dig out. Barrels should be stored lying down.

Pilots. Survey crews will generally consist of one pilot and one observer. Pilots must have experience at off-airport landings in winter conditions, knowledge about winter survival, experience using topographical maps, willingness to stay in remote settlements for extended periods, and be equipped with necessary items to start the aircraft in cold conditions without electricity. If pilots are brought in from Alaska, arrangements should be made to acquire work permits for the duration of their stay in Canada, and their planes must have the appropriate certification from Transport Canada.

First Nations communities. Investigators should seek permission for overnight stays from all relevant First Nations communities and inform them of the intent to conduct surveys in traditional use areas.

9.3 Budget

Disclaimer. This section is intended for planning purposes only. Values are in 2008 Canadian dollars, and are estimates only.

Pilot costs. Pilots generally have separate rates for ferrying (travel en route to survey area), survey, and down days. For a PA-18 Supercub in 2008, these costs were $190/hr, $225/hr, and $385/day, respectively. Hence, engaging two pilots for 120 hours (seven to eight survey days), including ferry from Alaska to Ontario and assuming a total of 10 bad weather days would cost approximately $34,000.

Fuel. Average price per drum of 100LL fuel in 2008 ranged from $300–400, not including taxes, and depending on whether the barrel was sealed or unsealed. Hence, 20 drums of fuel will cost between $7,000–8,000. If fuel needs to be transported to a remote community, this will come at an additional cost. Pilots will have to change the airplane oil on a regular basis so this will need to be factored into the cost as well.

Box 12. Considerations when choosing aircraft for aerial surveys of wolverine tracks.

Winter aerial track surveys of the type described in this document are most efficiently accomplished from slow-flying (ca. 110–130 km/h), small, fixed-wing aircraft on skis or wheel-skis with a pilot and observer team who are both experienced at track identification. It is advantageous for both the pilot and observer to be able to see the same track at the same time and discuss its identity, which makes two-place, tandem-seat (where the observer's seat is located behind that of the pilot) aircraft such as the Piper PA-18 (Supercub), Christen Husky, Piper PA-12, and other such aircraft are ideal.

Certain features of the Supercub make it particularly suitable for these types of surveys and the preferred choice over other aircraft when it is available:

- The Supercub has become the most widely used wildlife survey aircraft in the world and many modifications have been made to it over the years to improve its performance, safety, and reliability, including larger engines, long-range fuel tanks, higher useful loads, better cabin heaters, stronger landing gear, larger baggage compartments for bulky gear like wing and engine covers, etc.
- The Supercub is safer when flown slowly and close to the ground compared to many other planes, given equal skill level of the pilots (see below). Its stall speed is 68 km/h compared with stall speeds closer to 80-84 km/h for the Husky, PA-12, Scout, and the Cessnas. The Supercub also has a high power to weight ratio, a low gross weight, and can be quickly recovered from stalling with a slight increase in power.
- Although a fully-loaded piston or turbine Beaver has a low stall speed (about 64 km/h), it has a relatively high stall speed when banking, and it is slower to recover and takes more altitude to recover from a stall because of its relatively high gross weight.
- Because aircraft such as Huskies, Scouts, Maules, and Cessnas have higher stall speeds, they also have larger turning radii. It is necessary in this type of survey to have the ability to circle tracks to confirm identification, and these faster aircraft can be difficult to get back to a spot on the ground when the turn is completed. Also, for the pilot to circle a spot continuously requires being further away from the spot or higher over the spot than with an aircraft with a lower stall speed. Slowing down and pulling up the nose of these higher-speed aircraft makes them vulnerable to stalling and spinning.
- The modern piston engines of Supercubs can be safely operated down to about -35 to -40C and they are easily preheated in remote locations with or without electricity. They are relatively easy to put to bed at night, requiring wing and engine covers that are simple to put on without stepladders.
- Helicopters can be used but they are more expensive, have less fuel endurance (meaning that surveyors cannot venture as far from a home base), and do not have tandem seating. Additionally, when doing extensive surveys with helicopters, an engineer may need to be present to perform regular maintenance.

Ultimately, the skill of the pilot is the single most important part of performing snow-tracking surveys. If the pilot is not experienced or interested in identifying tracks, too much time can be lost in communication, circling back and trying to find tracks, and positioning the aircraft for observing problem tracks. In addition, working with a pilot who is not experienced at low-level survey flying (regardless of total experience) can be dangerous.

If winter snow-tracking surveys are to become regular practice in a jurisdiction it would be beneficial to have skilled pilots with optimal survey aircraft (i.e. Supercubs) on staff or available as locally-based contractors. Interested pilots can learn to become good trackers quite quickly and once they have been taught basic skills at identifying tracks, they can develop their skills incidentally to other flying they may be doing during the winter.

It is sometimes helpful to land to identify problem tracks and for this, straight skis are best because wheel-skis provide less flotation, cause more problems if slush (overflow) is encountered, and wheel-skis require a longer takeoff run when snow is deep. Nevertheless, for the ability to land and takeoff from airports in remote regions, or to conveniently obtain fuel at regional airports, wheel-skis are often necessary, so there is a trade-off.

Airport/landing fees. Although modest, costs per day of plugging in airplanes at any airport can accumulate over the course of the survey, so investigators must budget about $100/day/airplane.

Observer wage. Budget $3,700 per month per employee.

Lodging. Budget for $120 per day per person.

9.4 Field protocol

Surveys are undertaken with one or more fixed-wing aircraft with a pilot and one observer in each, both of whom are available to search survey units for wolverine tracks. The pilot can program the coordinates of hexagon centroids for the day's route in his/her global positioning system (GPS) unit in advance, or along the way as desired. The observer records data (tracks and position; Appendix 1), takes detailed notes, and acts as another set of eyes to spot tracks.

9.4.1 Survey conditions

Flights should be conducted on days with sunny or bright, overcast skies and only when wind conditions are favourable for safely maneuvering the aircraft at low levels. Wildlife species other than wolverines can be used as indicators that snow conditions are suitable for registering tracks and that light is suitable for detecting them. Generally, lighting will be best between 10:00 and 15:00. However, this can vary depending on weather, time of year, and location relative to time zones, with surveys beginning as early as 08:30 and ending as late as 17:00. Often the total flight time in a day must be increased to accommodate travel between the base and the starting point of the day's survey and/or from the end-point back to base. Survey teams should wait at least 24 hours following a deposit of at least three centimetres of fresh snow or a windstorm with average wind gusts of >50 km/hr before beginning a survey flight. When weather conditions deteriorate during a flight causing poor tracking conditions, the survey route should be terminated at that point. Survey altitude (ca. 100 m above ground level is ideal for track observations) should be adjusted depending on habitat type and survey conditions. Groundspeed is usually 110–130 km/hr if PA-18 Supercubs are used.

9.4.2 Timing of surveys

The best time for sampling wolverine tracks in snow in Ontario is January, February, or March, when daylight and snow conditions are most suitable. During the course of our fieldwork, however, we observed that detection rates seemed to increase in the period after mid- February, possibly due to increasing movements of young wolverines in late winter (Magoun 1985, Vangen *et al*. 2001) or the increased density of snow in late winter facilitating movement in open areas. We therefore recommend that wolverine surveys take place in late winter whenever possible. Magoun *et al*. (2007a) used a binomial statistic for before and after February 15 as a co-variate in the analysis.

9.4.3 Skill of survey team

Variance in skill level between survey teams is a potential source of bias, as failure to detect tracks that are present may be more pronounced in certain regions of the study area than others. For example, inexperienced teams may not be able to identify wolverine tracks when there are many tracks of other species in an area, especially older tracks, or be able to detect tracks in small forest openings where track segments are short. The skill of a tracking team is measured not only by their ability to distinguish between wolverine tracks and tracks of other species (section 9.4.4) but also by their ability to search for and examine wolverine tracks from aircraft under a variety of difficult tracking conditions. With less experienced teams, the number of repeat surveys required to obtain sufficient detections is likely to be higher and the tracking season longer. We recognize that the use of wolverine track surveys will be limited by the relative shortage of skilled wolverine trackers and by the expense and time necessary to develop tracking skills. Adequate training will be an integral step in the success of this method in Ontario. When observers come across tracks that they cannot identify with certainty, several geo-referenced photographs should be taken with the goal of identifying them later with the help of more experienced colleagues.

One of the benefits of the modeling approach used here is that the models can accommodate variability in observer skill levels, with survey skill considered as a covariate in the model at the data analysis stage. It is ideal to minimize the potential effects of this variable by distributing skill levels equally across the study area and alternating teams in repeat surveys (MacKenzie and Royle 2005).

9.4.4 Track identification

To identify tracks, the survey team must use a combination of track size, shape, depth, and gait, and most importantly track pattern, which includes changes in types and spacing of different gaits because of different habitats, snow conditions, and activities. In addition, body print patterns in deeper snow and behaviour of the animal can be used to help identify tracks. Wolverine tracks are usually easier to identify from the air than from the ground, especially when tracks are not fresh or are mixed in with tracks of other species. See Appendix 2 for a guide to track identification from the air.

9.4.5 Data collection and management

Information on the time and location of wolverine track detections (as well as those of other target species, e.g. caribou, moose, deer, wolves, etc.) should be recorded in a standard data sheet (Appendix 1). Additional information to be recorded are qualitative descriptions of habitat, including significant changes en route, and human sign, including roads and snowmobile tracks. Wolverines are almost always solitary, but when actual animals are seen (including other possible target species, such as caribou, moose, and wolves), the survey team should take photographs to estimate the number of animals and group composition (e.g. cow/calf ratio) where appropriate. As much time as necessary can be spent to verify the identity of tracks, including circling tracks, following tracks to observe changes in track pattern or behaviour, following fresh tracks until an animal is seen, photographing tracks, and landing the aircraft to investigate tracks on the ground if conditions permit. After a track is investigated, the team should return to the route heading. All detected tracks with positive identifications should be considered evidence of occurrence regardless of track age or condition.

While en route, there is no need to keep track of which unit is being surveyed at any given time, except for the first and last unit on each leg of the route (i.e. each straight-line segment flown), because up to 60 units per day will be surveyed. Instead, track locations are plotted and assigned to sample units after the survey is completed. Therefore, previous track history in a sample unit will not affect detection on subsequent surveys of the unit. However, it is important to record the coordinates or waypoints of the entire route surveyed (for example, using the airplane's GPS track log) and the coordinates or waypoint of each track location (Appendix 1). If there was any doubt about the identity of a track, it should not be included in the analysis to avoid false positives (Sargeant *et al*. 2005).

Survey data should be recorded in separate spreadsheets for each route (day). Following completion of the survey, locations will then be plotted and assigned to hexagons. The final spreadsheet consists of a list of all hexagons in the survey and a record of whether wolverines (or any other target species, each with its own column) were detected, were not detected, or the hexagon was not surveyed. Other columns can be filled with information on covariates that will later be incorporated to control for detection probability (e.g. time of survey, year, major habitat type, observer skill). Although this spreadsheet provides the primary input to the analysis described below, a separate spreadsheet for each species, with individual point locations and hexagon units, is useful for mapping locations.

9.5 Data analysis

This aerial survey technique uses readily available software to implement a hierarchical spatial model that estimates probability of occurrence (Magoun *et al*. 2007a). Results rest on the assumption that occurrence probability is greater in units with greater local abundance (Mackenzie 2005) and that the ability of skilled trackers to find wolverine tracks that are present across a vast landscape has a strong positive correlation with the abundance of wolverine tracks, and, consequently, with the relative abundance of wolverines at that same scale.

To examine wolverine distribution based on survey results, we recommend a hierarchical spatial model using Bayesian Markov chain Monte Carlo (MCMC) methods to estimate the occurrence probability in each sampling unit (Banerjee *et al*. 2004). A hierarchical model is a complex model with many dependencies among data and process models. This dependence is constructed by building a large model from several small models which are then linked together.

In the spatial occurrence model for wolverines there are three main models which are linked together to form a large model for making inference on the presence of wolverines. The first model is the “data” model. This component models the detection of wolverine tracks given wolverines are present. The second model is the “process” model. This models the occurrence of wolverines given a spatially correlated covariate (e.g. forest cover). Finally, the random effect model specifies a distribution for the spatial random effect.

MCMC is one of the most effective methods for making inference for parameters in hierarchical models. MCMC takes advantage of the hierarchical structure to draw a sample from the distribution of the parameters (or other quantities) given the data. See Link *et al*. (2002) for a description of Bayesian inference in ecology. The MCMC analysis can be accomplished using the program OpenBUGS (Bayesian inference using Gibbs sampling; Table 1) to fit the model and produce a map of occurrence probabilities. MCMC and OpenBUGS are becoming increasingly common in wildlife applications because they allow researchers to fit far more complex models to data than has been feasible using conventional approaches (Link *et al*. 2002) and are robust to the constraints of these particular sampling designs. In Appendix 3 we provide a description of how data should be set-up for input into program R, and the code is available in Appendix 4, Appendix 5, and Appendix 6.

We refer to our model for wolverines as an occurrence model rather than an occupancy model (MacKenzie *et al*. 2006), reserving the term “occupancy” for areas with resident animals. MacKenzie (2005) stated that a wide-ranging carnivore might be considered to “use” a survey unit if the unit is smaller than its home range or sampling takes place over a long season (i.e. physically present in a unit only at random points in time), but “occupancy” might be a better term if survey unit size is similar to home range size and the sampling season is short. However, wolverine track occurrence may not fall strictly within either of the above categories, regardless of survey unit size and survey season length, because a track may be a single, unrepeated occurrence of a non-resident animal transiting an area not ordinarily used by wolverines (i.e. dispersal or exploratory movements; Vangen *et al*. 2001). Clumping of units that have high occurrence probabilities can be used to identify areas on the landscape that are highly used by a species (MacKenzie 2006, Wintle and Bardos 2006); however, units with high occurrence probabilities that are isolated from similar units may hold little information about wolverine occupancy.

Results from aerial surveys can be used for monitoring changes in wolverine occurrence over time. Here, the metric is probability of occurrence; specifically, changes in this probability for individual hexagons over several years. Magoun *et al*, (in prep.) used simulations to show that this technique could be used to detect changes in the size of wolverine core range area in northern Ontario. They defined core range area as contiguous hexagons with an occurrence probability of ≥0.15. They simulated a reduction in the number of hexagons with detected tracks (assumed to represent a reduction in population size, as track occurrence is assumed to correlate with wolverine abundance), and compared the size of the core range area before and after the simulated population size reduction (Ray *et al*. in prep.).

Box 13. Summary of the protocol for implementing aerial surveys for tracks in snow

- Create a digital map of the study area with numbered hexagons (100 km^2 or 1,000 km^2, depending on the size of the study area) using GIS.
- Determine base communities and make arrangements for landing and lodging of survey team.
- Plan ideal flight paths (50–60 100-km^2 hexagons, or 24–36 1,000-km^2 hexagons per flight, in relation to base communities following design specifications in this manual.
- Determine and measure co-variates for analysis (factors that might contribute to detection heterogeneity)
- Conduct survey flights as described in this manual and record data as in Appendix 1. If weather conditions change and the flight plan needs to be adjusted, record all changes so that later it will be evident which hexagons were surveyed.
- Transfer data to the three tables specified in Appendix 3.
- Open the script in program R as specified in Appendix 3. These codes are available in digital format, along with example data, at: http://people.trentu.ca/jebowman/.
- Transfer results (i.e. mean occurrence probability for each hexagon) to a GIS. Core and peripheral wolverine range can be discerned with a threshold value (i.e. probability of occurrence). For example, contiguous hexagons with a probability of occurrence of ≥0.15 could be considered core range area. Changes in wolverine occurrence over time can be detected by monitoring changes in core area size over time.

10.0 Ground-based Snow Track Surveys

We recommend ground-based snow track surveys for estimating relative abundance of wolverines at small spatial scales (<100,000 km^2) in Ontario and other lowland, boreal forests, in areas where most sample units are accessible by truck or snowmobile. Unlike other survey protocols that we have recommended, we have not tested this method in Ontario for wolverines. Rather, our survey design is modeled after the Alberta Biodiversity Monitoring Institute's winter tracking protocol for monitoring mammals (Bayne *et al*. 2005) and Finnish wildlife triangles (Linden *et al*. 1996). The main difference between our protocol and those of Bayne *et al*. (2005) and Linden *et al*. (1996) is the study objectives: we aim to survey and monitor wolverines (although investigators can choose to record the identity of all tracks encountered), whereas the latter two studies aim to survey and monitor a variety of mammalian and avian species.

10.1 Survey design

Ground-based snow track surveys can be used to count tracks as an index of abundance, based on the assumption that areas with more wolverine tracks have higher wolverine abundance. Triangular transects are traversed and all wolverine tracks that cross a given transect are recorded, as described in Bayne *et al*. (2005). Ideally, the track count will be dependent on the number of tracks present and independent of transect placement and time of year. Differences in the rate of track encounters between two areas or two time periods can be used as an index to monitor differences or changes in wolverine abundance.

Sample unit size. The study area should be divided into a tessellation of 100-km^2 hexagons, as described for aerial snow track surveys (section 9.0, Magoun *et al*. 2007a) for several reasons:

- Survey effort will be distributed equally across the entire area of interest;
- 100 km^2 is the minimum home range size reported in the literature for female wolverines, and while smaller than that of reproducing female wolverines in Ontario (300 km^2; Ontario Boreal Wolverine Project, unpublished data), this size will help to ensure that all wolverines in the study area have the potential to be detected;
- Transect density will be greater than that used by Bayne *et al*. (2005), who reported low detection probabilities for wolverines in Alberta;
- Survey results may be comparable to results of small-scale aerial surveys based on 100-km^2 hexagons (section 9.0, Magoun *et al*. 2007a).

Triangle size and placement. We propose a systematic sampling scheme (Box 4), whereby sample units (hexagons) are systematically and permanently placed throughout the study area. One triangle with a 9-km perimeter would be placed as close as possible to the centre of each hexagon (Figure 10). We propose that the triangle be oriented and shifted within the hexagon to maximize the length of transect that is within potential habitat. Halfpenny *et al*. (1995) suggested a similar sampling regime, whereby surveyors searched 10-km^2 sample units for wolverine tracks in snow, focusing effort on areas most likely to be inhabited by wolverines and utilizing trails and roads. Likewise, Magoun *et al*. (2007a) focused aerial survey efforts within hexagons in areas where they were likely to detect wolverine tracks. If surveys are repeated over several years for monitoring purposes, we suggest that the same triangles are surveyed each time. However, if habitat characteristics at triangle locations are expected to vary considerably over time (i.e. due to forest harvest), then varying triangle locations across surveys might be more appropriate. We suggest triangular rather than straight-line transects so that surveyors spend the entire survey time sampling, rather than spending part of their time walking back to the starting point.

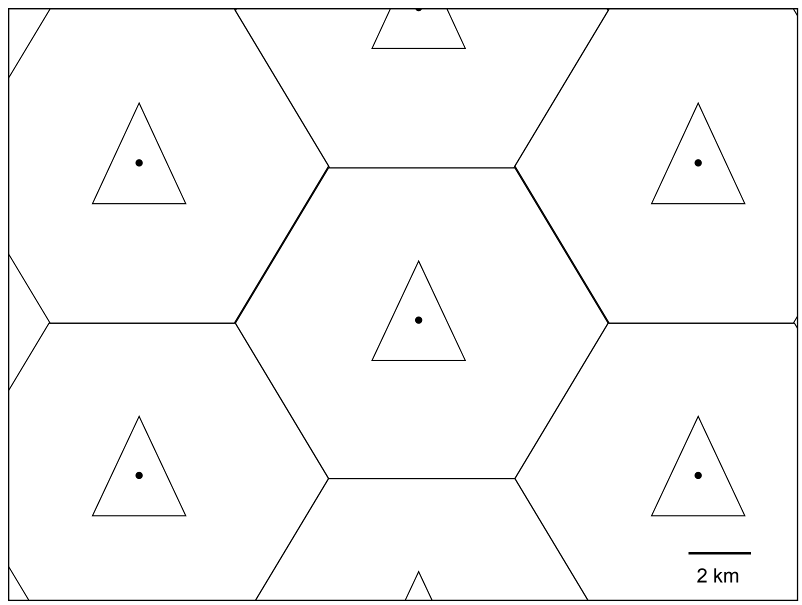


Figure 10. A scaled example of a tessellation of 100-km^2 hexagons with a survey triangle (9-km perimeter) at the centre of each hexagon.

10.2 Effort

It is difficult to predict how many hexagons should be surveyed to provide enough statistical power to detect differences in track counts over time or between areas, as we have not tested this method in northern Ontario. Effort recommendations will ultimately depend on the between- and within-transect variation in wolverine track counts in northern Ontario, which is currently unknown. However, we can suggest a range of estimates of the number of sample units necessary to obtain sufficient power based on variance estimates from other studies in other areas (Appendix 7). We assume that this range encompasses the variation in Ontario. We must stress that the following recommendations are intended only as guidelines upon which a pilot study should be based.

Change in track counts over time. Based on the assumptions outlined in Box 16 of Appendix 7, we used program MONITOR (Gibbs *et al*. 1998; Table 1) to estimate the power to detect changes in track counts with one survey every five years for 10 years (surveys at year 0, 5, and 10; Figure 26 in Appendix 7). In other words, we estimated the probability that we would detect a given difference in track counts over a 10-year period and given a range of possible variations on the mean count. Estimates are based on annual exponential declines of 5%, 3%, and 1%, resulting in cumulative declines of 54%, 37%, and 14%, respectively, over the 10-year period. For the purposes of this study, we consider statistical power of >0.8 to be sufficient.

If the CV (coefficient of variation; standard deviation/mean) of wolverine track counts in Ontario is 10 (as it is in Alberta [Bayne *et al*. 2005]), only when >100 sample units are surveyed is there a >80% chance of detecting an annual 5% decline in track counts (Figure 26a). Only when >200 sample units are surveyed could a 3% annual decline be detected with 80% probability (Figure 26b). However, if the CV in Ontario is <10 (say, five), between 70 and 80 sample units need to be surveyed to achieve adequate power to detect 5% annual declines (Figure 26a), and between 180 and 190 sample units to detect a 3% annual decline (Figure 26b). If resources are such that only minimum effort can be afforded (50 sample units surveyed once every five years for 10 years), CV would have to be less than five to detect a 5% annual decline (Figure 26a), ≤1 to detect a 3% annual decline (Figure 26b), and even if CV=0.5, a 1% annual decline could not be detected (Figure 26c).

Difference in track counts between areas. We assessed the number of sample units per study area that would be required to detect, with 80% probability, varying differences in wolverine track counts between two areas (Figure 27 in Appendix 7). For these simulations, we used a two-tailed test with α=0.1. We assumed that each triangle was surveyed once, and the total CV (within- and between-sample unit variation) fell in the range of 0.5–10.

If the CV for wolverine track counts in Ontario is ≥2.5, there will not be enough statistical power to detect even two-fold differences between two areas (Figure 27a). Only when CV ≤1.5 and ≥100 sample units in each site are surveyed, will there be sufficient power to detect differences of 75% in track counts between areas (Figure 27b). Even if 200 sample units per site are surveyed, there will be <80% probability of detecting small (25%) differences in track counts between sites (Figure 27d).

Study area size. In section 3.1.1, we suggested that small-scale study areas should be, at minimum, 20,000 km^2 in order to encompass a viable wolverine population. We must clarify that, although in some cases as few as 100 sample units need to be surveyed to achieve adequate power for particular objectives, the study area should still be at least 20,000 km^2. In an area this size, there are 200 sample units (if sample units are 100 km^2). Therefore, if only 100 sample units need to be surveyed, one-half (at minimum) of the sample units in the 20,000-km^2 study area should be surveyed.

10.3 Logistics

The following section outlines logistical considerations for executing ground-based snow track surveys to estimate wolverine relative abundance.

Transect delineation. Transect coordinates should be uploaded to a GPS or PDA prior to the survey and saved so that the exact same route can be surveyed during repeat surveys or over multiple years.

Days since last snowfall. The number of days since last snowfall for each hexagon must be recorded, as relative abundance is estimated as the number of tracks encountered per transect per 24 hours or could be used as a covariate in estimating detection rate. The date of the last snowfall could be difficult to obtain for study areas that are remote and potentially far from where the surveyors reside. Bayne *et al*. (2005) outlined several options, including Environment Canada data, local knowledge, information from remote contacts, or on-site assessments.

Optimal timeframe. After an obliterating snowfall (one which covers all tracks completely), surveyors must allow time for new tracks to accumulate; Bayne *et al*. (2005) recommended a minimum of three days and recommended against surveying beyond 10 days since the last snowfall, as the number of tracks accumulated no longer correlated with time, making it difficult to correct for time since last snowfall (see section 10.6). However, if the objective is simply to evaluate the presence of wolverines then the maximum number of days since last snow becomes less important.

Not every day during the survey period will have adequate snow tracking conditions, and investigators will need to factor this into their study plan; Bayne *et al*. (2006) assumed that roughly 35 days between January and the end of March were suitable for snow tracking in Alberta. We anticipate that in northern Ontario, there will be adequate snow conditions between December and March, and 15 days of every month will fall in the "three to 10 days since last snowfall" window.

Snowshoe versus snowmobile. Bayne *et al*. (2006) found more wolverine tracks in Alberta when surveying sample units with straight-line transects via snowmobile than they did with triangular transects via foot. They took advantage of seismic lines and existing trails in Alberta to drive snowmobiles on. In the boreal forest of northern Ontario, roads and trails will rarely be oriented in triangular or linear transects within each hexagon. Thus, it is unlikely that using snowmobiles for surveys in Ontario will be possible, although this will have to be assessed on a study-by-study basis.

Equipment. In addition to basic gear for navigation (GPS, topographic maps) and data recording, survey teams should carry with them necessary equipment in the event that tracks cannot be identified. This includes extra flagging tape to mark when surveyors leave the trail, a camera and measuring tape to record difficult to identify tracks, tweezers and envelopes to collect hair samples (Box 15), and vials to collect scat samples. Surveyor should also carry with them appropriate survival gear (safety considerations are outlined in Appendix 8).

Manpower. In Finland, over 1,500 wildlife triangles are surveyed by over 6,000 volunteers (Lindén *et al*. 1996). Bayne *et al*. (2006) assessed the possibility of engaging volunteers in their surveys in Alberta. Given the remoteness and low human population density of northern Ontario, we anticipate that the number of local volunteers will be small. Therefore, several seasonal employees will likely need to be hired to conduct surveys. Bayne *et al*. (2005) used two employees per transect for safety reasons (see Appendix 8 for other safety considerations). The number of pairs of employees necessary for surveys in northern Ontario will depend on how many sample units need to be surveyed, and how many days per winter will have adequate snow conditions. We address this with an example in section 10.4.

Roads, housing, and fuel. Much of wolverine range in northern Ontario is remote, and it is therefore probable that some or most of the sample units cannot be accessed by road. Investigators need to consider this when choosing their study area. For example, some sample units might be so far from roads that it will not be possible for surveyors to get from the nearest road to the beginning of the transect, survey the transect, and get back to the truck in a reasonable amount of time. In these cases, investigators should consider using snowmobiles or helicopters to access interior sample units.

Investigators also need to consider housing for employees and fuel for vehicles in the study area. Employees will need to be housed close enough to the study area that they can commute to and from the sample units and complete the surveys in a reasonable amount of time. This is by no means a simple task, as study areas at small spatial scales will likely be larger than 20,000 km^2 and existing human settlements and roads in northern Ontario are sparse. Several pairs of employees will likely need to be housed in different parts of the study area and camping may be required. Obtaining fuel for vehicles is also a logistical constraint that needs to be considered when planning a ground-based study in a remote area.

10.4 Budget

The following budget is based on a survey of 100 triangles, assuming that there are four months with adequate snow conditions (December–March), and 15 days of every month have suitable conditions for conducting surveys. Assuming that one triangle can be surveyed per team of two people per day, two two-person teams will need to be hired for a four-month period. We assume that one truck, one trailer, two snowmobiles, and two GPS units will be required per team.

Disclaimer. This section is intended for planning purposes only. Values are in 2008 Canadian dollars, and are estimates only.

Vehicle lease. We estimate $1,400 per month per truck and $1,200 per month per snowmobile for short-term leases. For two field crews for four months, budget $11,200 for trucks and $19,200 for snowmobiles. Investigators should compare the cost of leasing versus purchasing snowmobiles. Vehicle maintenance and trailer rental must also be factored into the cost.

Vehicle fuel. We estimate fuel costs of $2,000 per month (for truck and snowmobile combined). For two field crews for four months, budget $16,000 for fuel.

Supplies. Each field crew will need two GPS units, for a total cost of approximately $600. Budget $500 for safety gear and extra supplies such as flagging tape.

Lodging. This will depend on the location of the study area relative to the permanent dwellings of staff. If a field house is required, however, a typical cost in 2008 would be about $1,000/month/team of two. Also, owing to the large nature of the study area, hotels may be needed on some nights (budget $250 per week for hotels × 16 weeks = $4,000 per team of two). Total lodging budget equals $16,000.

Staff. Budget $3,700 per month per crew member. For four crew members for four months, the total cost would be about $59,200.

10.5 Field protocol

Triangles are traversed, usually by snowshoe or ski, by teams of two surveyors. The coordinates of each track crossing are recorded, even if it is clear to the surveyor that multiple crossings were made by the same individual wolverine. If no wolverine tracks are encountered after traversing the entire triangle, surveyors record this information as well. Repeat surveys are not required. If investigators are interested in using these data to estimate the statistical power to detect trends over time (using program MONITOR; Table 1), repeat surveys will be necessary in order to estimate both inter- and intra-transect variation in track counts.

If a track cannot be unequivocally identified as that of a wolverine, surveyors should temporarily deviate from the transect and follow the tracks. While doing so, surveyors will perhaps find better quality tracks or animal sign that will confirm species identification. If identification is still uncertain, track measurements and photographs with a scale should be taken. Additionally, hair or scat samples encountered along the trail should be collected and brought back for DNA analysis (Ulizio *et al*. 2006). Surveyors should then resume the survey where they left off. This protocol can be amended to include the tracks of all species encountered. See Appendix 2 for a guide to identifying snow tracks of northern Ontario mammals from the air, which also provides a starting point for wolverine track identification from the ground.

In the event that there have not been any recent obliterating snowfalls, surveyors can still complete the surveys if snow conditions are such that tracks can be identified. In this case, transects are traversed twice; the first time, surveyors mark each wolverine track that crosses the transect with a stick, and after waiting a defined amount of time (say, two days), surveyors walk the transect again, recording only new, unmarked tracks (Lindén *et al*. 1996). In this scenario, we suggest also recording the presence of tracks on the first survey, even though time since last snowfall is presumably unknown, as this information could be used for other purposes, such as distribution estimates based on presence-absence data.

10.6 Data analysis

The data for estimating relative abundance consist of a count of the number of wolverine tracks (Figure 11) that cross the transect per triangle per 24 hours since last snowfall. These data, averaged over all transects in a defined study area, are assumed to correlate with actual population abundance (see section 2.2).

The number of tracks encountered per transect must be corrected for the number of days since last snowfall, since tracks will accumulate as time progresses. A common approach is to divide the number of tracks encountered by the number of days since last snowfall, which assumes that track abundance increases at a constant rate over time. However, Bayne *et al*. (2005) found that this was not true for most species: as time since last snowfall increased, the number of *new* tracks decreased (although total accumulation still increased). Therefore, Bayne *et al*. (2005) suggested including days since last snowfall as an independent variable in the model, or estimating track accumulation rates in the field to correct for this directly.

To assess change in relative wolverine abundance over time, the average number of tracks/triangle/24 hours can be compared using a paired *t*-test if two time periods are to be compared, or a repeated measures ANOVA if more than two time periods are to be compared. The test must be a paired test because the same sample units are measured each time, and therefore are not independent.

To compare relative abundance between two areas, the mean track count/triangle/24 hours for each area should be compared using a *t*-test. If no difference between the two areas is found, investigators should perform a power analysis to ensure that failure to reject the null hypothesis of no difference is because there is no difference, and not because of a lack of statistical power. Additionally, investigators could calculate the observed effect size to determine how many more sample units must be surveyed to increase survey power to an adequate level (see Bayne and Hobson 1998 for an example, and section 2.4 for a discussion of statistical power).

Alternatively, these data could be manipulated in the data analysis stage to be used for other study objectives. For example, the presence or absence of wolverine tracks in each triangle could be used to estimate occupancy rate (MacKenzie *et al*. 2002). Because our survey design does not include repeat surveys, investigators could divide the triangle into smaller, equal segments and use each segment as an independent repeat survey to estimate the probability of track detection (see MacKenzie *et al*. 2002). This technique has been used to estimate lynx occurrence in Alberta (Bayne, Boutin, and Moses, submitted). Such approaches relax the assumption that the number of tracks within a triangle correlate with the number of animals present. If investigators intend to use this approach, *a priori* power analyses, as described by MacKenzie *et al*. (2006), should be performed to estimate the number of transects that must be surveyed to obtain adequate statistical power. These data could also be used to estimate relative abundance by dividing the triangle into segments (such as in Bayne *et al*. 2006) and using the proportion of segments per triangle with tracks present as a correlate of abundance.

Figure 11. Wolverine tracks from the ground, with a glove to show scale.

11.0 Hair Snare Surveys

We recommend using hair snare surveys along with capture-recapture methodology for estimating wolverine abundance at small spatial scales (<100,000 km^2; section 6.3). Captured hair can be genetically profiled to identify individual wolverines, and these profiles can serve as genetic tags for counting unique individuals in a population. Although numerous analytical methods are available, we propose an approach similar to Mowat and Strobeck (2000) and Mulders *et al*. (2007).

11.1 Survey design

A variety of techniques exist for snagging hair from carnivores (Kendall and McKelvey 2008). Examples include barbed wire corrals (Mowat 2001), currycomb-rigged traps (Belant 2003), and baited glue-patch traps (Mowat and Paetkau 2002). Based on our review of the literature, and our assessment of what would work best for wolverines in lowland, boreal forests, we recommend a modification of the design employed by Mulders *et al*. (2007) and Fisher (2005). This design involves a strand of barbed wire wrapped around a baited tree (but also see Magoun *et al.* 2008). The objective is to attract a wolverine to the site, have it climb the tree to access the bait, and deposit hair with follicles attached on at least one barb (Figure 12).

Figure 12. A hair snare, showing wolverine hair snagged on a barb.

A network of these hair snares should be deployed in accordance with study objectives (see section 6.3). For example, in our Red Lake, Ontario study, we were interested in identifying the distribution of wolverines. Therefore, we spatially stratified (Box 4) the study area into 20 cells of 100 km^2, based on the assumption that adult female wolverines have a minimum home range size of 100 km^2 (Ontario Boreal Wolverine Project, unpublished data). In this way, each female within the study area would be exposed to our survey. We deployed three hair snares within each of these cells (i.e. the density of snares was three per 100 km^2) which resulted in identifying 15 unique wolverines in an area of about 2,000 km^2.

Finally, we note that a major potential objective of estimating population abundance is to assess changes in abundance over time. Therefore, a one-year, hair snare capture-recapture study might be used as part of a multi-year assessment of population trend. Annual or multi-annual surveys could be planned as part of such a program (Kendall and McKelvey 2008).

11.2 Logistics and Effort

It appears that wolverines do not shed hair easily until late winter (Magoun *et al*. 2007b). As such, we do not recommend deploying hair snares until mid- to late February. Moreover, late spring would be an ineffective time to monitor wolverines with hair snares due to disturbance by black bears (*Ursus americanus*). Thus, the potential effective survey period for this method appears to extend from mid-February until mid-April. Over this period, snares should be checked every 10-day session for bait (see Section 11.4) and for the presence of hair. Missing baits should be replaced immediately. Given the short duration of this deployment, we recommend that snares be completely distributed throughout a study area within a season, rather than cycled through for shorter periods of time. We suggest a design where 10-day periods are treated as sessions for the purposes of capture-recapture modeling. Thus, a two-month deployment would produce about five sessions, with some additional room for setting up and taking down the snares.

Based on the Ontario Boreal Wolverine Project experience, a team of two people working full time with one truck and one snow machine could deploy 80 hair snares across 80–100 cells of 100 km^2 (snare density of one per 100 km^2) within about a week and then revisit these snares to bait and sample them each 10 days thereafter until the end of the approximately two-month survey. The better part of a third month would probably be taken up scouting field sites prior to the survey, so for planning purposes, we suggest that this would be a three-month survey.

Study area size is an important consideration for a capture-recapture assessment of wolverine population size, as two important assumptions of most methods are affected by study area size (Box 14; see also Box 2). As the study area increases in size (and hair snare density concomitantly declines), there is an increased likelihood that assumptions related to homogeneous capture probability will be violated. On the other hand, as the size of the study area is reduced, closure violations become more likely. Therefore, the ideal study area size strikes a balance between these two considerations. We have not yet adequately tested these assumptions in Ontario, so we must make an informed guess as to an appropriate study area size.

Our radio telemetry estimates tell us that the small spatial extent that was used in our previous hair snare work (about 2,000 km^2) likely violated the closure assumption. We propose a spatial extent of at least 8,000 to 10,000 km^2, with one snare in each of the 80–100 100-km^2 cells. Our radio-telemetry data suggest that female home ranges in Ontario are actually >100 km^2; a minimum density of one snare per 100 km^2 would ensure homogeneous capture probability across individuals. Within each cell, hair snares should be placed in suitable locations (with respect to habitat) to maximize capture probabilities (Woods *et al*. 1999). We emphasize that one snare per 100 km^2 should be considered a minimum, and if logistically possible a greater snare density should be used.

Realistically, deployment of a large network of hair snares requires road or trail access, where trucks or snow machines can be used. However, investigators should recognize that roads and trails lead to biased population estimates and unrepresentative vegetation (e.g. Betts *et al*. 2007). Therefore, care should be taken to gain as much access as possible to interior sites. In the Ontario Boreal Wolverine Project, we worked in a managed forest landscape, and had our best success detecting wolverines in coniferous riparian corridors left behind following logging. These corridors appeared to funnel wolverines through the landscape, making detection more probable. At a local scale, such sites should be targeted for sampling (Woods *et al*. 1999).

Box 14. Typical assumptions of capture-recapture models.

Closure assumptions. Many capture-recapture models assume that the studied population is closed. This includes demographic closure, where there are no births or deaths during the study, and geographic closure, where animals do not move in and out of the study area boundary (White *et al*. 1982).

Homogeneity of detection assumptions. There are at least three types of variation in capture probability. First, trap-shy or trap-happy behaviour can lead to unequal capture probabilities among individuals. Second, time may lead to varying capture rates, for example owing to seasons. Finally, unexplained heterogeneity may exist (see Box 2).

Although it is not possible to know in advance whether assumptions of capture-recapture models have been violated, a benefit of using models such as those available in CAPTURE or MARK (White and Burnham 1999; Table 1) is that these assumptions can be tested.

It is worthwhile to note that there is a potential for wolverines to become habituated to the hair snares (i.e. they learn that they cannot get the bait, and therefore do not climb the tree and leave a hair sample) or to learn to climb the tree without touching the barbs. Both of these scenarios would result in a heterogeneous capture probability, where previously captured individuals are less likely to be captured twice.

11.3 Budget

Principal budget items for a hair snare survey during one year include the following, based on a team of two people working full time with one truck and two snow machines for three months (two-month survey plus one-month planning and scouting), deploying 80 hair snares across 80 cells of 100 km^2 (one snare/100 km^2), with snares revisited every 10 days.

Disclaimer. This section is intended for planning purposes only. Values are in 2008 Canadian dollars, and are estimates only.

Vehicle lease. At a minimum, a truck is required for accessing hair snares. Typical short-term lease costs for a truck in 2008 were $1,400/month, and therefore $4,200 for a three-month study. Depending on the study design, snow machines might also be required, and the cost for these would be over and above the truck. Vehicle maintenance must also be factored into the cost.

Vehicle fuel. For a truck and two snow machines, budget $2,000 per month for gas.

Hair snare supplies. Principally, these include double stranded barbed wire, nails and spikes, a hammer, bailing wire (for wiring bait), and commercial trapper's lure. Budget $1,500. GPS units and safety gear must also be factored into the cost.

Lodging. This will depend on the location of the study area relative to the permanent dwellings of staff. If a field house is required, however, the typical cost in 2008 would be about $1,000/month. Also, owing to the large nature of the study area, hotels may be needed on some nights (budget $250 per week for hotels × eight weeks = $2,000). Total lodging budget equals $5,000.

DNA profiling. Based on our experience in Ontario, a two-month deployment of 80 hair snares could be expected to generate about 400 hair samples that cannot easily be distinguished morphologically from wolverine hair. Therefore, we suggest planning for about 400 hair samples to be processed by a DNA lab for confirmation of species at a cost of $20 per sample, or $8,000 total. Of the 400 hair samples, perhaps 200 would be identified as wolverines, and would need to be sequenced at microsatellite markers at an additional cost of $30 per sample (total $6,000).

Staff costs. If contract staff are hired for this work, a team of two people will cost about $22,200 for three months.

11.4 Field protocol

The first phase of a hair snare survey would be to obtain digital forest maps of the proposed study area. These can be used for an initial spatial stratification (i.e. the establishment of sample units such as 100-km^2 hexagonal cells). The objective of the stratification is to have the snares more or less evenly distributed throughout the study area. Within each cell, an even number of snares should be deployed, and they should be deployed at sites likely to attract wolverines. Based on this digital stratification, sites should then be scouted and selected in the field. Snares can be established at this time, and left unbaited. The coordinates of each site should be marked with GPS for subsequent mapping and analysis.

To set-up the hair snare, researchers should locate a tree with few or no branches low on the bole, and wrap 10–15 m of double-stranded barbed wire (with four-point barbs) around the bole from ground level to about 2 m in height. The barbed wire should be nailed into place such that stiff lobes of wire remain sticking out from the bole (Figure 13). Above the barbed wire at a height of 2 m, bait should be nailed with spikes or wired to the tree. Bait should be large and frozen. The bait and tree can also be scented with trapper's lure such as beaver castor. As much as possible (and depending on objectives) baits and lures should be standardized among snares.

To commence the survey, large, frozen baits (roughly 30 cm x 30 cm) should be obtained, for example beaver (*Castor canadensis*) carcasses from trappers, or road-killed cervids (such as deer [*Odocoileus virginianus*]). Baits and lures should then be deployed at hair snares over as short a duration as possible (less than one week for two people to deploy 80 snares). In our experience, 15 or 20 cm galvanized spikes can be used for putting holes through baits. It is then best to use these holes and bailing wire to wire the bait to a tree. Using frozen baits makes the wiring procedure easier. Barbs should be inspected at this time to ensure they are free from hairs prior to commencement of the survey.

Hair snares should then be revisited at least once per 10-day session, rebaited if necessary, and each barb should be carefully inspected for hair. Hair samples should be collected, labeled, and stored (Box 15). At the end of the survey, a final search of the snares should be completed. Snares can then be removed using a hammer to pull out the nails.

Figure 13. A technician for the Ontario Boreal Wolverine Project demonstrates a hair snare set for wolverines in northern Ontario, showing barbed wire wrapped around a tree and bait placed from above.

11.5 Data collection and management

A spreadsheet should be established for daily records of the snares deployed, baits deployed, and the snares checked. Even snares that are checked but have no bait deployed and no hairs identified should be noted in the spreadsheet. This spreadsheet should be maintained on a daily basis if possible. Otherwise, this information should be carefully recorded on an appropriate data sheet.

Each hair that is collected from a barb (Box 15) should also be recorded in this spreadsheet. In most cases, the species that deposited the hair will be unknown. Where species is known, these hairs should still be collected, and the potential identity should be recorded as it will potentially reduce costs associated with genetically profiling wolverines. It is also possible to screen hair samples in a lab to identify species prior to submitting them for genetic profiling. Hair shaft morphology can be studied microscopically to make species identifications (Adorjan and Kolenosky 1969). If following this approach, hair should not be thawed and refrozen, as this process may introduce bacteria. The distal end of hair shafts contain no DNA and can be clipped from the proximate end of the sample to make this morphological assessment without reducing DNA availability. In general, DNA labs prefer at least 15–20 hair follicles, and a minimum of five follicles, in order to extract sufficient high-quality DNA for analysis.

Hair should be collected from barbs such that hair from each barb in a snare is packaged and labeled separately. Small index cards should be purchased or made so that double-sided tape is sticking to one side of the card, with wax backing still in place on the facing side. The wax backing can be lifted, and distal ends of hair from a barb (and not follicles) are stuck to the tape. The wax backing can then be re-stuck to hold the hair in place. It is very important that the follicles are not stuck to the tape, but rather are left sticking out. A single card so employed can then be inserted into an envelope, and the envelope labeled with appropriate information such as snag number and location, barb number (barbs should be numbered in a consistent way), date, time, putative species, and collector name. These envelopes can be stored at room temperature for short periods of time (weeks or months) providing that conditions are dry. When possible, the envelopes should be stored in a freezer at least -20° C. It is important that hair samples be stored in paper envelopes rather than plastic, etc., to prevent moisture from decaying the sample.

Box 15. Collecting and storing hair samples for DNA analysis.

11.6 Data analysis

11.6.1 Genotyping

Samples should be sent to a DNA lab for species identification and profiling at microsatellite markers. These are rapidly evolving markers that can be used for identification of individuals (Box 5). Markers should be sufficient in number and sufficiently variable that analysts are confidently able to distinguish between individuals (McKelvey and Schwartz 2004b, Paetkau 2004). The Ontario Boreal Wolverine Project has worked with the Natural Resources DNA Profiling and Forensic Centre at Trent University in Peterborough, Ontario, where wolverine samples have been profiled at 11 microsatellite loci. An ancillary benefit of such analyses is that information can also be obtained about genetic diversity and effective population size (see section 4.5). It is important for researchers to contact genetic labs to gauge interest and availability in participating in genetic analyses of samples prior to the start of the study. Researchers should also figure in time required for analyses into the study plan.

11.6.2 Capture-recapture models

Closed capture-recapture models involve estimating initial capture probability, subsequent recapture probability, and finally population size (Box 2). These quantities can be estimated using encounters with unique genotypes as indicative of captures and recaptures. A number of studies have used this type of design in recent years with mixed success (Mowat and Strobeck 2000, Boulanger *et al*. 2004, Mulders *et al*. 2007). The most important issues appear to be meeting model assumptions (Box 14) and achieving sufficient power. Mulders *et al*. (2007) provide an excellent overview of using this approach to estimate wolverine densities in the Northwest Territories. The Ontario Ministry of Natural Resources currently implements a capture-recapture hair snare design to estimate black bear abundance.

Programs such as MARK (White and Burnham 1999) or CAPTURE (which is available within MARK) enable the testing of various assumptions associated with these models, and application of the most appropriate model given the study conditions. Detailed documentation for implementing MARK is available online (Table 1).

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Appendices

Appendix 1. Example of a data collection sheet for aerial snow track surveys.

Ontario Wolverine Aerial Survey Data Sheet 2008		**Page** 1 of 8	**Temp (°C):** -22	**Last snowfall:** Yesterday morning, 5 cm	**V= Visual observation**
Date: Feb 16 2008	**Start:** 0920	**Pilot**: Smith	**Tracking Conditions (circle):** Excellent Good Fair Poor		**F= Fresh tracks** **O= Old tracks**
Route #: 5	**Finish:** 1400	**Observer:** Johnson	**Sky (circle)**: Clear Partly cloudy Light overcast Heavy overcast		**Wind:** Light, no turbulence

Target Hex	**Distance**	**Latitude**		**Longitude**		**Wolverine**	**Wolf**	**Caribou**	**Moose**	**Lynx**	**Fisher**	**Other**	**Comments:** (habitat, activity, human sign, etc.)
S-296c													
296	98.7								F		✔		A lot of moose in cuts and burns
	81.3								F				A lot of moose in cuts and burns
	80.5	51	12.90	94	19.00		F						~6 on Peisk Lake
	71.8	51	15.36	94	22.95			F/O				✔	Caribou tracks and cratering on lakes (marten tracks)
	70.3	51	15.80	94	25.35			F/O					Caribou tracks and cratering on lakes; mature forest around lakes
	27.6												Snowmobile tracks
470	42.2	51	26.09	94	30.82		F~2	V3			✔		Caribou tracks and cratering on all lakes; mature pine/spruce
	20.4	51	27.16	94	32.40			F					Caribou tracks on all lakes; mature pine/spruce
	11.5	51	31.75	94	30.58		F~2		F				~2, into a burn
688	11.0										✔	✔	Hares in burn

The datasheet (above) is a sample datasheet that can be used to record data during an aerial survey. Note that the latitude/longitude can be replaced by the waypoint number. This particular example is from a survey undertaken in northern Ontario; the hexagons, or sampling units, were 100 km^2 (see Figure 14). The flight route should be decided by the beginning of the day. Although all flight routes should be plotted out prior to the survey, it is not uncommon to adjust these routes from one day to the next. It is helpful to have a printout ready with all the hexagon centers in the study area so that flight routes and waypoints can be easily constructed and transmitted to the pilots on the fly. It is vital to keep an up-to-date database recording actual survey effort with hexagon identities (numbers) and coordinates so that the route can easily be plotted, and repeat surveys and overall effort can be calculated.

A day's outing (about seven hours in duration) is assigned a route number; if more than one airplane is out, each survey team assigns a unique route number to their data. The weather conditions of the day should be recorded, as well as the time and amount of the last snowfall. The pilot and observer names, the date, and finally the start and finish times of the survey should be noted at the top of the first survey sheet of each route.

The most critical information to record en route is the GPS position and identity of tracks of target species. There is, however, a wealth of other useful or important information that can be easily recorded along the way. These include back-up information related to the location of tracks if GPS information is misrecorded, the occurrence of other species of interest, and habitat descriptions. In this sample sheet, wolverines were the principal target species, but caribou and wolves were also of high interest, with moose and lynx observations of secondary interest. We decided in advance, therefore, that we would take latitude/longitude coordinates of wolverines, wolves, and caribou (primary target species) but would note the presence of moose, lynx, fisher, and relative abundance of hare (secondary target species) along the way. It is important to ensure that all survey teams agree in advance on the priorities of the survey so that the same quality of data are recorded and that the priority species receive the most attention if there is a lot of track activity.

Except for the starting hexagon, which is noted with "s-" at the beginning of the datasheet, the *target hex* column refers to the hexagon to which the plane is heading along a straight-line path before it changes direction. In this sample, the route started in hexagon 296, headed in the direction of the center of 470 (Figure 14), after which it changed direction en route to hexagon 688. The distance column is a place to record where along the straight-line flight path between target hexagons the data are recorded. This serves both as insurance against loss of data due to misrecording of latitude/longitude coordinates, but also gives information about where secondary target species were located (but coordinates were not recorded) so that approximate locations can be plotted later if desired. It should be noted, however, that both the latitude/longitude and the distance information can easily be replaced by the waypoint number. Waypoints can generally be downloaded from airplane GPS units at the end of the day and matched with data taken by the observer.

Additional information to be recorded is whether the track is fresh (F), old (O), or both (F/O) if there are several tracks. If the animal is visible (and not just the tracks), the observer should note "V" and the number of individuals. For wolves, sometimes the number of individuals in the pack can be estimated from the tracks and those data should also be recorded. For secondary target species, coordinates are not necessary and are up to the discretion of the observer.

Other columns can be added; for example, we found it important to keep track of the location of human infrastructure (e.g. roads, buildings, snowmobile trails, communities, mineral exploration camps or other activity) along the route. The extent to which these are of interest will depend largely on the relative remoteness of the site and how much of this information is already accessible through available GIS layers. The comments column provides an opportunity to record qualitative descriptions of habitat or changes in terrain or forest cover, changes in survey conditions or weather, and descriptions of the tracks.

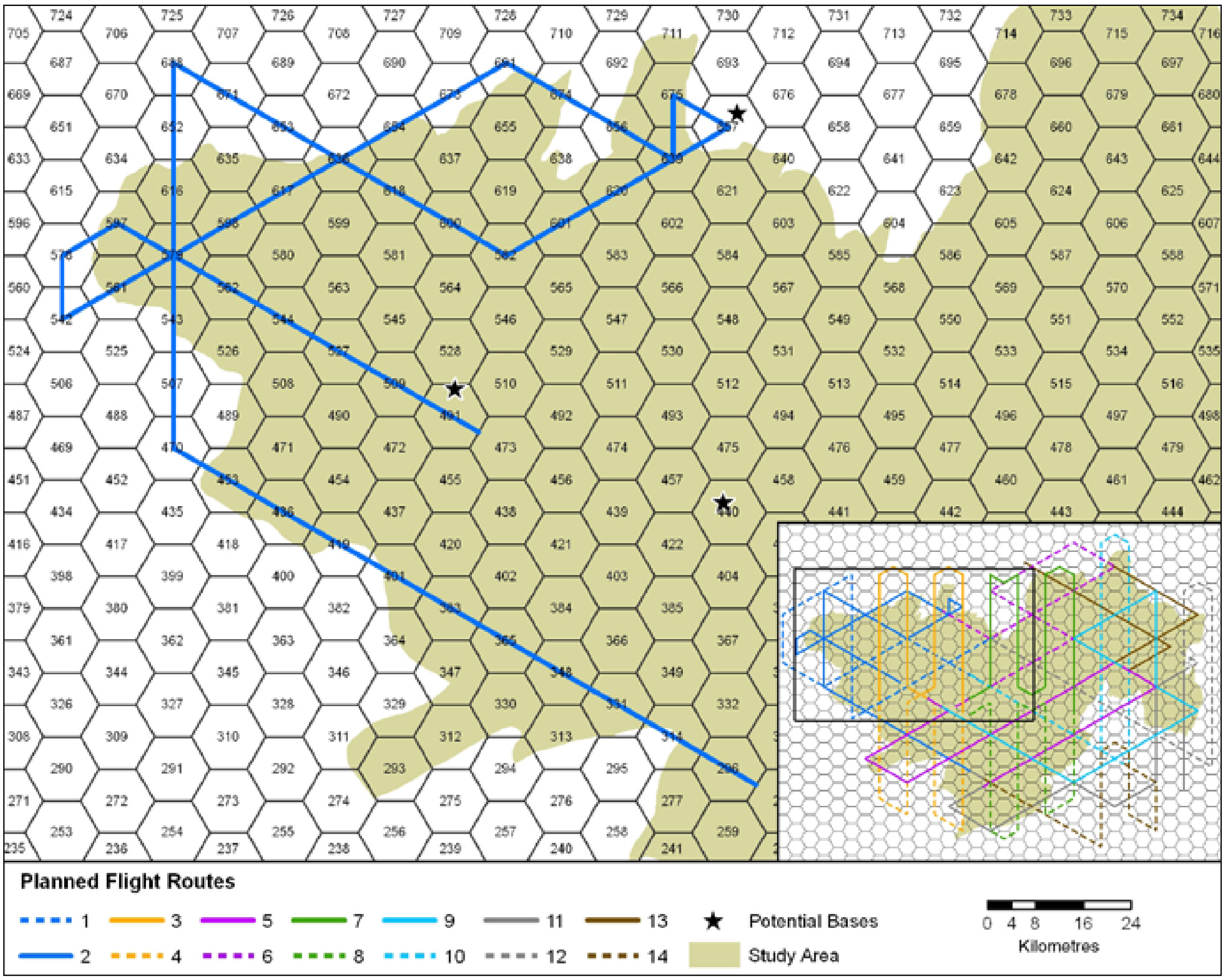

Figure 14. An example of a flight plan, starting in hexagon 296 and ending in hexagon 491. See an example of a portion of a data collection sheet for this flight (above).

Appendix 2. Identifying snow tracks of northern mammals from the air.

By Patrick Valkenburg and Audrey J. Magoun
Wildlife Research and Management (WRAM)
3680 NON Rd.
Fairbanks, Alaska 99709

Aerial tracking of animals in snow is a science and an art. Unlike tracking from the ground that often relies on identification of individual footprints, aerial tracking is based primarily on the appearance of track patterns. In many ways it is easier to identify tracks from the air than from the ground because observers have a vertical view of a large segment of tracks and can follow animals for long distances to observe their behavior. It is also sometimes possible to follow the tracks directly to the animal if there is doubt about its identity. On the other hand, it is seldom possible to observe footprints and count toes from the air (except with wolves and ungulates) and this can sometimes be advantageous.

Winter aerial track surveys are most efficiently done from slow-flying (about 110–130 km/h), small, fixed-wing aircraft on skis with a pilot and observer team who are both experienced at track identification (see Box 12). To observers in small aircraft, the visual appearance of an animal's tracks in snow is dependent on altitude, depth and hardness of snow, age of the snow surface, melting, wind, and lighting conditions. All of these factors can change during a survey, from day to day, and gradually or abruptly over the course of a winter. The gait (i.e. track pattern) that an animal tends to use also depends to a great degree on snow density. The ability of observers to distinguish tracks under these varying conditions is largely a function of the observer's and the pilot's previous exposure and currency of experience. Before trying to learn to track animals from the air, we recommend that observers become thoroughly familiar with the tracks of animals on the ground. Without doubt, the best way to learn tracks is to participate in trapping furbearers or to accompany trappers in a variety of habitats and snow conditions. Familiarity with animal behavior and biology is also very helpful in identifying difficult tracks. Animal behavior and biology can be learned with a combination of reading and observing. It is important to recognize that even experienced observers can misjudge the size of an animal's track from the air because of subtle, unnoticed, changes in altitude. Observers should continuously calibrate their perspective by observing the apparent size of common and easily identified tracks such as those of snowshoe hare, marten, and otter. Size of animals can also vary from region to region. For example, moose and wolves in Alaska are larger than in the Canadian boreal forest, and woodland caribou in the boreal forest are relatively large compared with some other caribou. It is therefore more likely to confuse caribou and wolf tracks in Alaska than in the Canadian boreal forest. Conversely, moose and caribou are easily confused in the Canadian boreal forest but this is not much of a problem in Alaska.

It is important to realize that not all tracks can be identified from the air (or from the ground). Sometimes track segments are too short, snow has melted too much, or tracks are covered with too much snow for positive identification. It is sometimes necessary to call a track unknown and just move on. However, observers must be careful not to bias results by ignoring difficult tracks through lack of experience. Appearance of tracks can be quite variable, and there are many things to keep in mind while conducting surveys and observing tracks. The following guide and photographs will hopefully serve as a useful guide to experienced observers and as an aid to those who are just learning the technique. We have brought together a combined total of about 25 years of experience trapping wolves, marten, mink, lynx, wolverines, muskrats, beaver, otter, and ermine in Alaska, and many years of hunting experience in snow in Maine, Alaska, and the western United States. In addition, we have conducted approximately 5,000 hours of winter survey flying as pilots and observers in Alaska, Ontario, Manitoba, and Labrador. We also have over 40 years of collective experience catching, radio collaring, and studying wildlife, especially wolverines, marten, wolves, caribou, moose, and other northern mammals in Alaska and Ontario. We have learned a great deal from many other pilots and biologists with the Alaska Department of Fish and Game, and trappers in Alaska and Ontario. We particularly thank Marty Webb and Rick Swisher.

Table 6. A guide to identifying snow tracks of northern mammals and birds from the air (Alaska, Ontario/Manitoba, Labrador).

Species	Relative track size	Usual track cluster	Primary distinguishing characteristics	Species most easily confused with	Remarks
Bison (*Bison bison*)	Very large	Single footprint	Tracks offset, with almost continuous, heavy drag marks, rounded hoof prints; usually in groups; grazing on grasses and sedges	Moose	
Moose (*Alces alces*)	Very large	Single footprint	Tracks offset, with drag marks, sinking deep; large, oval beds; feeds on hardwood species or cedar; often one or two individuals in group; seldom galloping (leap marks)	Woodland caribou; bison	
Caribou (*Rangifer tarandus*)	Very large	Single footprint	Tracks offset, with drag marks, sinking deep; cratering in pine, spruce, or tundra; rounded beds on lakes; often more than two individuals in group; often galloping (leap marks)	Moose in boreal forest; wolves in Alaska	
White-tailed deer (*Odocoileus virginianus*)	Large	Single footprint	Tracks offset, with drag marks, sinking deep; often see leap marks; meanders along lake shores, frequently feeds on cedar or hardwood browse	Wolf; coyote in boreal forest	
Wolf (*Canis lupus*)	Large	Single footprint	Tracks in line, sinking deep; often travel on roads, trails, rivers, and lakes, often more than two; often see leap marks	Deer; caribou in Alaska; coyote; lynx	
Lynx (*Lynx canadensis*)	Large	Single footprint	Almost always walking; oval, clean tracks, with very few or no drag marks	Red fox; walking wolverine; single wolf	February breeding season often has two animals traveling together
Coyote (*Canis latrans*)	Medium	Single footprint	Very similar to red fox but usually larger and sinking deeper in snow, often see leap marks, especially in deep snow; often in pairs; commonly associated with snowshoe hare concentrations; often follows trails, roads, lakeshore, and river banks	Red fox; wolf	February breeding season
Red fox (*Vulpes vulpes*)	Medium-small	Single footprint, but two by two (diagonal) is common on hard surfaces with little snow	Round tracks, usually walking, except in February and March when galloping tracks become much more common because of the breeding season; does not usually sink much, even in deep snow; often meanders in brushy areas and lake margins; usually single animal, except in February when often in pairs	Coyote; lynx; marten and fisher when two by two on hard surface and shallow snow	February breeding season often has two animals traveling closely together, appearing as a single animal and more galloping tracks are seen

Table 6. (continued).

Species	Relative track size	Usual track cluster	Primary distinguishing characteristics	Species most easily confused with	Remarks
Wolverine (*Gulo gulo*)	Large	Two by two (diagonal); three by three (diagonal); single, slightly offset track in deep snow	Usually shows three by three diagonal pattern at some time in the track segment; large feet, does not sink much unless snow is very feathery; in two by two feet are much larger than marten and fisher; usually solitary; drag marks are common and often curved; "pigeon-toed" appearance	Lynx; single wolf; fisher; otter (when walking)	When snow is very soft, walking track is common
Otter (*Lontra canadensis*)	Medium	Sliding trail; sometimes bounding; rarely walking	Sliding trail with parallel pairs of footprints; rarely walks; rarely lunges, except in very deep snow; usually around water but not uncommon going overland	Walking tracks of wolverine and otter are similar	
Fisher (*Martes pennanti*)	Medium-small	Two by two (diagonal, but feet usually more parallel than marten and wolverine)	Sinks deeply in deep snow; narrow trail; looks like very large marten or a very small wolverine in shallow snow on a hard surface or on very firm snow; generally only shows three by three diagonal pattern when snow is very firm	Wolverine, especially when snow is firm; marten; red fox when in two by two	
Marten (*Martes americana*)	Small-medium	Two by two (diagonal); sometimes walking (offset single)	Floats on the snow, almost always two by two	Red fox two by two in very shallow snow on hard surfaces; fisher	Most common track seen, except when snowshoe hares are abundant; very useful track for helping observers calibrate sizes of animal tracks
Mink (*Mustela vison*)	Small	Two by two (almost parallel)	Sinks deeply because of small feet; usually leaves a small trench; almost always near beaver dams, beaver lodges, lakeshores, and rivers; rarely travels overland; occasionally slides like otter, but is much smaller	Otter; red fox	
Snowshoe hare (*Lepus americanus*)	Small	Four by four with large hind feet parallel and in front, smaller hind feet parallel in rear	Very distinctive, always four by four; often in trails with several other hares; in boreal forest usually in young jackpine or hardwood regeneration		
Grouse and ptarmigan	Small	Single footprint but footprints so close together they blend as a trail	Abrupt beginning and end where birds land or take off		
Arctic or tundra hare (*Lepus arcticus*)	Small-medium	Four by four with large hind feet parallel and in front, smaller hind feet parallel in rear	Much larger than snowshoe hare, often 1.2–1.8 m between track clusters	Red fox or arctic fox when running	

Table 6. (concluded).

Species	Relative track size	Usual track cluster	Primary distinguishing characteristics	Species most easily confused with	Remarks
Porcupine (*Erethizon dorsatum*)	Medium	Deep trench	Confined to small areas	Beaver	
Beaver (*Castor canadensis*)	Medium	Deep trench	Confined to small areas near water	Porcupine	

Distinguishing problem tracks from the air

Moose vs woodland caribou. Positive identification must be based on sightings of large, oval beds (moose), browsing around hardwoods (moose), cratering in pine and spruce forest floors (caribou), or roundish beds on lakes (caribou). Both moose and caribou sometimes feed on muskrat pushups or paw in slush. See Figure 15.

All photographs in Appendix 2 by Audrey Magoun

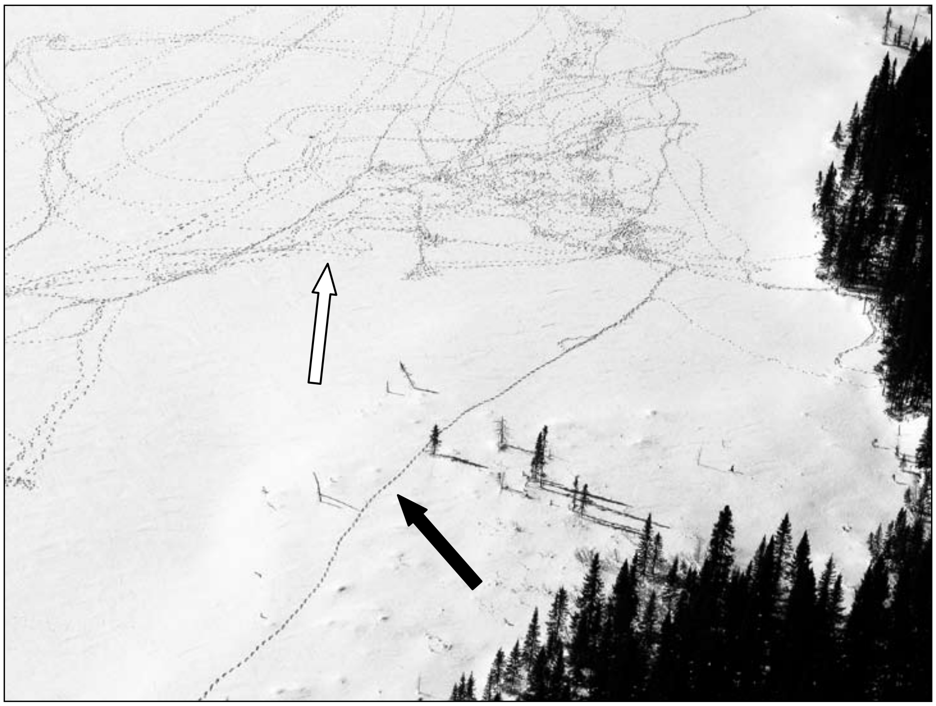

Figure 15. Wolverine (black arrow) and caribou (white arrow) tracks.

Moose vs bison. Positive identification should come from observing feeding patterns such as grazing (bison), or browsing (moose), or from herding nature (bison).

White-tailed deer vs wolf. Wolves tend to travel in straight lines in the open on rivers and lakes where pad prints can be seen on hard surfaces with shallow snow. Wolves will also urinate on stumps and objects in the snow. Deer meander along lakeshores, feed on cedar and hardwood shrubs, and seldom travel long distances in a straight line.

Lynx vs wolverine (walking) and single wolf. Lynx tracks are usually very clean and oval or egg-shaped, seldom with drag marks. Walking wolverines have extensive drag marks (often curved) on the outsides of the track, and their foot prints are closer together than lynx footprints. In February and March when lynx are breeding, tracks of a pair of lynx can look like those of a single wolverine (Figure 16). Single wolves sink more deeply than lynx, and wolf tracks are diamond shaped and often have some drag marks connecting the front and rear of the diamonds. Wolf tracks are also generally further apart than lynx tracks.

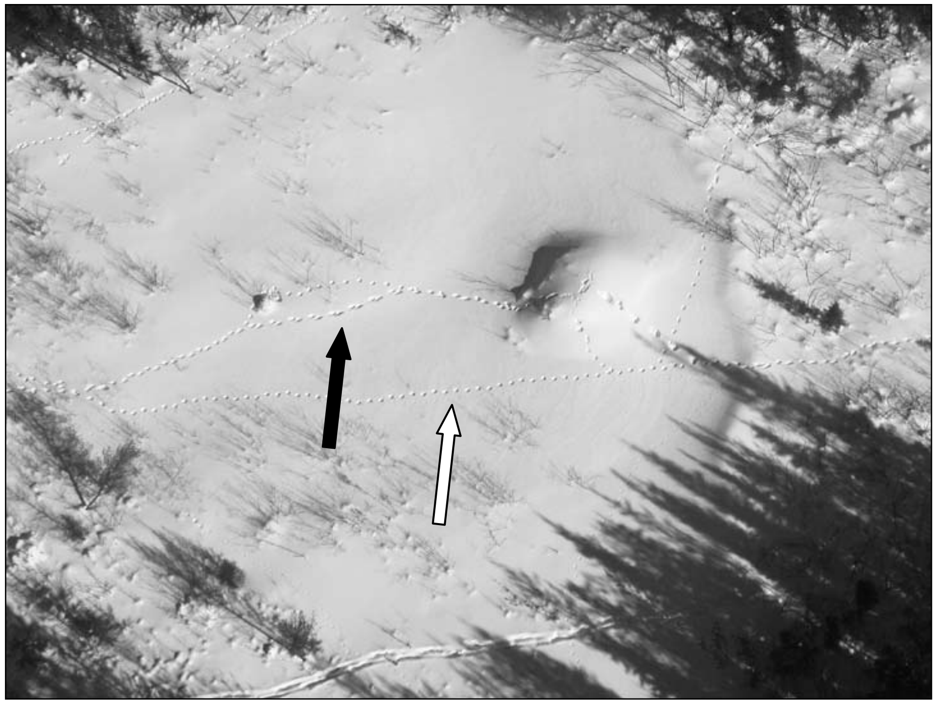

Figure 16. Wolverine (black arrow) and lynx (white arrow) tracks.

Coyote vs red fox and wolf. Coyotes often travel as pairs or singles and tend to be associated with snowshoe hares. Coyote and red fox tracks overlap in size and it is often not possible to distinguish the two when they are walking on hard surfaces with little snow. In deeper snow coyotes sink deeper than red foxes and red foxes wander to and fro rather than traveling in straighter lines. Single wolf tracks are much larger than coyote tracks and confusion usually occurs when observers lose their size perspective because of inadvertent changes in altitude of the aircraft. See Figure 17.

Figure 17. Wolverine (black arrow) and red fox (white arrow) tracks.

Red fox vs lynx, marten, and fisher. Red fox have smaller feet than lynx and steps are closer together. Red fox are only confused with marten (Figure 18) when the two are walking on hard surfaces with little snow and the trotting fox forms a two by two offset pattern. It is sometimes necessary to follow the track to see if the pattern changes. In February and March when foxes are breeding, pairs often follow in each other's footprints and these tracks can resemble those of the fisher. Tracks have to be followed to find where the foxes separate.

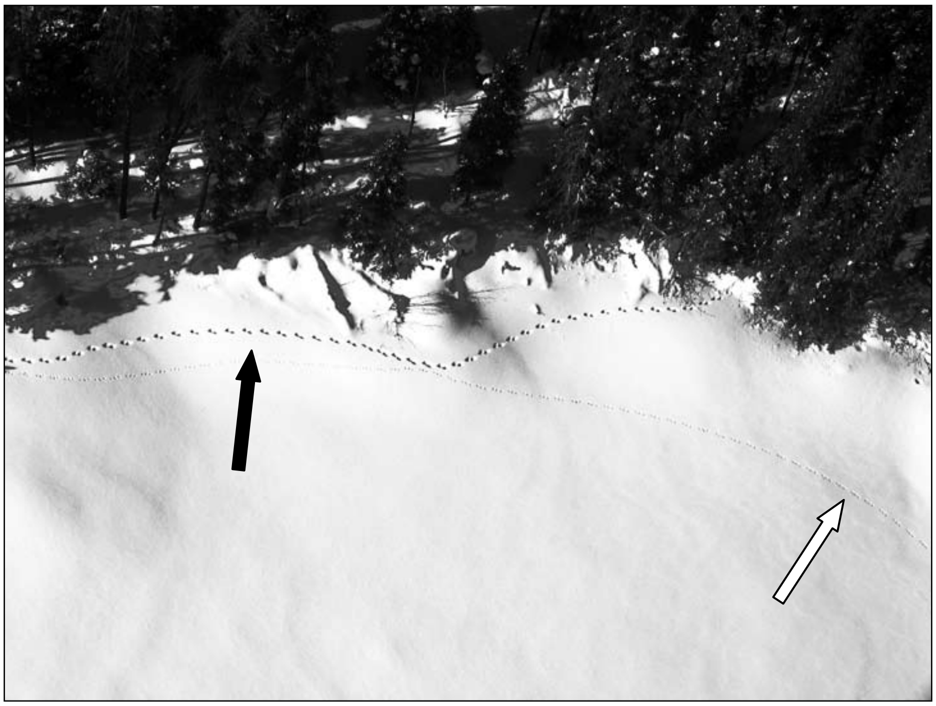

Figure 18. Wolverine (black arrow) and marten (white arrow) tracks.

Wolverine (walking) vs single wolf, and otter (walking). Wolverines will walk extensively if snow is very soft or where they are investigating food or scents (Figure 19). They have very large feet for their body weight and thus do not sink much unless snow is new and very fluffy (Figure 20 and Figure 21). They always leave drag marks (usually curved) but these can be obscured by wind or light snowfalls. Wolves sink deeply unless snow is very firm, and wolf tracks are much further apart than walking wolverine tracks. Otters very seldom walk far, but their walking tracks can look almost identical to walking wolverine tracks. Tracks have to be followed to see if the otter begins sliding (Figure 22).

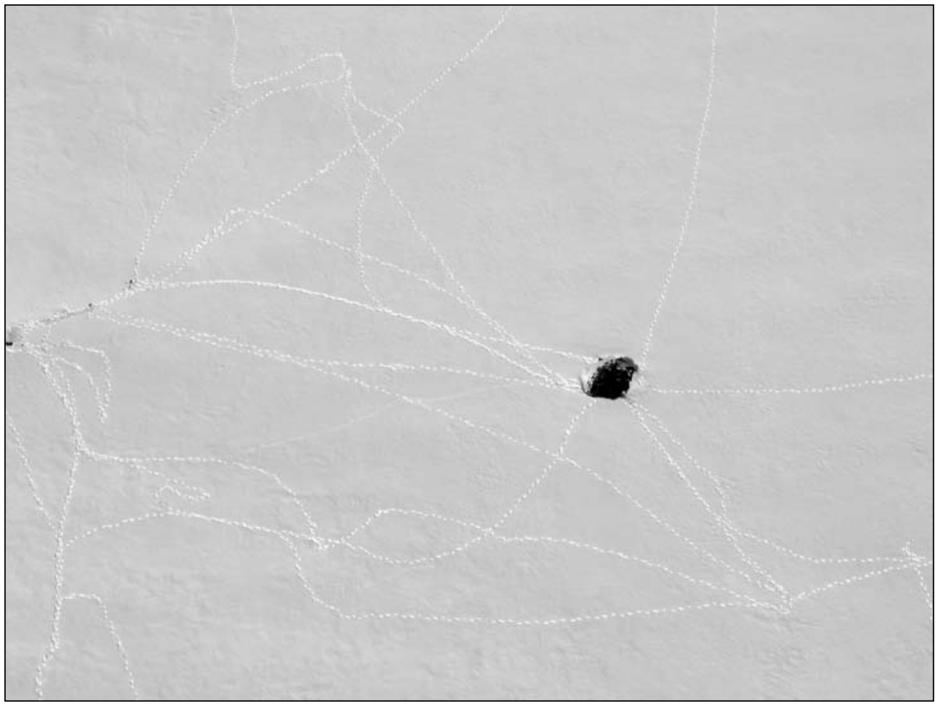

Figure 19. Wolverine tracks on a beaver house (note at least three types of tracks).

Figure 20. Tracks of a wolverine walking on crusty snow.

Figure 21. Typical wolverine two by two pattern in deep snow.

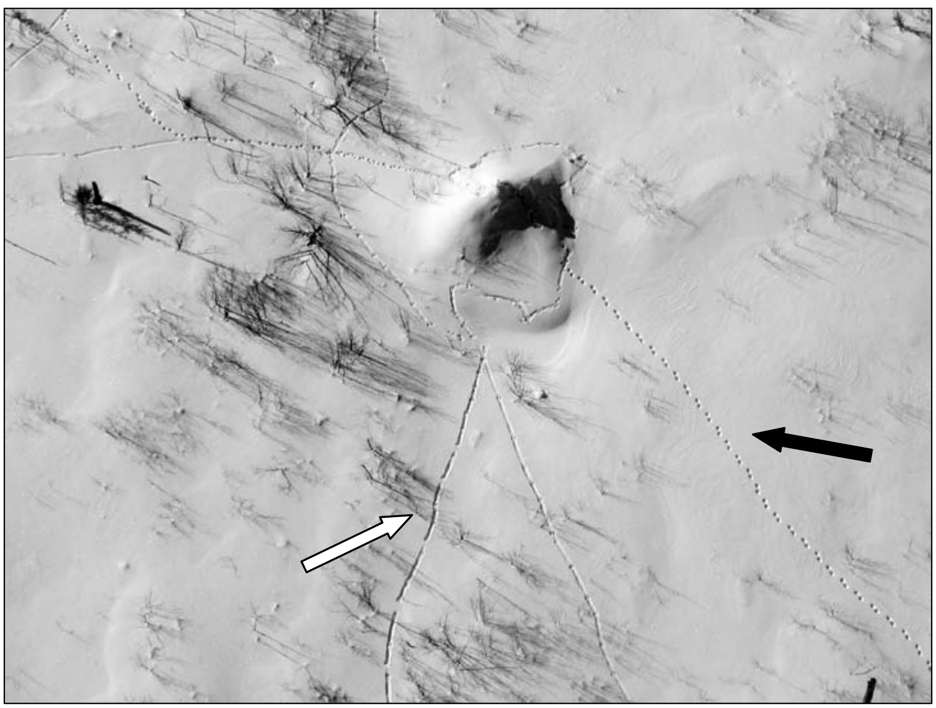

Figure 22. Tracks of a sliding river otter (white arrow) and a wolverine (black arrow).

Wolverine and fisher. In deep snow wolverines either walk (discussed above) or use the two by two diagonal (Figure 23) which can be confused with fisher. However, fishers sink deeper than wolverines, their tracks look narrow, and their bounds are further apart than those of wolverines. The wolverine has lynx-sized feet while the fisher has fox-sized feet. When snow is very dense the pattern of wolverine and fisher tracks can be virtually identical, the only difference being size. Both animals will use the three by three diagonal and two by two diagonal frequently (Figures 24 and 25). In cases like this it is often necessary to have a very low look and to have known-sized animal tracks (like marten) nearby for comparison. It is also useful to land and verify size a few times when these conditions are encountered. Once observers have landed several times, they will develop confidence in distinguishing between these animals in firm snow.

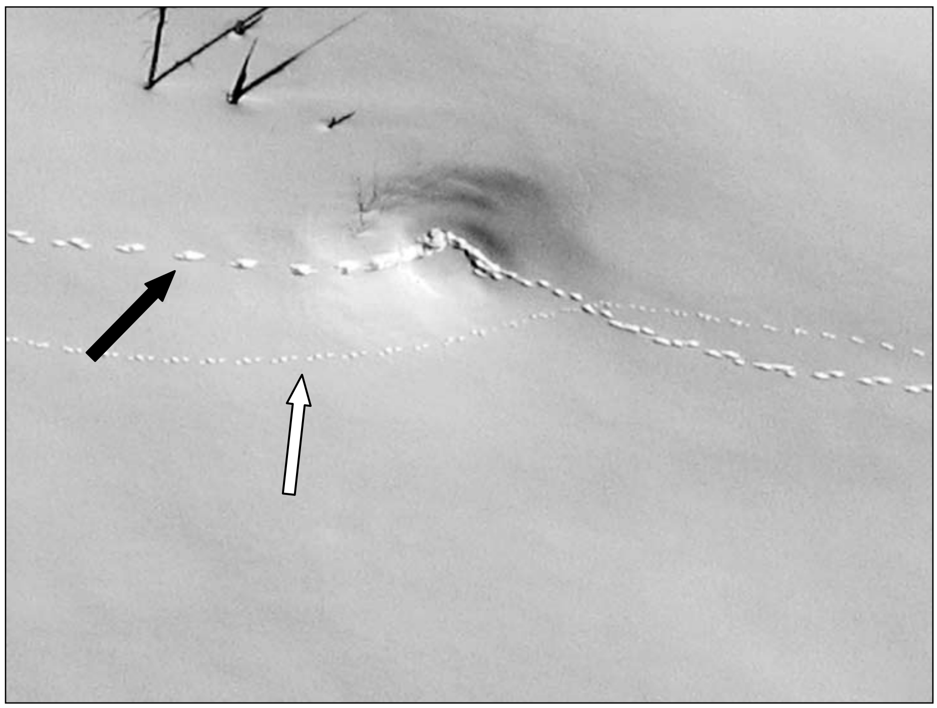

Figure 23. Tracks of a wolverine (black arrow) and fisher (white arrow).

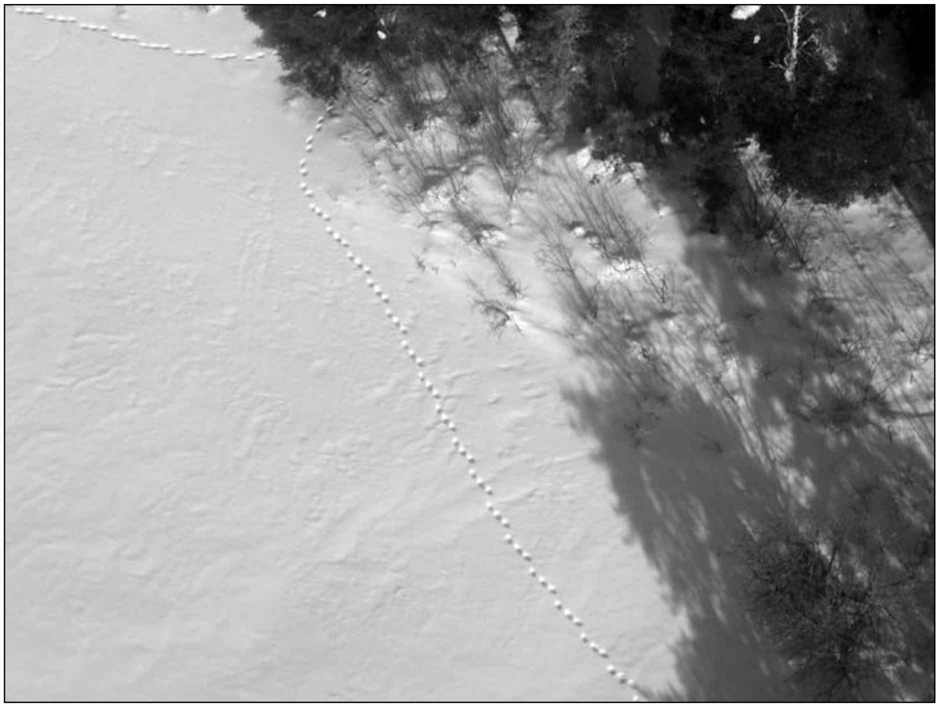

Figure 24. Wolverine tracks, showing three by three pattern.

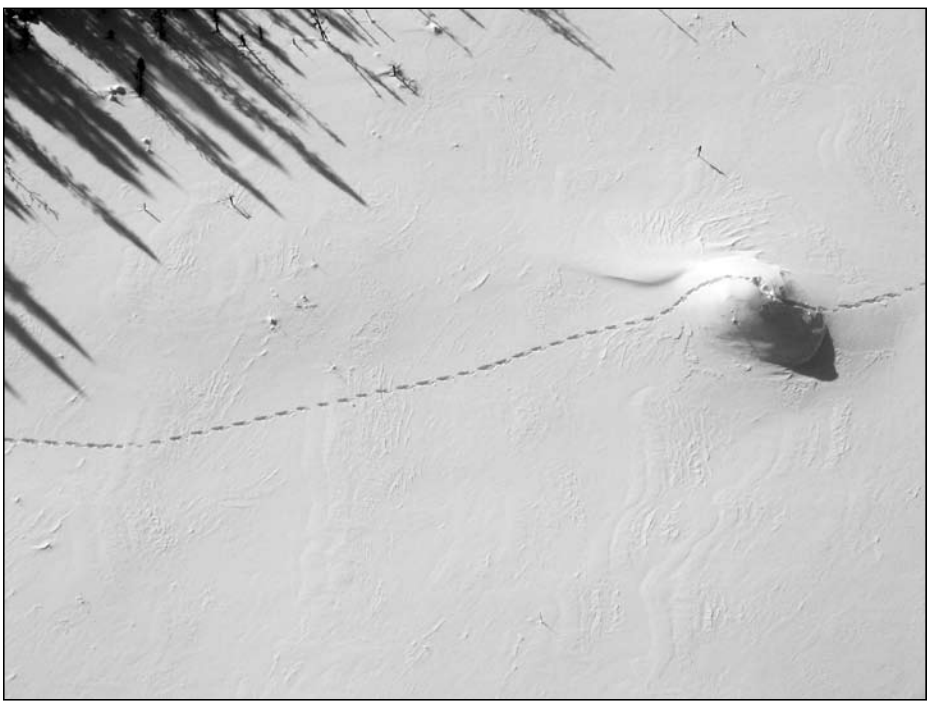

Figure 25. Typical wolverine two by two pattern in new, slightly windblown snow common in more open boreal forest.

Appendix 3. Analyzing spatial occurrence survey data with R and OpenBUGS.

In this appendix, we provide a "cookbook" description for analyzing spatial occurrence survey data using Markov chain Monte Carlo (MCMC) as described in Magoun *et al*. (2007a). MCMC simulation is accomplished with the freeware program OpenBUGS (www.mathstat.helsinki.fi/openbugs/). The freeware statistical package R (http://www.R-project.org) serves both as a front end to OpenBUGS, as well as a package for manipulating data to get it formatted for input into OpenBUGS.

To begin analysis, the raw data from the site surveys should be placed into three tables. The first is a table of hexagon IDs and dates of repeat surveys (Table 7). The second is a table of observed occurrences on each of the repeated surveys (see Table 7, with dates replaced by 1 or 0; whether or not occurrence was observed at the hexagon). Finally, Table 8 is an example of hexagon IDs, hexagon locations (site centroids), and any site-specific covariates that might be included in either the model for detection probabilities or occurrence probabilities. The procedures which are given here can be directly modified if there are time-varying covariates (e.g. weather conditions) for which it might be desirable to include in either model.

Now analysis can begin in R:
1. First open the file "data_manipulation.r" (Appendix 4) and paste all of the commands into a R session. This file constructs the necessary objects for an OpenBUGS session.
2. Open "analysis.r" (Appendix 5) and paste the commands into an R session to perform the MCMC analysis in OpenBUGS. The results are output to the file "spatialOccurenceResults.csv".
Appendix 6 holds some additional R code that is necessary for the computation of the spatial neighborhoods. These codes are also available in digital format, along with example data, at: http://people.trentu.ca/jebowman/.

Table 7. Dates of repeated surveys[1].

HEXID	DATE1	DATE2	DATE3	DATE4	DATE5
9	20050131	20050302	NA	NA	NA
10	20050115	20050207	NA	NA	NA
11	20050115	20050203	NA	NA	NA
12	20050120	NA	NA	NA	NA
13	20050120	20050129	NA	NA	NA
14	20050120	20050308	NA	NA	NA
15	20050129	20050302	NA	NA	NA
16	20050115	20050131	NA	NA	NA

[1] The dates can be represented in many different forms (this is just an example). How these dates will be used will ultimately determine the best form. NAs represent missing values (R uses NA). The number of columns should be equal to the maximum number of surveys for any sample unit.

Table 8. Site-specific information.

HEXID	CENTER_X[1]	CENTER_Y[1]	OPEN[2]
9	338312	12781985	0.5015
10	356925	12781985	0.3281
11	375537	12781985	0.5360
12	394149	12781985	0.7884
13	412761	12781985	0.6878
14	431373	12781985	0.5059
15	319700	12771239	0.7492
16	338312	12771239	0.5900

[1] UTM coordinates of hexagon centroids.
[2] An example of a covariate.

Appendix 4. R Code for data processing (data_manipulation.r).

```
# Data manipulation for input to OpenBUGS

# Load BRugs library
library(BRugs)

# Load function for CAR spatial models
source('carFunc.r')

# Load observation data
obsData = read.table('2005_intensive_wolverine_obs.csv', sep=',', head=T)

# Load dates for surveys
datesData = read.table('2005_intensive_wolverine_dates.csv', sep=',',
head=T)

# Load site specific data
siteData = read.table('2005_intensive_wolverine_sites.csv', sep=',', head=T)

## Calculations for spatial CAR specification ########################
#########

# Get UTM coordinates for sites and convert to km
UTM.km = siteData[,c('UTM_X', 'UTM_Y')]/1000

# Get distance to neighbor sites for CAR spatial model
thresh = ceiling(min(dist(UTM.km)))

# Get lists for OpenBUGS
nLst = ndef.disthresh(UTM.km, 'UTM_X', 'UTM_Y', thresh)
####################################

# Calculate date indicator for surveys conducted after Feb
dateInd = ((datesData[,-1]-20050214)>0)*1.0

# Insert dummy dateInd value for unsurveyed sites
dateInd[,1] = ifelse(is.na(dateInd[,1]),1,0)

# Calculate observed occurence
pres = ifelse(apply(obsData[,-1],1,sum,na.rm=T)>0,1,NA)
```

```
# Write data files for OpenBUGS
bugsData(
 list(
 Nsites = nrow(siteData),
 Nsurv = apply(!is.na(dateInd),1,sum,na.rm=T),
 y = as.matrix(obsData[,-1]),
 Dates = dateInd,
 Open = siteData[,'OPEN'],
 x = pres,
 adj = as.vector(nLst$adj),
 num = as.vector(nLst$num),
 C = as.vector(nLst$C),
 M = as.vector(nLst$M)
 ),
 fileName = 'spatialOccurenceData.txt'
)

# Initialization values for MCMC
bugsData(list(b0 = 0, b.date=0, b.open=0, b.pres=0,
eps=rep(0,nrow(siteData)),
 sig = 0, gamma=0.99), fileName='spatialOccurenceInits.txt')
```

Appendix 5. Analysis of occurrence model in OpenBUGS via R (analysis.r).

```
# Spatial Occurrence model for OpenBUGS

library(BRugs)

# Check for correct syntax
modelCheck('spatialOccurenceModel.txt')

# Load Data
modelData('spatialOccurenceData.txt')

# Compile MCMC code
modelCompile(numChains=1)

# Intialize sampler
modelInits('spatialOccurenceInits.txt')
modelGenInits()

# Begin updates for burnin
modelUpdate(10000)

# Set Parameters to monitor
samplesSet(
 c(
 'theta1',
 'theta0',
 'gamma',
 'b.date',
 'b0',
 'b.pres',
 'x',
 'sd.eps',
 'b.open'
 )
)

# Draw sample for inference
modelUpdate(50000)

# Calculate posterior summary
summTable = samplesStats("*")
write.table(summTable, "spatialOccurenceResults.csv", sep=",", quote=F)
```

Appendix 6. Additional spatial neighborhood function.

```
ndef.disthresh <- function(data, xcol, ycol, threshold)
{
	thresh <- threshold
	distmat <- as.matrix(dist(cbind(data[,xcol], data[,ycol])))
	dist.ind <- (distmat < thresh)*1.0
	diag(dist.ind) <- 0
	n <- length(distmat[,1])
	adj <- NULL
	C <- NULL
	num <- NULL
	M <- NULL
	for(i in 1:n) {
		neigh <- (1:n)*dist.ind[i,]
		neigh <- neigh[neigh > 0]
		neigh.C <- rep(1/length(neigh), length(neigh))
		adj <- rbind(adj,matrix(neigh, ncol = 1))
		C <- rbind(C, matrix(neigh.C, ncol=1))
		num <- rbind(num, length(neigh))
		M <- rbind(M, 1/length(neigh))
	}
	list(adj = adj, num = num, C=C, M=M)
}
```

Appendix 7. Power to detect changes in wolverine relative abundance (using number of tracks that cross a transect).

Bayne *et al*. (2005) found wolverine tracks in 0.1% of the triangles that they surveyed in Alberta, and estimated the average (between-sample unit) track count over a 72-hour sampling period to be 0.01 (SD = 0.10). Thus, the coefficient of variation (CV; standard deviation/mean) was 10.0. Bayne *et al*. (2005) found that between-sample unit CVs ranged between 1.1 and 11.8 for the 24 species that they detected, and within sample CV from a limited data set ranged from 0.5–1.7. Gibbs *et al*. (1998) concluded that temporal and sampling error variation (CV), estimated from 512 time-series data sets, ranged from 0.1 to 1.3. Thus, we used a range of CVs (between 0.5 and 10) for our power analysis. We assume that the CV of track counts in Ontario will fall in this range. Given a range of CV and number of sample units, we used program MONITOR (Gibbs *et al*. 1998; Table 1) to estimate the statistical power to detect differences in wolverine track count data over time, and G*Power (Faul *et al*. 2007; Table 1) to estimate the power to detect differences in the index between two areas (Figures 26 and 27).

Box 16. Monitoring number of tracks/triangle/24 hrs: Assumptions used in the power analysis.

Since we have not done a pilot study to evaluate the effects of varying effort on small-scale, ground-based snow track surveys to detect differences in wolverine relative abundance over time, we must make several assumptions, outlined below.

- The CV of number of wolverine tracks found on a 9-km transect in Ontario over 24 hours ranged from 0.5 to 10 (Schieck 2002, Bayne *et al*. 2005).
- We imputed a value of 10 tracks per transect into programs MONITOR and G*Power (therefore, when CV=2, standard deviation was 20). Note that the ratio of mean count to standard deviation is important, not the value of the count per se.
- We assumed triangles were surveyed once per year (except during the pilot study, where some hexagons need to be surveyed several times per year to estimate within-site variation).
- We assumed that the mean and standard deviation of track counts were the same for each hexagon. Therefore, the range of variances was modeled as within-sample unit variability; we did not model between-sample unit variability.
- The monitoring period was 10 years, with three complete surveys conducted once every five years.
- α=0.1 (we had a 90% chance of correctly detecting a change in the index).
- Tests were two-tailed.
- Results were based on exponential declines in track counts (Schieck 2002, Bayne *et al*. 2005).
- Results are the mean of 10,000 iterations (performed in program MONITOR).

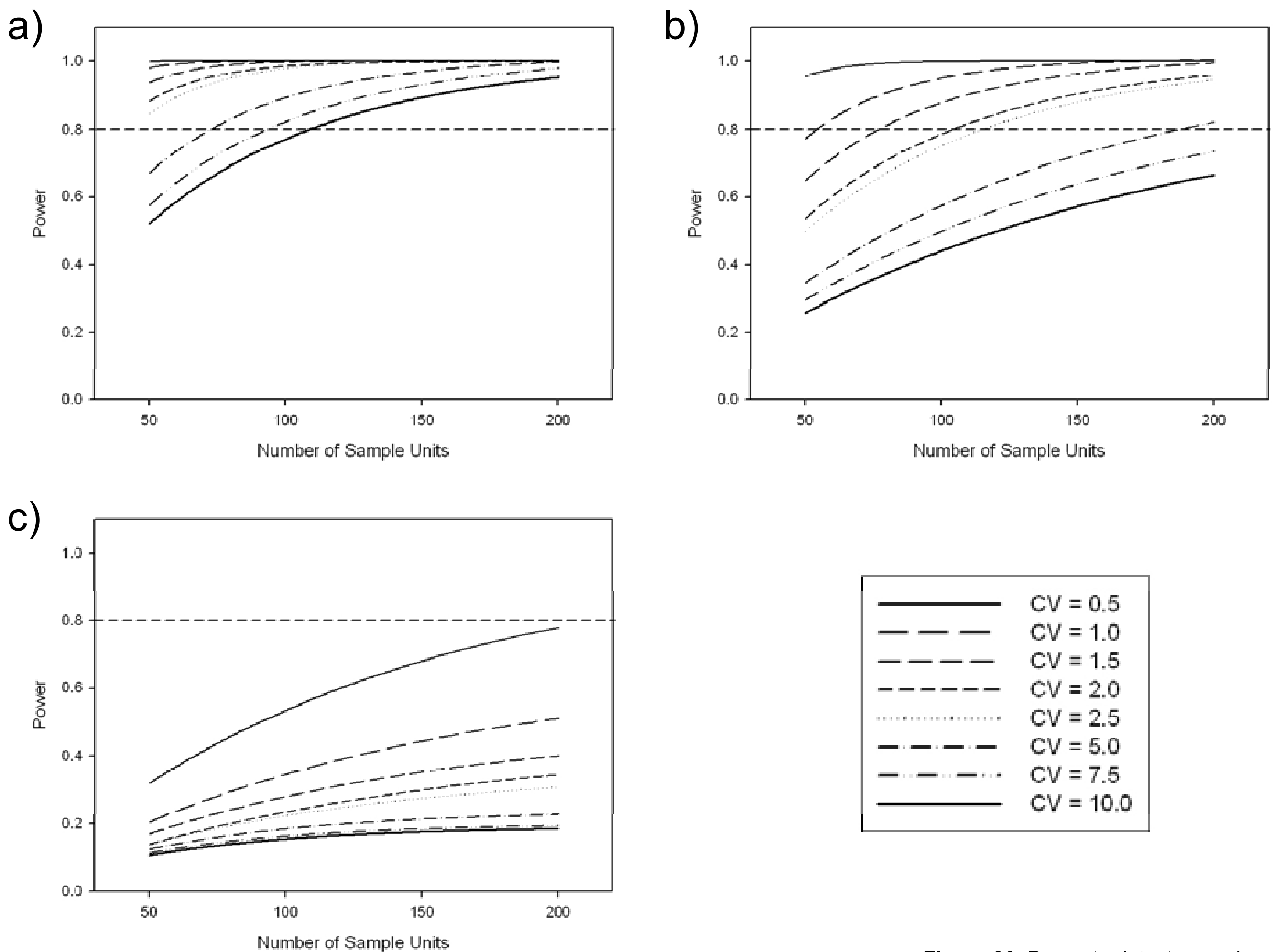

Figure 26. Power to detect annual exponential declines in track counts of a) 5%, b) 3%, and c) 1% over 10 years when 50–200 sample units per year were surveyed, for varying CVs. Additional assumptions are detailed in Box 16. Curves above the horizontal dashed line indicate circumstances in which power is >0.8.

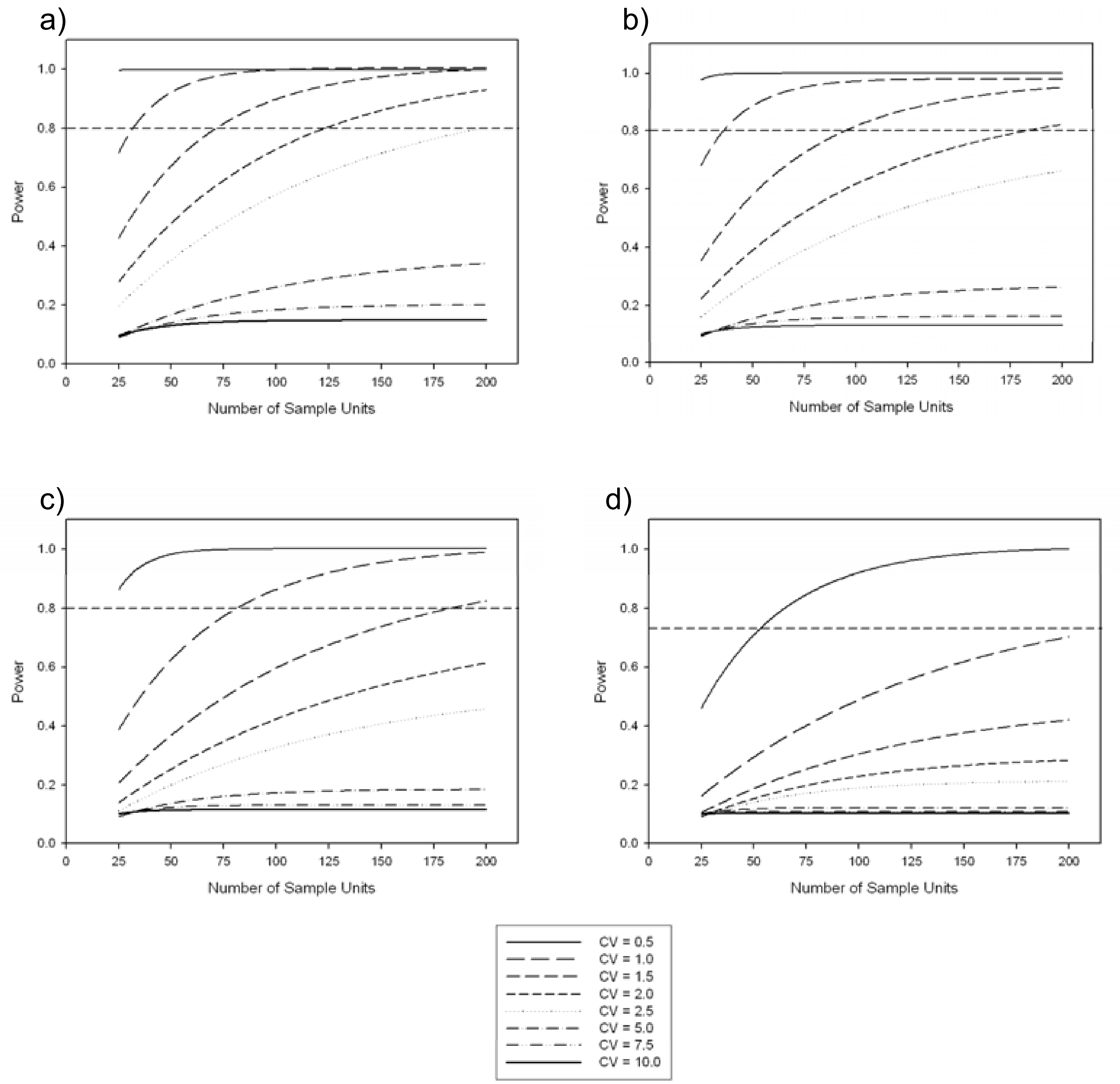

Figure 27. Effect of number of sample units surveyed per area on the power to detect differences in track counts of a) 100% (2-fold), b) 75%, c) 50%, and d) 25% between two areas. Curves above the horizontal dashed line indicate circumstances in which power is >0.8.

Appendix 8. Winter field safety considerations.

Winter is a time of extremes and it is important to be cognizant of safety issues and concerns during this season. It is the supervisor's responsibility to ensure that employees are adequately trained and aware of the potential hazards when working in winter conditions. The following are some of the more important items staff need to be aware of and all staff should be familiar with the complete health and safety indoctrination package for their office/work location.

All staff should have up-to-date Standard First Aid and CPR certification. Because equipment may break down or become bogged down more easily in the winter it is highly recommended that staff be trained in winter survival techniques in case they are required to spend an unscheduled evening in the bush. All staff should carry personal winter survival gear with them.

It is absolutely imperative that field crews have an emergency plan on file which outlines procedures for call-in at start of the work shift and call-in at the end of the shift to let office staff know when workers are leaving, where they will be working for the day, and when they have returned from the field. The plan must also outline the procedure for follow-up should the crew not call in at the end of the day so that search efforts can be mobilized in an effective and efficient manner.

Staff must have a satellite phone with them at all times and be properly trained in its use. A phone in the truck is of no use when your snow machine is broken down or you are injured on the far end of a transect.

Employees must be evaluated to ensure they are capable of driving in winter conditions and must be familiar with the "Winter Driving – Be Prepared, Be Safe" bulletin. If trailers are required for transporting snowmobiles then staff must be familiar with the OMNR "Trailer Operation and Maintenance Guideline" and "Introduction to Snowmobiling" bulletins. For a two-person crew, it is preferable that each crew member have a snowmobile for the following reasons: should one machine break down in a remote area then a machine is still available to get the crew back to their vehicle, and when breaking new trail it is much easier on the snowmobile if only a single person is on the machine to minimize bogging down and wear and tear on the machine.

Staff who will be working on or near ice must be familiar with the OMNR "Working On Ice Policy" and have completed ice safety training.

For aerial surveys, staff must be familiar with the Aviation services "Aircraft Safety and Winter Flying Requirements" bulletin regarding appropriate clothing and personal safety/survival gear.

Frost-bite, hypothermia, snow blindness, and sunburn are also potential winter hazards and staff should be aware of requirements and recommendations in the safety bulletins "Preventing Cold Injuries" and "Personal Protective equipment – Skin and Eye UV Radiation Protection Guideline".

When conducting hair snare surveys, employees will be in contact with baits in the form of road-killed deer or moose and trapper donated beaver carcasses. It is important to avoid, or at least minimize, bare-handed or direct contact with the animal and to wash hands thoroughly. Employees should be familiar with the "Handling Dead Animals – Safe Operating Procedures" bulletin.

Winter survival gear/kits should include the following items:

Ice picks	First aid kit (including pain relievers)
Windproof/waterproof matches/lighter	Personal flotation device (PFD)
Hatchet or small saw	Water, filter, and/or purification tablets
Polypropylene rope	High energy food
Flashlight	Sunglasses
Sound signaling device	Extra batteries
Flares	Spare GPS
Maps and compass	Watertight bag
Extra clothing	Survival knife
Candles	Duct tape
Sleeping bag	Survival blankets
Coffee can (for storage and boiling water)	Special medicine you may require

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