

## Comparative woodland caribou population surveys in Slate Islands Provincial Park, Ontario

Natasha L. Carr<sup>1</sup>, Arthur R. Rodgers<sup>2</sup>, Steven R. Kingston<sup>1</sup>, Peter N. Hettinga<sup>3</sup>, Laura M. Thompson<sup>4</sup>, Jennifer L. Renton<sup>5</sup>, & Paul J. Wilson<sup>4</sup>

<sup>1</sup> Ontario Ministry of Natural Resources, Ontario Parks, Thunder Bay, ON P7E 6S8, Canada (natasha.carr@ontario.ca).

<sup>2</sup> Ontario Ministry of Natural Resources, Centre for Northern Forest Ecosystem Research, Thunder Bay, ON P7B 5E1, Canada.

<sup>3</sup> Natural Resources Institute, University of Manitoba, Winnipeg, MB, R3T 2N2, Canada,

<sup>4</sup> Trent University, Natural Resources DNA Profiling and Forensic Centre, Peterborough, ON K9J 7B8, Canada.

<sup>5</sup> Centre for Forest Interdisciplinary Research, University of Winnipeg, Winnipeg, MB R3B 2E9, Canada.

*Abstract:* We evaluated three methods of estimating population size of woodland caribou (boreal ecotype) on the Slate Islands in northern Ontario. Located on the north shore of Lake Superior, the Slate Islands provide a protected and closed population with very limited predator influence that is ideal for a comparison of survey methods. Our objective was to determine the costs and benefits of three population estimation techniques: (1) forward looking infrared (FLIR) technology to count the number of caribou on regular-spaced transects flown by fixed-wing aircraft; (2) observers to count the number of caribou seen or heard while walking random transects in the spring; and, (3) mark-recapture sampling of caribou pellets using DNA analysis. FLIR and the genetics 3-window approach gave much tighter confidence intervals but similar population estimates were found from all three techniques based on their overlapping confidence intervals. There are various costs and benefits to each technique that are discussed further. Understanding the costs and benefits of different population estimation techniques is necessary to develop cost-effective programs for inventorying and monitoring this threatened species not only on the Slate Islands but for other populations as well.

**Key words:** forest-dwelling woodland caribou; population size; genetic profiling; forward looking infrared; FLIR; mark-recapture; transects; protected areas; *Rangifer tarandus caribou*; Slate Islands Provincial Park.

**Rangifer**, Special Issue No. 20: 205–217

### Introduction

The forest-dwelling ecotype of woodland caribou (*Rangifer tarandus caribou*) is listed as a threatened species in Canada. Population size estimates are a basic parameter used to assess and monitor a variety of caribou related programmes (e.g., evaluate the status of woodland caribou, track temporal population changes, assess the effectiveness of various management actions to maintain and/or restore populations). However, accurate and precise population estimates

have been notoriously difficult for woodland caribou (Thomas, 1998; Courtois *et al.*, 2003) due to very low densities and small groups dispersed over large areas.

A variety of survey methods are available to estimate population size and trend data for ungulates (Leopold, 1933; Caughley, 1977; Davis & Winstead, 1980; Seber, 1982; Sinclair & Caughley, 1994) but there are a number of problems with their application (Caughley, 1977; Seber, 1982; Sinclair & Caughley, 1994; Vincent *et al.*, 1996). Many of these

techniques, particularly aerial surveys, are hampered by the size of areas to be surveyed and difficulties in observing animals due to dense vegetation, as well as logistics and costs (McDonald 2004; Pollock *et al.* 2004). These problems are exacerbated for species such as caribou that are sometimes sparsely distributed and difficult to detect. Recently, non-invasive sampling methods such as genetic analyses of faecal or hair samples and thermal infrared imaging in aerial surveys, have increased in popularity and use for estimating abundance of rare or elusive species (Thompson, 2004).

Slate Islands Provincial Park provides an ideal setting to compare various population size estimation techniques for caribou as this archipelago represents an essentially closed population, with minimal immigration and emigration for the past 75 years and little influence of predation (Bergerud, 2001; Bergerud *et al.* 2007). The objective of this study was to evaluate three different population estimation techniques (four different methods) to assess the caribou population on the Slate Islands and discuss the pros and cons of each.

## Study area

### *Slate Islands Provincial Park*

Slate Islands Provincial Park, which is approximately 224 km east of Thunder Bay, came under regulation as a natural environment class provincial park in 1985. The total size of the protected area is 47.3 km<sup>2</sup> (OMNR, 1986). The park is comprised of two proximate groups of islands situated roughly 13 km southeast of the coastal mainland town of Terrace Bay (Fig. 1). The relatively small Leadman Islands group (which includes Leadman, Cape, Spar and Fish Island) is located approximately 2 km northeast of Patterson Island, which, along with Mortimer, McColl, Edmonds, Bowes, Delaute and Dupuis Island, constitutes the major grouping of islands included within park boundaries (Fig. 2). The total area of these islands, which were surveyed or sampled, was 37.2 km<sup>2</sup>.

The Slate Islands fall within the southern range limits of Ontario's boreal region and consequently they contain floral species and communities that are generally characteristic of the province's southern boreal, including balsam fir (*Abies balsamea*), black spruce (*Picea mariana*), white spruce (*Picea glauca*), white birch (*Betula papyrifera*), trembling aspen (*Populus tremuloides*) and Showy mountain-ash (*Sorbus decora*) (McGregor, 1974). The last major wildfire on the islands was believed to have occurred around the beginning of the 20<sup>th</sup> century (Cringan, 1956). Two logging operations

are thought to have taken place on the islands during the late 19<sup>th</sup> century (Cringan, 1956), while further logging activities were carried out during the 1930s (Cringan, 1956; Euler *et al.*, 1976). Lacking substantial wildfire or recent logging disturbance, natural succession processes are leading to a reduction of deciduous forest cover on the Slate Islands (W.J. Dalton pers. comm., 2002). Based on long-term observations and the preliminary results from exclosures, Bergerud (2001; Bergerud *et al.* 2007) suggested that several plant species are under threat of being extirpated from the islands as a result of intensive browsing and foraging pressure by caribou.

The first definitive evidence of woodland caribou on the Slate Islands dates back to the winter of 1907, when tracks (crossing both to and from the mainland) were noted along the surface of the ice that had formed between the islands and the mainland (Middleton, 1960 cited in McGregor, 1974). Bergerud (2001) has suggested that from 1907 to the mid 1930s, the caribou population was relatively small, with movements of individuals across the 13 km between the islands and the mainland during the occasional winters when an ice bridge formed between them. No definitive evidence for the consistent year-round presence of caribou on the islands existed prior to the 1940s (Bergerud, 2001). Bergerud (2001) has argued that as a result of the end of selection logging activities on the islands in approximately 1935, combined with a possible increase in predation pressure on the mainland, movements of caribou both to and from the islands ceased and the Slate Islands population became relatively isolated. The last recorded solid ice that occurred between the mainland and the Slate Islands was in the winter of 1993-1994 (Bergerud, 2001; Bergerud *et al.* 2007). Movements of caribou to the mainland were not recorded during that winter but two wolves crossed the ice to the Slate Islands and substantially reduced calf survival and overall population numbers until 1996, after which the wolves were no longer observed (Bergerud *et al.*, 2007). Wolf sign was again observed on the Slate Islands in 2003 and 2004 (Bergerud *et al.*, 2007).

Caribou population surveys on the Slaters Islands were completed every year from 1974-2003 using the "King census" strip transect technique (King, 1937) and from 1975-1997 using a mark-recapture Lincoln Index (Lincoln, 1930). During this period, Bergerud (2001; Bergerud *et al.* 2007) suggests that the population began to increase and eventually entered a "boom and bust" cycle that he believes has persisted to the present day, whereby the number of individuals has fluctuated between 100 and 600 animals and major "die-offs" are experienced at five year intervals.



Fig. 1. Regional Context for Slate Islands Provincial Park.

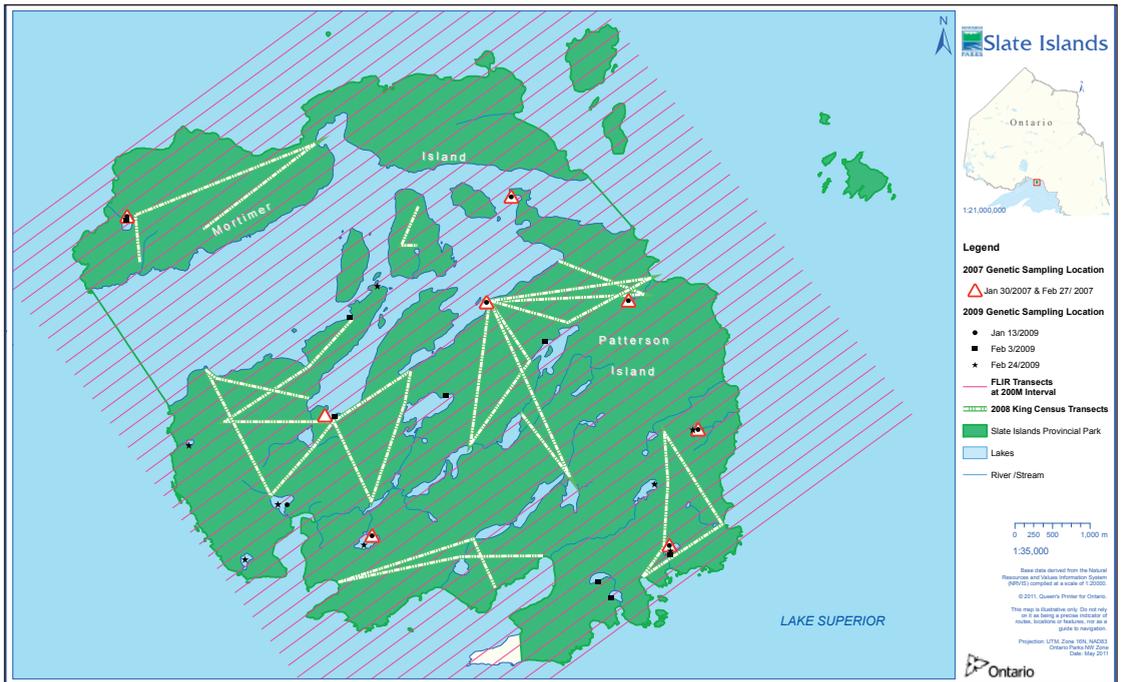


Fig. 2. Locations of faecal pellet collections, ground and FLIR transects surveyed to obtain population estimates of caribou on the Slate Islands, Ontario.

## Methods

### *Distance Sampling Techniques*

#### Forward Looking Infrared (FLIR)

The FLIR surveys of the Slate Islands were conducted by Vision Air Research Inc. (Boise, Idaho) on January 29-30, 2009. They used a PolyTech Kelvin 350 II gimbal (Eskilstuna, Sweden), which included a high resolution Agema Thermovision 1000 (FLIR Systems, Inc., Wilsonville, Oregon) infrared sensor with a spectral range of 8-12 microns and a Sony video camera (Sony Corporation, Minato, Tokyo, Japan), mounted under the left wing of a Cessna 206 "Stationair". The thermal delta of the infrared sensor was less than 1 °C, so it could detect objects with less than 1 °C temperature difference from the background. The sensor gimbal allowed 330° of azimuth and 90° of elevation providing complete coverage except directly behind the airplane. The FLIR system had both a wide (20°) and narrow (5°) field of view (FOV). At 305 m above ground level looking straight down using the wide FOV, the footprint or area covered by the sensor was 110 m in width x 71 m in length, while the narrow FOV provided a footprint of 27 m x 18 m. The sensor operator / wildlife biologist sat in the rear seat of the aircraft and watched a high resolution 38 cm monitor to aim and focus the sensor, which had 800 x 400 pixels resolution. The operator identified animals by their morphology and luminous intensity (Fig. 3). The pilot had >1000 hours of experience flying FLIR surveys and the sensor operator had > 5000 hours experience with FLIR use and interpretation.

Survey flights took place between 1000 and 1400 hrs. Survey transects were oriented to run northeast – southwest to take advantage of the islands' terrain. Transects were spaced 200 m apart to give complete coverage of the area and some overlap to allow more viewing angles of cliffs and steep terrain. Transects were navigated using a Global Positioning System (GPS). For safety reasons, flight altitude was 305 m above ground level of the highest point along each transect flown and the adjacent transect. The sensor look angle was approximately 30° in elevation to nearly straight down. The sensor operator scanned side to side to allow multiple fields of view and additional overlap. Animals were initially sighted using the infrared sensor wide FOV then checked with the narrow FOV and verified using real time video imagery.

The portion of the flight within the study area was recorded on video. The pilot and sensor operator communicated to verify the start and end of each transect to turn the video recorder on and off. The video recorder had slow motion, still image display,



Fig. 3. FLIR images of (a) an adult moose near Marathon, Ontario, taken at an altitude of 610 m (2000 ft) a.g.l. and (b) an adult caribou on the west side of Patterson Island taken at an altitude of 305 m (1000 ft) a.g.l. Even at higher altitude, the moose is obviously much larger and has greater luminous intensity than the caribou. Images courtesy of Susan Bernatas, Vision Air Research, Boise, ID, USA.

and zoom modes. Caribou were located by observing their level of emitted infrared energy versus background levels (Fig. 3). Caribou were mapped at their observed position in relation to physical features (Gill *et al.*, 1997; Bontaites *et al.*, 2000) on an enlarged 1:50 000 topographic map (Energy, Mines and Resources Canada, Ottawa, Ontario, 1986) rather than the position of the airplane. Plotting individual caribou locations allowed identification and omission of duplicate sightings (Haroldson *et al.*, 2003).

#### Analyses

Following the survey, all video recordings were reviewed frame by frame, forward and backward and in slow motion to confirm caribou sightings and locations and to verify the number of individuals that

may have occurred in groups. An additional check of the data was performed by sampling the videotape for detection verification and checking for duplicate groups.

Perpendicular distances between caribou locations and transect lines were determined in ArcGIS 9.2 (ESRI, 2006). A caribou population estimate and associated confidence intervals from the FLIR survey were then calculated using Distance 6.0 release 2 (Thomas *et al.*, 2010). The population estimate, assuming 100% sightability along transect lines, was based on a half-normal detection model with simple polynomial adjustment that was chosen by minimisation of Akaike's information criterion (AIC<sub>c</sub>) from a variety of hazard rate and half-normal models examined (Buckland *et al.*, 2001). Although caribou could have occurred on ice-covered lakes, no animals were observed more than 100 m from land along any shoreline (i.e., half the distance between transect lines) so density estimates used only those portions of transect lines that occurred on land. The total length of the transect lines that occurred on land was 284.4 km.

#### *Walking transects*

The Slate Islands caribou population was estimated using the King census technique described by Bergerud *et al.* (2007). Single persons walked straight line transects by compass over a 30-day period in July 2008 (Fig. 2). Transects were walked on days with little or no wind and with damp ground litter, resulting in good listening conditions. Transects were walked at a normal walking pace, with frequent stops for compass bearings. Noise was kept to moderate levels to limit disturbing or alerting caribou. Transect routes were chosen to cover different habitat classes across the islands (mostly sparse/dense coniferous habitat, taking turning points (topographic features, lakes, bays, etc.) and boat pick-ups and drop offs into consideration. Routes were selected to avoid areas disturbed in recent days by previous transects. Observers estimated the distance to any caribou seen or heard. As indicated previously, no other large mammals are usually present on the Slate Islands other than when wolves are occasionally observed. All transect routes and caribou observations were recorded on a map of the park. The average length of the 11 transect lines that were walked was 4.2 km and the total length was 63.4 km.

#### *Analyses*

Caribou density and associated confidence intervals from the ground transect survey were calculated using Distance 6.0 release 2 (Thomas *et al.*, 2010).

The population estimate was based on a uniform detection model without adjustment that was chosen by minimisation of Akaike's information criterion (AIC<sub>c</sub>) from a variety of uniform models with different adjustment terms that were tried (Buckland *et al.*, 2001).

## **Mark – Recapture Technique**

#### *Genetics*

In 2007, faecal pellets were collected on January 30 and February 27 (2-window approach) for mark-recapture analysis. The eight sampling sites were chosen by randomly selecting lakes and/or sheltered bays within the study area that were appropriate for landing a helicopter to collect samples. At each site, four people searched for approximately 20-30 minutes in each of the 4 cardinal compass directions by searching lakes and shorelines. The same random sites were visited on January 30 and February 27. Each faecal sample was placed in a sealable plastic bag to prevent DNA contamination and stored at -20 °C. All samples were shipped frozen to the Natural Resources DNA Profiling and Forensic Centre at Trent University in Peterborough, Ontario for DNA analysis to identify unique individuals.

More sophisticated mark-recapture models, allowing for variation in capture probabilities, can be constructed when 3 or more sampling periods are assessed, so we also estimated caribou population size using a 3-window approach (Otis *et al.*, 1978). In 2009, faecal pellets were collected on January 13, February 3 and February 24 (3-window approach). Sampling sites were again randomly chosen, however, a different random set was chosen for each of the three sampling periods. As with the 2-window approach protocol, all samples were stored in a sealable plastic bag, frozen, and shipped for analysis.

#### *Laboratory analyses*

Caribou DNA was extracted from faecal samples using the methods of Ball *et al.* (2007). DNA was amplified using 9 polymorphic, microsatellite markers (Rt6, Rt7, Rt24, Rt30 (Wilson *et al.*, 1997); Map2C, BM848 (Moore *et al.*, 1992); BM888, RT5 (McLoughlin *et al.*, 2004); BMS1788 (Cronin *et al.*, 2005). Each reaction was composed of a 10- $\mu$ l volume containing: 1x PCR buffer, 2.0  $\mu$ M MgCl<sub>2</sub>, 0.2  $\mu$ g/ml of BSA, 0.4-0.5  $\mu$ M of each primer (forward primer fluorescently labelled with NED, FAM, or HEX; Applied Biosystems [ABI], Foster City, California, USA); 0.2  $\mu$ M of each dinucleotide triphosphate; 1 unit of Taq polymerase (Invitrogen Life Technologies, Carlsbad, California, USA) and 2.0  $\mu$ l of DNA

template. The amplification cycle consisted of an initial denaturing of 94 °C for 5 min followed by 30 cycles of 94 °C denaturing for 30 seconds, 56-60 °C annealing for 30 seconds, and 72 °C extension for 30 seconds. The cycling culminated with a final extension of 60 °C for 45 minutes. Thermal cycling was performed in an MJ DNA Engine PTC 200 (MJ Research, Watertown, Massachusetts, USA) configured with a heated lid.

Generally, 0.5 µl of each desalted sample was added to 10 µl of deionized formamide and 0.002 µl of the internal size standard GENESCAN-500 (ROX; ABI). That mixture was subjected to capillary electrophoresis on an ABI 3730 Genetic Analyzer (i.e., automated sequencer) and GENEMARKER AFLP/Genotyping Software (version 1.6; Soft Genetics LLC®, State College, Pennsylvania, USA) was used to score, bin, and output allelic (and genotypic) designations for each caribou sample.

#### Statistical analyses

We compared genotypes at each of the 9 microsatellite loci to identify the number of unique individuals sampled. We calculated the probability that 2 or more individuals within the population shared the same genotype using the probability of identity for siblings calculations ( $PI_{sib}$ ; Evett & Weir, 1998) where caribou genotypes were accepted as unique individuals when  $P \leq 0.05$ . All calculations were performed in program GENECAP (Wilberg & Dreher, 2004). Information on matching genotypes based on sampling time for 2007 and 2009 was also retained for use in applying mark-recapture models.

Population closure is defined as a population size that remains constant over the period of investigation; that is, where no recruitment (births or immigration) and no losses (death or emigration) occur. Because immigration and emigration of woodland caribou to/from the Slate Islands were unlikely and caribou faecal pellets were collected over relatively short time periods (winter months prior to calving; Pollock *et al.*, 1990), we only considered closed models for population size estimation. Those included the modified Lincoln-Petersen estimator (2-window approach; Seber, 1982) and the multiple mark-recapture models (3-window approach; Otis *et al.*, 1978). Based on guidelines given by Otis *et al.* (1978) and White *et al.* (1982), estimates for all models were produced with the objective to obtain a coefficient of variation (CV) of  $\leq 20\%$  and capture probabilities  $\geq 20\%$ . The examination of woodland caribou population parameters in the application of genetically-based mark-recapture estimates has been applied in other caribou populations (Hettinga, 2010).

The modified Lincoln-Petersen model (Chapman, 1951) was used to estimate caribou abundance based on individual genotypes collected from 2 sampling occasions in 2007. That estimator is based on the ratio of marked and unmarked individuals captured within 2 sampling periods (i.e., 2-window approach; Seber, 1982) and relies on the following assumptions: the population is closed to additions (births or immigrants) and deletions (deaths or emigrants), all animals are equally likely to be captured in each sample, and marks are not lost and are not overlooked by the observer (Pollock *et al.*, 1990). Ninety-five percent confidence intervals for calculated Lincoln-Petersen estimates were estimated using the inverse cube root method (Arnason *et al.*, 1991).

It is widely recognized that the assumption of equal catchability is not met in most mark-recapture studies conducted on natural populations (White *et al.*, 1982). Consequently, the use of multiple mark-recaptures using the 3-window approach (i.e., individual genotypes sampled in 2009) allowed the application of multiple models to assess sampling covariates in the estimation of population size (Otis *et al.*, 1978; White *et al.*, 1982). Following closed population modeling assumptions in acquiring mark-recapture data over multiple sampling intervals, animal capture histories can be used to model variability in estimated capture probability rates and increase the precision and accuracy of calculated estimates (Otis *et al.*, 1978). Models often used in examining variation in capture probability include those assessing time effects, behavioural capture effects, individual heterogeneity or interactions between any and all sampling factors present. The utility of using alternate models to assess variation in capture probability based on sampling covariates is limited by the quality of data available, where increasing sampling times and recapture rates can be important in increasing estimator accuracy and precision (White *et al.*, 1982).

Models run in the interpretation of capture history information from the 3-window approach included the Mo, null model, Mt, time effects models, and Mh, the heterogeneity jackknife model. The Mh estimator is a model derived to look at individual differences in capture probability and has relatively widespread use (Chao & Huggins, 2005). The Mh model is ideal with non-invasive genetic sampling where variability in sampling frequency for identified individuals is often apparent (Mills *et al.*, 2000; Frantz *et al.*, 2003; Hansen *et al.*, 2008) and has been used previously in the estimation of population size for woodland caribou populations (Hettinga, 2010).

Estimation of the Mo, Mt and Mh model was done using the CAPTURE (White *et al.*, 1982) application

within program MARK (White & Burnham, 1999). Model ranking was done in CAPTURE where likelihood ratio tests were used to determine if models used could serve as accurate indicators for calculated capture probability values when compared to the null, Mo model, or other imbedded models (Otis *et al.*, 1978). Where a model ranking was given to a model that was unusable (due to the limited number of sampling times or sparseness in sampling data) the next highest ranking model was selected as the candidate model for use in estimating population size.

## Results

### *Forward looking infrared (FLIR)*

The FLIR survey of the Slate Islands was completed in 5.3 hours of flying over two days. Follow-up tape review and analysis took 12 hours. The FLIR survey recorded 58 caribou at 46 locations on the Slate Islands; two groups of three individuals, eight groups of two and the remainder were singles. Individuals were not classified by age or sex but most groups of two were cow-calf pairs. The estimated density was  $1.56 \pm 0.50$  caribou/km<sup>2</sup> with a CV of 19.4%, producing a population estimate of 58 caribou (95% CI 40-85) (Table 1).

The costs of the FLIR survey included the actual flight time of 5.3 hrs over two days at CDN\$ 750 per hr, 12 hrs of videotape review and analysis at CDN\$ 100 per hr, and daily crew support (i.e., food, accommodations, etc.) of CDN\$ 310 per day. So, the total cost of the FLIR survey of the Slate Islands was about CDN\$ 5800. The ferry costs of bringing the crew and their aircraft to the survey location (CDN\$ 370 per hr) have not been included (Table 2).

### *Walking transects*

A total of 11 caribou were observed on transects that were ground surveyed on the Slate Islands. No groups were observed and individuals were not classified by age or sex. The density estimate calculated from the ground survey data was  $3.62 \pm 0.17$  caribou/km<sup>2</sup> with a CV of 29.3%, producing a population estimate of 134 caribou (95% CI 71-255) (Table 1).

The costs of the ground transect survey were minimal and, excluding wages, only included the costs of transportation by boat to the Slate Islands and provisions (i.e., food, camping equipment) for the field crew. We estimated the total cost of the ground transect survey to be < CDN\$1000 (Table 2).

### *Genetics*

One hundred faecal samples were analyzed from the 2007 field season and 49 unique individual genotypes were identified. The  $PI_{\text{ind}}$  calculated for individuals captured during 2007 (2-window approach) was  $4.32 \times 10^{-4}$ . That probability corresponded to a 1 in 2315 chance that 2 individuals had the same genotype at the loci examined. The Lincoln-Petersen model calculated for the 2-window approach in 2007 produced a population estimate of 151 caribou or 4.1 caribou/km<sup>2</sup> (Tables 1 and 4). However, the precision (CV = 37%) and the capture probability of that model was low (10%).

In 2009, 164 faecal samples were analyzed (based on 3 sampling occasions) and 57 unique individual genotypes were identified. The  $PI_{\text{ind}}$  was  $1.53 \times 10^{-3}$  for the 57 individuals captured during 2009, corresponding to a 1 in 654 chance of encountering identical genotypes. In the use of models examining sampling covariates where three sampling windows were considered, the Mo, Mt and Mh models, alternate population estimates were calculated (Tables 1, 3 and 4). The Mh, heterogeneity jackknife estimator,

Table 1. Estimates of woodland caribou population size on the Slate Islands, Ontario, using three different techniques.

	FLIR	Walking Transects	Genetics (2 sampling periods)	Genetics (3 sampling periods)
Population Estimate	58 (program DISTANCE)	134 (program DISTANCE)	151 (Lincoln- Petersen)	99 (program CAPTURE)
Variability around N (95% Confidence Interval)	40-85 (Thomas <i>et al.</i> , 2010)	71-255 (Thomas <i>et al.</i> , 2010)	80-349 (Arnason <i>et al.</i> , 1991)	85-122 (White <i>et al.</i> , 1982, Otis <i>et al.</i> , 1978)
Confidence in population estimate	High due to narrow confidence intervals	Low due to large confidence intervals	Low due to large confidence intervals	High due to narrow confidence intervals

Table 2. Comparison of potential advantages, disadvantages and costs of three different techniques used to estimate woodland caribou population size on the Slate Islands, Ontario.

	FLIR	Walking Transects	Genetics (2 sampling periods)	Genetics (3 sampling periods)
Correction factor	Possible – marked individuals would provide an estimate of detectability	Possible – marked individuals would provide an estimate of detectability	Inherent in calculation	Inherent in calculation
Male to female ratio	Possible, if image permits	No, few observed individuals can be sexed	Yes, high confidence	Yes, high confidence
Calf ratios	Possible, if image permits and animals are flushed	Unlikely due to few calves observed	Possible, based on size of pellets	Possible, based on size of pellets
Location accuracy	High	Low	High	High
Group sizes estimate	Yes, with high confidence	Yes, with low confidence due to disturbance of individuals by observer	Depends on sampling	Depends on sampling
Time restrictions	Preferably not during leaf out period	Preferably not during leaf out period	Winter	Winter
Additional values of sampling	Census other species		<ul style="list-style-type: none"> <li>· Contribution to metapopulation research</li> <li>· Collected pellets used for other testing (e.g., pregnancy, diet)</li> </ul>	<ul style="list-style-type: none"> <li>· Contribution to metapopulation research</li> <li>· Collected pellets used for other testing (e.g., pregnancy, diet)</li> </ul>
Costs	<ul style="list-style-type: none"> <li>· 5.3 hrs flying over 2 days@750hr = \$3975</li> <li>· 12 hrs. videotape review and analysis @100hr = \$1200</li> <li>· Daily support (food and accommodations) = \$620</li> <li>· Total \$5.8K</li> </ul>	<ul style="list-style-type: none"> <li>· Time of 2 people for approximately 1-2 weeks)</li> <li>· Total &lt;\$1K (boat ferry from Terrace Bay plus food)</li> </ul>	<ul style="list-style-type: none"> <li>· 5 hrs flying @1200hr = \$6K</li> <li>· DNA analysis for 100 samples @ \$30 sample = \$3K</li> <li>· 4 people walking transects for 2 days</li> <li>· Total \$9.0K</li> </ul>	<ul style="list-style-type: none"> <li>· 7.5 hrs flying @1200hr = \$9K</li> <li>· DNA analysis for 164 samples @ \$30 sample = \$4920</li> <li>· 4 people walking lines for 3 days</li> <li>· Total \$13.9K</li> </ul>

was selected as the best fit model for use in estimating population size and an estimate of 99 animals (95% CI 85-122), or 2.7 caribou/km<sup>2</sup> was calculated. Alternately use of the Mo and Mt models yielded

estimates of 115 (95% CI 83-185) and 100 (95% CI 75-157) animals, respectively. Those estimates corresponded to densities of 3.1 and 2.7 caribou/km<sup>2</sup>, respectively. Calculated coefficient of variation values

Table 3. Estimation of population size,  $n$ , based on genetic sampling using program CAPTURE (White *et al.*, 1982) with 2009 sampling information from Slate Islands Provincial Park woodland caribou population.

	All animals			Males			Females		
	$n$	SE	95% CI	$n$	SE	95% CI	$n$	SE	95% CI
Mo	115	23.32	(83,185)	42	8.76	(31,71)	68*	26.91	(38,191)
Mt	100	18.35	(75,157)	38	6.81	(29,63)	58	20.69	(34,158)
Mh	99*	9.41	(85,122)	44*	6.09	(36,60)	47	6.59	(38,64)

\* selected as best fit model using Likelihood Ratio Tests in program CAPTURE (White *et al.*, 1982).

Table 4. Sampling information in the collection of woodland caribou faecal pellet samples from the Slate Islands Provincial Park in 2007 and 2009.

	2007		2009		
	Jan 30	Feb 27	Jan 13	Feb 3	Feb 24
Sites Sampled	8	8	8	8	8
Samples Collected	37	63	34	46	84
Genotypes ID'd	18	31	14	16	41
Capture Probability	10%	17%	14%	16%	41%

\* Capture probability calculated using time effects (Mt) model.

for the Mh, Mo and Mt models were 10%, 23% and 19%, respectively.

In the estimation of gender-specific population size estimates, the Mh model was again selected as the best fit model, using likelihood ratio tests in CAPTURE (White *et al.*, 1982), in modeling male sampling information, whereas the Mo model was selected in modeling female sampling information. Calculated estimates for sampled males, using the Mh model, was 44 (95% CI 36-60) and for females, using the Mo model, was 68 (95% CI 38,191). In the estimation of females, likelihood ratio tests ranked the Mh model only slightly below (0.80) that of the Mo model (0.83). An estimate of the number of females from the Mh model was 47 (95% CI 38, 64). Calculated CV values for the Mh model were 14% in estimating males and females while, for females alone, the Mo model returned a high CV value of 40%.

The costs of the genetics surveys included the actual flight time of 5.0 hrs over two days at CDN\$ 1200 per hr for the 2-sampling period survey in 2007 and 7.5 hrs flight time over three days for the 3-sampling period survey in 2009. DNA analysis for 100 samples at \$30 per sample totalled CDN\$ 3000 for the two sampling periods and CDN\$ 4920 (164 samples) for

the three sampling periods. So, the total cost of the genetics survey of the Slate Islands was approximately CDN\$ 9000 for the 2-sampling period approach in 2007 and CDN\$ 13 920 for the 3-sampling period approach in 2009. Costs do not include aircraft ferrying costs and times are based on flights originating from Terrace Bay (Table 2).

## Discussion

All three techniques of estimating the population size of caribou on the Slate Islands gave results with large, overlapping confidence intervals. However, the population estimate based on walking transects and genetic sampling with the 2-window approach had much wider confidence intervals than the FLIR survey or genetic sampling using the 3-window approach. Whereas confidence in population estimates from genetic sampling can be improved by adding more sampling effort and periods, estimates based on walking transects are greatly influenced by observer bias (i.e., experience) that cannot be readily corrected; increasing the number of transects walked will help but observer bias remains high. The most common source of bias in walking transects is the human error associated with a false observation or

failure to record an animal along a transect. Careful consideration must also be given to the dispersion of transects through areas to provide appropriate sampling. Ground-based estimates are also limited in capacity to determine sex ratios, cow-calf ratios and group sizes due to limited visibility and observer disturbance. Nonetheless, ground-based transects were the least expensive of the survey options that we compared on the Slate Islands with boat access. Ground-based surveys work best in small, easily accessible areas, but would lose any cost advantage if required over larger and more remote areas requiring access by aircraft. One must also consider timing restrictions with each survey technique. Both FLIR and ground surveys are best conducted when deciduous vegetation has lost its leaves and DNA extraction for genetic analysis produces better results with winter collections of faecal pellets (Ball *et al.*, 2007).

The caribou population estimate from the FLIR survey represents a minimal value that may have been limited by the rugged landscape of the Slate Islands and possibly dense conifer forest cover; subsequent FLIR surveys for moose and caribou in a conifer-dominated landscape on the mainland north of the Slate Islands, however, indicated this forest type does not severely limit detection (A. Rodgers, unpubl. data). Detection rates using FLIR are greater than those achieved by standard aerial census (Naugle *et al.*, 1996; Havens & Sharp 1998; Gill *et al.*, 1997; Bontaites *et al.*, 2000) and are subject to less observer bias caused by experience, fatigue, air sickness, etc. (Caughley, 1974; LeResche & Rausch, 1974) but the possibility of not detecting all animals and undercounting remains a potential source of error (Thompson, 2004; Drake *et al.*, 2005). As with virtually all wildlife survey methods, double counting can lead to biased population estimates. Because the FLIR survey of the Slate Islands was carried out over two days, we cannot completely eliminate the possibility that caribou were double-counted. Consequently, the caribou population estimate from the FLIR survey may be even lower than reported (Table 1).

Similar to standard aerial census methods, FLIR has the additional advantage of objectively detecting multiple species (e.g., moose, wolves) in the same survey. However, the window of opportunity for FLIR surveys is wider than for aerial surveys that require appropriate snow conditions in winter; an important consideration in a period of climate change that may produce mild winters with less snow. Although FLIR surveys require an experienced sensor operator and specialized equipment, they are usually less expensive (Adams 1995; Bernatas & Nelson, 2004) and require less expertise and special equipment than genetic

sampling. Occupancy estimation using FLIR and subsequent modeling may provide a cost-effective approach to broad-scale caribou population monitoring covering much larger geographical extents.

Comparison of the FLIR estimate of caribou population size with genetic sampling in three periods, suggests the detection rate of FLIR on the Slate Islands was about 60%. Thus, combining genetic sampling with other survey methods such as FLIR or aerial surveys can provide a correction factor for detection rate. Alternatively, a correction factor could be determined by marking individuals (e.g., radio collars) in a population prior to a survey (Bernatas & Nelson, 2004).

Genetic sampling can provide population estimates with high confidence in a closed system like the Slate Islands and in populations where population modeling assumptions can be verified in the use of mark-recapture models (Hettinga, 2010). However, in this study, variation in the number of animals sampled at each sampling time may have introduced bias in calculated estimates. In particular, in the sampling of caribou faecal pellets in 2007, low recapture rates (17%) were apparent and likely led to a positive bias in the calculated estimate which was also relatively imprecise (CV = 37%). In the calculation of population size using the 2009 collected samples, the incorporation of three sampling periods, as well as the use of mark-recapture models in program CAPTURE (White *et al.*, 1982), likely reduced the amount of bias in calculated population size estimates; despite variation in the number of animals sampled at each sampling time (Table 3). Regardless, because the two methods were applied in different years, comparisons of population size estimated from the 2-sample and 3-sample approach must assume there was no substantial change in population growth rate ( $\lambda$ ) between sampling periods.

Genetic sampling may be an expensive option if a lot of helicopter time is required; however, the collection of faecal samples can be an easy addition to an existing survey (e.g., aerial census by helicopter), thus being very cost effective. Additional benefits provided through the collection of faecal pellets include the potential for other genetics based testing in assessing population bottlenecks (Petersen *et al.*, 2010), meta-population structure (Ball *et al.*, 2010) and sex-ratios (Vors, 2006) and other faecal-based parameters, including: hormonal information to assess pregnancy and stress indicators (Messier *et al.*, 1990; Vors, 2006), the size of pellets as an indicator of age-range (Ball, 2010), diet information (Boertje, 1990) and parasite load (Gray & Samuel, 1986). In conducting multi-year sampling events there is also the potential for

the use of open population models where population demographic parameters including population rate of growth and recruitment rates can be estimated (Hettinga, 2010). Non-invasive genetic sampling for estimating population size has been done for mainland mountain and boreal-dwelling populations (Hettinga, 2010). Isolated populations like the Slate Islands are well suited to meeting the assumptions of closed population modelling; however, mainland populations must work within stricter definitions. Notably, additional attention should be paid to the boundaries of the study area and the timing of sampling periods to minimize chances of individuals moving out of or into the study area.

Ultimately, the best survey method to use will depend on the monitoring/research question(s) asked and resources available. Ground-based surveys may be sufficient if a rough estimate of population size is required, but more expensive surveys may be required if a more accurate and/or precise estimate is needed. FLIR and the 3-window genetic approach of sampling provided the most precise estimates in our comparisons. Given a known detection error for a study area, FLIR may be a cost effective monitoring method, but if the detection error is unknown, the 3-window genetic sampling approach will provide a more accurate and precise estimate. A combination of techniques may also be a productive approach, as the benefits of each technique are unique and convergence of population estimates will provide greater certainty to management plans for caribou recovery.

## Acknowledgements

We are grateful to the numerous individuals who participated in field and laboratory work that contributed to this study. We also appreciate additional GIS support from Yuewei Wang and Jevon Hagens. Thanks to Dr. Brian McLaren for supporting the King Census component of this project. Thanks to the staff of the Ontario Ministry of Natural Resources who participated in genetic collections and field surveys on the Slate Islands. Funding for this study was provided by the Ontario Public Service Innovation Fund, Ontario Parks program, Ontario Ministry of Natural Resources Species at Risk program, NSERC and Lakehead University. We appreciate the constructive comments and thorough review of this manuscript by Dr. Martin-Hugues St-Laurent and Dr. Isabelle Schmelzer.

## References

Adams, K.P. 1995. *Evaluation of moose population monitoring techniques and harvest data in New Hampshire*. Masters Thesis, University of New Hampshire, Durham, USA.

- Arnason, A.N., Schwarz, C.J., & Gerrard, J.M. 1991. Estimating closed population size and the number of marked animals from sighting data. – *Journal of Wildlife Management* 55:716-730.
- Ball, M.C. 2010. Faecal pellet size can be used to differentiate age-classes in caribou: implications for non-invasive genetic studies. – *Conservation Genetic Resources* 2:239-241.
- Ball, M.C., Finnegan, L., Manseau, M., & Wilson, P.J. 2010. Integrating multiple analytical approaches to spatially delineate and characterize genetic population structure: an application to boreal caribou (*Rangifer tarandus caribou*) in central Canada. – *Conservation Genetics* 11:2131-2143.
- Ball, M.C., Pither, R., Manseau, M., Clark, J., Peterson, S.D., Kingston, K., Morrill, N., & Wilson, P.J. 2007. Characterization of target nuclear DNA from faeces reduces technical issues associated with the assumptions of low-quality and quantity template. – *Conservation Genetics* 8:577-586.
- Bergerud, A.T. 2001. The herbivores of the Slate Islands: 1974-2001. Unpubl. progress report prepared for the Ontario Ministry of Natural Resources.
- Bergerud, A.T., Dalton, W.J., Butler, H., Camps, L. & Ferguson, R. 2007. Woodland caribou persistence and extirpation in relic populations on Lake Superior. – *Rangifer* Special Issue No. 17:57-78.
- Bernatas, S. & Nelson, L. 2004. Sightability model for California bighorn sheep in canyonlands using forward-looking infrared (FLIR). – *Wildlife Society Bulletin* 32:638-647.
- Boertje, R.D. 1990. Diet quality and intake requirements of adult female caribou of the Denali herd, Alaska. – *Journal of Applied Ecology* 27:420-434.
- Bontaites, K.M., Gustafson, K.A., & Makin, R. 2000. A Gasaway-type moose survey in New Hampshire using infrared thermal imagery: preliminary results. – *Alces* 36:69-75.
- Buckland, S.T., Anderson, D.R., Burnham, K.P., Laake, J.L., Borchers, D.L., & Thomas, L. 2001. *Introduction to Distance Sampling*. Oxford University Press, Oxford, UK.
- Caughley, G. 1974. Bias in aerial survey. – *Journal of Wildlife Management* 38:921-933.
- Caughley, G. 1977. *Analysis of Vertebrate Populations*. Wiley & Sons, London, UK.
- Chao, A. & Huggins, R.M. 2005. Classical closed-population capture-recapture models - In: Amstrup, S.C., McDonald, T. L. & Manly, B.F.J. (eds.). *Handbook of Capture-Recapture Analysis*, pp. 22-35. Princeton University Press, Princeton, New Jersey, USA.
- Chapman, D. G. 1951. *Some properties of the hypergeometric distribution with application to zoological censuses*. *University of California Publications in Statistics* 1:131-160.
- Courtois, R., Gingras, A., Dussault, C., Breton, L., & Ouellet, J. 2003. An aerial survey technique for the forest-dwelling ecotype of Woodland Caribou,

- Rangifer tarandus caribou*. – *Canadian Field Naturalist* 117:546-554.
- Cringan, A.T. 1956. *Some aspects of the biology of caribou and a study of the woodland caribou range of the Slate Islands, Lake Superior, Ontario*. Masters Thesis, University of Toronto, Ontario, Canada.
- Cronin, M.A., MacNeil, M.D., & Patton, J.C. 2005. Variation in mitochondrial DNA and microsatellite DNA in caribou (*Rangifer tarandus*) in North America. – *Journal of Mammalogy* 86:495–505.
- Davis, D.E. & Winstead, R.L. 1980. Estimating the numbers of wildlife populations. – In: Schemnitz, S.D. (ed.). *Wildlife management techniques manual*. 4<sup>th</sup> ed., pp. 221-245. The Wildlife Society, Bethesda, Maryland, USA.
- Drake, D., Aquila, C., & Huntington, G. 2005. Counting a suburban deer population using Forward-Looking Infrared radar and road counts. – *Wildlife Society Bulletin* 33:656-661.
- ESRI. 2006. ArcMap 9.2. Environmental Systems Research Institute, Redlands, California, USA.
- Euler, D.L., Snider, B., & Timmermann, H.R. 1976. Woodland caribou and plant communities on the Slate Islands: Lake Superior. – *Canadian Field Naturalist* 90:17-21.
- Evelt, I.W. & Weir, B.S. 1998. *Interpreting DNA evidence: statistical genetics for forensic scientists*. Sinauer, Sunderland, MA, USA.
- Frantz, A.C., Pope, L.C., Carpenter, P.J., Roper, T.J., Wilson, G.J., Delahay, R.J., & Burke, T. 2003. Reliable microsatellite genotyping of the Eurasian badger (*Meles meles*) using faecal DNA. – *Molecular Ecology* 12:1649-1661.
- Gill, R.M.A., Thomas, M.L. & Stocker, D. 1997. The use of portable thermal imaging for estimating deer population density in forest habitats. – *Journal of Applied Ecology* 34:1273-1286.
- Gray, J.B. & Samuel, W.M. 1986. *Parelaphostrongylus odocoilei* (Nematoda: Protostrongylidae) and a protostrongylid nematode in woodland caribou (*Rangifer tarandus caribou*) of Alberta, Canada. – *Journal of Wildlife Diseases* 22:48-50.
- Hansen, H., Ben-David, M., & McDonald, D.B. 2008. Effects of genotyping protocols on success and errors in identifying individual river otters (*Lontra canadensis*) from their faeces. – *Molecular Ecology Notes* 8:282-289.
- Haroldson, B.S., Wiggers, E.P., Beringer, J., Hansen, L.P., & McAninch, J.B. 2003. Evaluation of aerial thermal imaging for detecting white-tailed deer in a deciduous forest environment. – *Wildlife Society Bulletin* 31:1188-1197.
- Havens, K.J. & Sharp, E.J. 1998. Using thermal imagery in the aerial survey of animals. – *Wildlife Society Bulletin* 26:17-23.
- Hettinga, P.N. 2010. *Use of fecal DNA to estimate population demographics of the boreal and southern mountain ecotypes of woodland caribou*. Master of Science Thesis, University of Manitoba, Winnipeg, Manitoba, Canada. 118pp.
- King, R.T. 1937. Ruffed grouse management. – *Journal of Forestry* 35:523-532.
- Leopold, A. 1933. *Game management*. Charles Scribner's Sons. New York, New York, USA. 481pp.
- LeResche, R.E. & Rausch, R.A. 1974. Accuracy and precision of aerial mouse censusing. – *Journal of Wildlife Management* 38:175–182.
- Lincoln, F.C. 1930. Calculating waterfowl abundance on the basis of banding returns. – *U.S. Department of Agriculture Circular*. No 118, May 1930.
- McDonald, L. 2004. Sampling rare populations. – In: Thompson, W.L. (ed). *Sampling rare or elusive species: concepts, designs, and techniques for estimating population parameters*, pp.11-42. Island Press, Washington, D.C., USA.
- McGregor, C.A. 1974. A vegetation inventory of the Slate Islands. – *Environmental Planning Series, Life Science Report*. Vol.V, No.17. Ontario Ministry of Natural Resources, Division of Parks, North Central Region. Park Planning Branch, Environmental Planning Section.
- McLoughlin, P.D., Paetkau, D., Duda, M., & Boutin, S. 2004. Genetic diversity and relatedness of boreal caribou populations in western Canada. – *Biological Conservation* 118:593–598.
- Messier, F., Desaulniers, D.M., Goff, A.K., Nault, R., Patenaude, R., & Crête, M. 1990. Caribou pregnancy diagnosis from immunoreactive progesterins and estrogens excreted in feces. – *Journal of Wildlife Management* 54:279-283.
- Middleton, H. 1960. Caribou crossing from the Slate Islands, March 15, 1960. Harold Middleton to District Forester Geraldton, Ontario. Cited in C.A. McGregor: A vegetation inventory of the Slates Islands. – *Environmental Planning Series, Life Science Report*. Vol.V, No.17. Ontario Ministry of Natural Resources, Division of Parks, North Central Region. Park Planning Branch, Environmental Planning Section.
- Mills, L.S., Citta, J.J., Lair, K.P., Schwartz, M.K., & Tallmon, D.A. 2000. Estimating animal abundance using non-invasive DNA sampling: Promises and pitfalls. – *Ecological Applications* 10:283-294.
- Moore, S.S., Barendse, W., Berger, K.T., Armitage, S.M., & Hetzel, D.J. 1992. Bovine and ovine DNA microsatellites from the EMBL and Genbank databases. – *Animal Genetics* 23:463–467.
- Naugle, D.E., Jenks, J.A. & Kernohan, B.J. 1996. Use of thermal infrared sensing to estimate density of white-tailed deer. – *Wildlife Society Bulletin* 24, 37-43.
- OMNR. 1986. *Slate Islands Provincial Park: Background Information and Optional Plans*. Ontario Ministry of Natural Resources, Thunder Bay, Ontario.
- Otis, D. L., Burnham, K.P., White, G.C., & Anderson, D.R. 1978. Statistical inference from capture data on closed animal populations. – *Wildlife Monographs* 162: 135pp.

- Petersen, S.D., Manseau, M., & Wilson, P.J. 2010. Bottlenecks, isolation, and life at the northern range limit: Peary caribou on Ellesmere Island, Canada. – *Journal of Mammalogy* 91:698-711.
- Pollock, K.H., Marsh, H., Bailey, L.L., Farnsworth, G.L., Simons, T.R., & Alldredge, M.W. 2004. Separating components of detection probability in abundance estimation: An overview with diverse examples. – In: Thompson, W.L. (ed.). *Sampling rare or elusive species: concepts, designs, and techniques for estimating population parameters*, pp. 43-58. Island Press, Washington, D.C., USA.
- Pollock, K.H., Nichols, J.D., Brownie, C., & Hines, J.E. 1990. Statistical inference for capture-recapture experiments. – *Wildlife Monographs* 107. 97pp.
- Seber, G.A.F. 1982. *The estimation of animal abundance and related parameters*. Second ed. MacMillan, New York, NY, USA.
- Sinclair, A.R.E. & Caughley, G. 1994. *Wildlife ecology and management*. Blackwell Science, London, UK.
- Thomas, D. 1998. Needed: less counting of caribou and more ecology. – *Rangifer* Special Issue 10:15-23.
- Thomas, L., Buckland, S.T., Rexstad, E.A., Laake, J.L., Strindberg, S., Hedley, S.L., Bishop, J.R.B., Marques, T.A., & Burnham, K.P. 2010. Distance software: design and analysis of distance sampling surveys for estimating population size. – *Journal of Applied Ecology* 47:5-14. DOI: 10.1111/j.1365-2664.2009.01737.x
- Thompson, W.L. 2004. Future directions in estimating abundance of rare or elusive species. – In: Thompson, W.L. (ed.). *Sampling rare or elusive species: concepts, designs, and techniques for estimating population parameters*, pp. 389-399. Island Press, Washington, D.C., USA.
- Vincent, J.P., Hewison, A.J.M., Angibault, J.M., & Cargnelutti, B. 1996. Testing density estimators on a fallow deer population of known size. – *Journal of Wildlife Management* 60:18-28.
- Vors, L.S. 2006. *Woodland caribou extirpation and anthropogenic landscape disturbance in Ontario*. M.Sc. Thesis, Trent University, 146pp.
- White, G.C., Anderson, D.R., Burnham, K.P., & Otis, D.L. 1982. *Capture-recapture and removal methods for sampling closed populations*. Report LA-8787-NERP. Los Alamos National Laboratory, Los Alamos, NM, USA.
- White, G.C. & Burnham, K.P. 1999. Program MARK: survival estimation from populations of marked animals. – *Bird Study* 46:S120 - S139.
- Wilberg, M.J. & Dreher, B.P. 2004. GENECAP: a program for analysis of multilocus genotype data for non-invasive sampling and capture-recapture population estimation. – *Molecular Ecology Notes* 4:783-785.
- Wilson, G.A., Strobeck, C., Wu, L. & Coffin, J.W. 1997. Characterization of microsatellite loci in caribou *Rangifer tarandus*, and their use in other artiodactyls. – *Molecular Ecology* 6:697-699.

