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Using temporary dye marks to estimate ungulate population abundance in southwest Yukon, Canada

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Abstract: We describe the protocols of two mark-resight abundance surveys, using temporary dye-marks, for the Aishihik woodland caribou (*Rangifer tarandus caribou*) and wood bison (*Bison bison athabascae*) populations (herds) in the southwest Yukon Territory, Canada. We also provide recommendations based on experiences from these surveys for biologists and managers considering this approach. The Aishihik woodland caribou herd was the focus of intensive management in the 1990s aimed at recovering the herd. Following recovery activities, a target size of 2000 animals was determined and the Champagne-Aishihik Traditional Territory Community-Based Wildlife Management Plan recommended an estimate of the herd's size be completed before the year 2013. We used an aerial mark-resight approach to estimate the herd's size in March 2009. Caribou (n = 59) were marked from a helicopter with temporary dye, delivered via a CO_2-powered rifle. Two independent resighting sessions were subsequently carried out via helicopter. The herd was estimated at 2044 animals (90% CI: 1768 – 2420) with an overall resighting rate of 0.47. The mean annual growth rate (λ) of the herd from 1997 – 2009 was 1.05 (SE = 0.01). The Aishihik wood bison herd was estimated at 1151 (90% CI: 998 – 1355). Our study suggests that ungulates temporarily marked with dye can be successfully used to obtain statistically sound population estimates.

Key words: abundance; dye-marks; mark-resight; *Rangifer tarandus caribou*; Yukon Territory.

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Introduction

Population abundance is a key parameter used by managers and other stakeholders for effective and sustainable management and conservation of wildlife populations (Milner-Gulland & Rowcliffe, 2007). This information is used, for example, to ensure harvest is sustainable (Sæther *et al*., 2001), to establish baseline conditions prior to anthropogenic activities on the landscape, and to subsequently assess the impacts of these activities (Sorensen *et al*., 2008). Furthermore, abundance estimates can be used to obtain a greater understanding of ecological processes influencing a single population's (e.g., Jenkins & Barten, 2005) or multiple populations' dynamics (e.g., Vors & Boyce, 2009; Wittmer *et al*., 2010).

Estimating abundance in large and remote areas can be expensive and time consuming. This is made more challenging when surveying animals occurring at low densities. A variety of methods are available to estimate population abundance (e.g., Schwarz

& Seber, 1999), and a number have been used to estimate woodland caribou abundance in the Yukon including total counts (e.g., Hayes *et al.*, 2003), a stratified random quadrat (SRQ) method (Farnell & Gauthier, 1988), and mark-resight surveys using radio-collared animals (Environment Yukon, unpubl. data).

The Aishihik caribou (*Rangifer tarandus caribou*) herd (AH) in the southwest Yukon Territory (Yukon), Canada, is a population of the Northern Mountain ecotype of woodland caribou. These caribou are legally listed in the Canadian *Species at Risk Act* as a species of *Special Concern* (COSEWIC, 2002). Following declines in the 1980s and early 1990s, AH was the focus of an intensive population recovery effort in the 1990s (Hayes *et al.*, 2003) and is one of the better-studied herds in the Yukon (Farnell *et al.*, 1998). Recovery actions for AH included limiting human harvest and reducing wolf (*Canis lupus*) populations through sterilization and lethal control (Hayes *et al.*, 2003). A community-based fish and wildlife management plan for the Champagne & Aishihik First Nation's traditional territory, in which AH is located, recommended the herd's size be estimated by 2013 to determine if the herd had reached a management target of 2000 animals. The most recent estimate of the AH was 1148 animals (90% CI: ± 6.5%; Hayes *et al.*, 2003) in 1997. Additionally, there are local concerns regarding the impact of the reintroduced Aishihik wood bison (*Bison bison athabascae*) population (AWB) on the AH. From 1988 to 1992, 170 bison were released into the wild in the Aishihik area and First Nations have expressed concern over the potential impact of reintroduced wood bison on sympatric caribou (Fischer & Gates, 2005).

We lacked radio-collared animals in the AH to correct for sightability, and were wary of attempting a total count approach given its unreliability (e.g., Caughley & Goddard, 1972). Furthermore, previous applications of an SRQ approach on the AH required greater resources (e.g., financial and personnel) than were available (Environment Yukon, unpubl. data). Thus, we applied a standard mark-resight approach using temporary dye (i.e., paintballs) to mark a subsample of animals in AH to estimate its size. This approach has been used to estimate abundance in other ungulate populations including elk (*Cervus elaphus*; Skalski *et al.*, 2005) and mountain goats (*Oreamnus americanus*; Cichowski *et al.*, 1994; Pauley & Crenshaw, 2006). We also adopted this approach to estimate the size of the AWB which has an overlapping range with that of AH.

The primary objective of this paper is to describe and provide practical recommendations for biologists and managers considering a mark-resight approach using temporary dye marks. These recommendations are based on our experiences estimating abundance of the AH and the AWB. Our purpose for briefly including the AWB in this paper is to develop and strengthen our recommendations based on two separate surveys, and species. A secondary objective of this paper is to discuss our findings with respect to the population dynamics of the AH, which was the initial impetus for us to use this method.

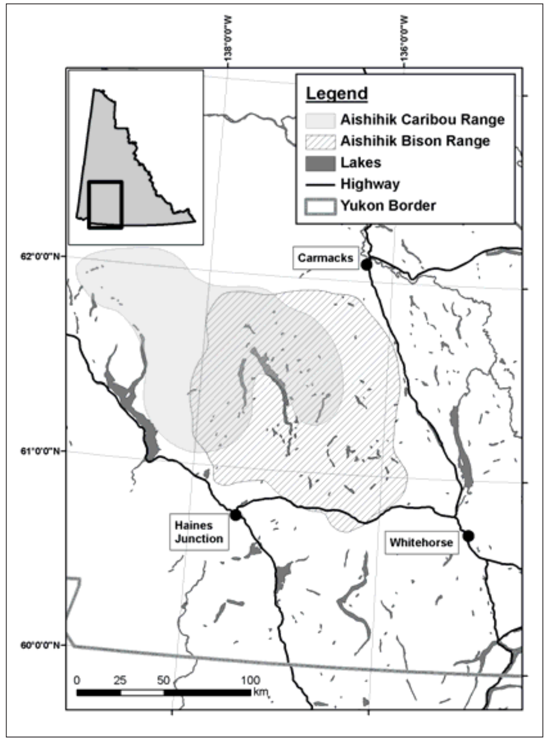

Fig. 1. Generalized range boundaries of the Aishihik caribou and wood bison populations in the southwest Yukon Territory, Canada.

Material and methods

Study area

Both the AH and AWB are located in the southwest Yukon (Fig. 1) within the Boreal Cordillera ecozone (Smith *et al.*, 2004). The area hosts a full complement of native ungulates and large carnivores, including woodland caribou, wood bison, moose (*Alces americanus*), thinhorn sheep (*Ovis dalli*), mule deer (*Odocoileus hemionus*), grizzly bear (*Ursus arctos*), black bear (*Ursus americanus*), wolverine (*Gulo gulo*), and wolves. Topography of the area is mountainous with high plateaus and is characterized by rounded and rolling hills in the east with more rugged terrain in the west. Mean elevation in the area is approximately 1400 m above sea level (asl) and ranges from approximately 800 to 2300 m asl.

The area lies within the St. Elias Mountains rain shadow and is semi-arid, with annual precipitation averaging approximately 250 – 300 mm/year. Mean annual temperature is approximately -3 °C (winter average: -17 °C; summer average: 10 °C). Lower elevation areas below treeline consist primarily of open canopy white spruce (*Picea glauca*) forest. Higher elevations are characterized by shrub (*Betula* spp. and *Salix* spp.) communities in the subalpine. Alpine communities include low lying shrubs, *Dryas* spp., and various graminoids, mosses, and lichens. A detailed description of the area is provided by Smith *et al.* (2004).

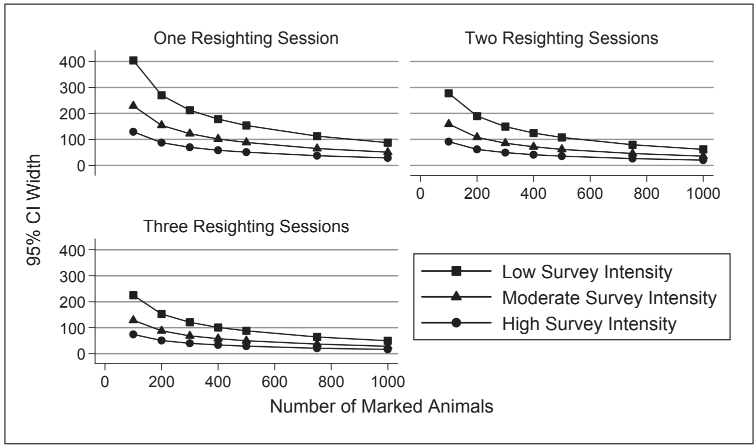

Fig. 2. Simulation results for determining a sampling strategy for the mark-resight survey of the Aishihik caribou herd. Survey intensity is defined by sightability rates and characterized as low (0.50), moderate (0.75), and high (0.90). Simulations were carried out assuming a true population of 2000 individuals. The y-axis represents the width of the 95% confidence interval of the estimate.

Survey protocols

Prior to the caribou study we used a Monte Carlo simulation approach, using the software NOREMARK (White, 1996), to guide our decisions for the survey design. Differing combinations of survey parameters (Fig. 2) were assessed with respect to their effect on precision (i.e., confidence interval width) of the estimate including the number of marked animals, the number of resighting surveys, and resighting rate (i.e., survey intensity). We used 2000 animals as the assumed population size for all simulations. This was based on the minimum number of animals known to be present in the herd from observations during recent fall composition surveys, and projections of a simple population model (Environment Yukon, unpubl. data). We used three resighting rates (0.50, 0.75, and 0.90) representing a range of rates. However, we acknowledge that of the three parameters considered in the simulations, this was the variable we would have the least control over largely due to the effect of environmental conditions (e.g., weather) on sightability.

Based on our computer simulations and financial considerations, our design was to attempt to mark a minimum of 150 animals, followed by two independent resighting sessions. To minimize potential biases associated with marking, and subsequently resighting, animals in different sized groups (Skalski *et al.*, 2005) we aimed to mark 20% of the animals in each group. Marking was carried out from a relatively fast and maneuverable helicopter (A-Star 350B1) with a three-member crew: navigator/data recorder, shooter, and a shooter's assistant. The shooter was positioned behind the pilot with the shooter's assistant seated in the adjacent rear seat. The navigator/data recorder occupied the front passenger seat. To mark animals (Skalski *et al.,* 2005; Pauley & Crenshaw, 2006) we used temporary oil-based dye (paint) pellets (Nelson Paint Company of Canada; Sault Ste. Marie, ON) delivered from a compressed CO_2 charged rifle (Tippmann A-5; Tippmann Sports LLC, Fort Wayne, IN). Dyes were non-toxic and of the same type used in the livestock and veterinary fields. Our primary choice of colours was bright orange and bright green as we assumed these colours would be highly visible for subsequent resighting. We also marked a smaller number of animals using yellow, blue, and red dyes, to assess their visibility in the field. To ensure marks were visible for the resighting sessions, we attempted to mark each animal with a minimum of five dye pellets. During marking operations we avoided the animal's head and attempted to place all marks near their hind quarters. Thus, for the purposes of this survey a marked animal was one hit with a minimum of five pellets on its hind quarters (e.g., rump or flank). Calves were not marked.

We used historical late-winter animal locations to guide our aerial search and to increase our efficiency by avoiding those areas where caribou had never been observed during late winter. Additionally, prior to marking we used a fixed-wing aircraft (Cessna 205) to survey the perimeter of the herd's known range to delineate the outer edge of the study area by locating tracks or animals.

During marking, we focused on alpine areas and flew contours along mountain sides and along valleys and alpine plateaus. When a group of animals was located we first tallied its size to determine the number of animals to mark. We then flew towards the group, attempting to move animals uphill if possible, and marked animals from behind at a distance of approximately 5 m (i.e., within the rotor wash of the helicopter). The helicopter was equipped with a sliding door that was opened prior to marking to enable the shooter to mark animals with a wide range of movement. Marking was generally completed within 30 – 45 seconds, after which the helicopter immediately lifted away from the target animal. Marking occurred during 4 – 7 March 2009.

Shortly after marking, we carried out two consecutive and independent aerial resighting sessions. Resighting crews were independent of one another and consisted of three observers and a pilot. Resighting surveys took place using a Bell 206B helicopter. The pilot remained the same for both surveys but was instructed not to impart any information on animal locations to the second resighting crew to ensure independence between the two resighting surveys. During the resighting surveys groups of animals were counted and the number of marked animals was recorded. The first resighting session (14 hours of flying) took place 7 – 9 March 2009 and the second (12.5 hours of flying) occurred on 10, 11, and 15 March 2009.

In July 2009 we conducted a mark-resight population estimate of the AWB. We used the same marking and resighting methodology as for AH described above. Bison were marked with blue dye on 25 July, followed by two independent resighting sessions during 26 – 29 July 2009. During July, this population of wood bison are typically aggregated and found in alpine areas (Environment Yukon, unpubl. data), which facilitates a relatively high resighting rate. Our target was to have 75 marked animals in the population, based on an assumed population size of 1200 wood bison. Blue was selected as the most visible and durable dye colour based on testing on captive bison at the Yukon Wildlife Preserve. Twenty-four bison fitted with radio-collars were also present in the herd during the survey and these animals were also included in the marked sample and were not dye-marked. The survey boundary was delineated based on observations of radio-collared animals located via fixed-wing aircraft 7 days prior to our marking session, and existing radio-telemetry survey data from previous years.

Statistical analysis

We used the program NOREMARK (White, 1996) to estimate abundance, for both the AH and AWB, by fitting the data to a joint hypergeometric distribution (Neal *et al.*, 1993). Given the relatively short time frame between marking and the resighting surveys we assumed the populations were both demographically and geographically closed (i.e., no animals died and no animals left or entered the study area). Confidence intervals were determined using a profile likelihood approach (White, 1996).

The average annual (geometric) growth rate (λ) was estimated for the AH using the equation $N_t = N_0\lambda^t$ (Caughley, 1977), where N_t is the 2009 estimate, N_0 is the 1997 estimate, and t =12 (i.e., the number of years between estimates). The SE of λ was estimated using the Delta method with the 'emdbook' package (version 1.3.1; Bolker, 2008) for the statistical software R (version 2.13.0; R Development Core Team, 2011).

Results

We marked 122 of 793 caribou observed over approximately 14 hours of flying time (approximately 1960 km) during the initial marking session. However, technical problems with the bright orange dye resulted in poorly marked animals and other colours such as yellow were deemed too difficult to observe during the resighting sessions, potentially leading to missed marks. Therefore, for analysis we only considered animals with either blue or bright green dyes to be "marked"; resulting in 59 marked animals. Due to larger than expected group sizes (mean = 27.3, range: 1 – 121) we were unable to mark 20% of the animals in each group, as doing so would have placed too much stress on the animals.

Resighting rates in each of the resight sessions were similar (Table 1) with an overall resighting rate of 0.47. Flight lengths for the first and second resighting sessions were approximately 1460 km and 1580 km, respectively. Based on observed marked and unmarked animals, the AH's size was estimated at 2044 (90% CI: 1768 – 2420). Because of the smaller number of marked animals used in the analysis, the reduced precision in the estimate was expected. The annual population growth rate (λ) of the herd, based on this estimate and the previous SRQ estimate from 1997, was estimated at 1.05 (SE = 0.01).

In July 2009, 59 bison were marked with blue dye during approximately 6 hours of flying and together with the 24 previously radio-collared individuals, 83 bison were "marked" in the population. As with the caribou census, resighting rates of the two resight sessions were also similar (Table 1). The size of the AWB was estimated at 1151 (90% CI: 998 – 1355).

Discussion

Survey recommendations

We obtained acceptable estimates of the AH and AWB, based on the relatively narrow 90% confidence interval coverage. Moreover, the population estimates were consistent with our expectations, based on previous estimates, demographic data, and anecdotal observations (e.g., observed numbers of animals during composition surveys; Environment Yukon, unpublished data). The use of temporary dye marks allowed us to mark more animals than we would have been able to had we relied on using a subsample of radio-collared animals as the marked population, given the resources available. Monte Carlo simulations suggested that an increased proportion of the population being marked would increase the precision of our population estimates. This is not surprising and was reported earlier (Neal *et al*., 1993) using the same Monte Carlo simulation procedures.

The two mark-resight population estimates we conducted provided us an opportunity to identify those features and aspects of the studies which were useful and effective. We provide our experiences and recommendations in the hope they may be useful for biologists and managers considering this approach in their own work. We acknowledge that our recommendations and opinions are qualitative and generally not based on any formal experimental or comparative approach. Nevertheless, we feel these lessons learned may be useful.

Assuming there is some basic quantitative or qualitative information available for the population of interest, the use of the program NOREMARK (White, 1996) can be a useful tool to guide biologists in allocating marking and resighting efforts. Perhaps the most useful aspect of running a simulation study was the ability to determine how many animals to mark and how many resighting sessions to be flown (e.g., Fig. 2). Given that resighting rates can be influenced by many factors (Caughley *et al*., 1976; Anderson *et al*., 1998), some out of the researcher's control, assessing sample size and the number of resighting sessions over a range of differing resighting rates is a valuable approach.

Prior to marking, trials of different colour dyes on live animals can guide the best choice of dye colour to use in the field. In both of our studies, observers noted that blue was the most readily visible colour during the resighting sessions. However, this may not be applicable for all species, seasons, or environments. Because the blue contrasts with many natural environments and terrain, it is a highly recommended colour for increased visibility by search and rescue agencies (National Association for Search and Rescue, 2005).

During animal marking we strongly recommend the use of a fast and maneuverable helicopter. Marking ungulates with temporary dye requires low-level and dangerous flying and a maneuverable helicopter will reduce chase times thus reducing stress on the animals and increasing the safety level for the crew.

Our crews were made up of three members: a navigator/data recorder, a shooter, and a shooter's assis-

Table 1. Resighting summary data from the Aishihik caribou (March 2009) and wood bison (July 2009) mark-resight surveys.

Session	Total Animals Observed	Marked Animals Observed	Resighting Rate (SE)[c]
		Caribou	
Resight 1	1012	29[a]	0.49 (0.07)
Resight 2	928	27[a]	0.46 (0.07)
		Wood bison	
Resight 1	355	33[b]	0.39 (0.05)
Resight 2	512	31[b]	0.37 (0.05)

a: 59 marked animals available; b: 83 marked animals available; c: Resighting rates did not differ between sessions of the caribou (Z = 0.37, P = 0.712, n = 59) and wood bison (Z = 0.31, P = 0.750, n = 83) surveys, respectively.

tant. Fewer crew members would have substantially slowed down the marking operations as personnel would have had too many tasks in the helicopter. In addition to having multiple crew members, backup equipment (e.g., CO_2 rifles and barrels) and cleaning gear is also highly recommended. If one rifle breaks or malfunctions, significant time can be lost if the crew must return to replace it. Additionally, dye pellets can break in the barrel of the rifles and must be cleaned by the shooter's assistant. Having multiple barrels available to be changed rapidly also increases the overall efficiency of the marking operations.

Care and maintenance of the equipment can prove to be critical and greatly increase the likelihood of a successful marking operation. We recommend that dye pellets do not freeze or be exposed to extreme heat as extreme temperatures may damage the outer shell or alter the shape of the pellets. The technical problems with the bright orange pellets which we experienced during the marking of AH during late winter may have been due to these pellets freezing during shipping from the manufacturer in February. Upon closer inspection following the survey we noticed that nearly all pellets had been warped to a more ovoid shape rather than spherical. This change in shape may have lead to pellets prematurely bursting in the barrel of the rifle.

Having a properly tuned rifle is important. Regulators govern the amount of CO_2 entering the rifle and hence influencing the speed and trajectory of the pellets. Adjustments on these can have a significant influence on the effectiveness of the rifles, which is critical when firing at a moving animal from a helicopter. Ensuring rifles are working properly prior to field effort may only require a short amount of time relative to the overall benefits obtained during marking operations.

A limitation of this approach is the potential to violate a key assumption of mark-recapture analyses: that all animals have equal probability of being marked and resighted (Skalski *et al*., 2005). For animals occurring in groups (e.g., *Rangifer* sp.), detectability is often positively related to group size (Anderson *et al*., 1998). Thus, animals in larger groups may have a higher probability of being marked, and resighted, than those in smaller groups which may bias abundance estimates. This becomes a greater issue when groups are unequal in size and remain constant in size between marking and resighting sessions, and when individuals exhibit at least partial group fidelity.

The nature of the marking approach used here requires that large amounts of time should not be allowed for between marking and resighting in order to avoid losing marks (another assumption of mark-recapture analyses). To account for the potential biases associated with varying group sizes, Skalski *et al*. (2005) recommend marking a constant proportion of animals within a group, which we attempted here. They also recommend marking animals when they occur in smaller and less stable groups. For populations having varying sized groups and individuals exhibiting partial fidelity to those groups, Skalski *et al*. (2005) provide information on the degree of bias based on the overall variation in group sizes and the degree of fidelity that individuals exhibit. The degree of fidelity may be challenging to estimate however, as it is based on the correlation in group size an individual is associated with over time. The inability to uniquely identify dye-marked animals is one challenge; as is the minimum number of resighting sessions required to adequately estimate this correlation (i.e., fidelity). Prior monitoring of uniquely identifiable marked animals (e.g., through radio-collaring) could provide this information.

In this study we lacked uniquely marked individuals, and therefore could not formally estimate group fidelity. Two factors may have minimized the bias in our abundance estimate for the AH. First, the AH is an alpine-wintering herd (Kuzyk *et al*., 1999) which may reduce the influence of group size on sightability. That is, large and small groups may have similar detectability in treeless alpine areas under the same surveying route. The Chisana herd in the southwest Yukon (Kuzyk *et al*., 1999) also occurs in high elevation habitats with low tree cover and a recent mark-resight estimate, using radio-collared individuals, of it found no strong relationship between detection and group size ($\beta_{\text{Group Size}} = 0.77$, SE = 0.64, $P = 0.23$; Environment Yukon, unpublished data).

Second, groups may not have been constant in size in the AH over the marking and resighting sessions. For example, a closer examination of groups observed during the marking and resighting session in one drainage (Raft Creek) of the study area found a range of group sizes with similar numbers of total animals counted. During marking 140 animals were observed in four groups ranging in size from 12 to 62. Subsequently, in one resighting session 140 animals were observed in one group and in the other resighting session 180 animals were observed in five groups (range: 4 – 90).

Aishihik caribou population dynamics

The roughly 5% annual increase in the AH from 1997 to 2009 contrasts the broader pattern of *Rangifer* declines observed globally (Vors & Boyce, 2009) and at the local population level elsewhere (Wittmer *et al*., 2005). Licensed bull harvest of the herd was stopped prior to recovery efforts in the 1990s and a permit-based hunt began in 2002 with approximately 19 bulls harvested annually (Environment Yukon, unpubl.). Aboriginal subsistence harvest of the herd, not regulated by the Government of Yukon, was voluntarily halted during this time. Thus, cow harvest of the herd has been minimal, if present at all, from the 1990s to the present. Given the strong influence of adult female survival on ungulate population dynamics (Gaillard *et al*., 2000) the lack of a cow harvest may have contributed to this increase.

The increasing trends in the AH and AWB suggests there is little evidence that interspecific competition between caribou and bison has resulted in a decline of the caribou population. Fischer & Gates (2005) found little basis for competition between AH and AWB based on winter habitat and diet overlap. However, we are unable to assess what the trend in AH would have been in the absence of the AWB.

Conclusion

Overall we deemed the temporary dye mark-resight approach effective in our estimation of abundance of two ungulate populations. A key benefit of this approach is the relative speed in which many animals can be marked, greatly increasing the precision in the population estimate. Dye marks can be the sole marking source or can be used to augment existing marks (e.g., radio-collars) as was the case with the AWB survey. Augmenting existing marks may be useful in situations where, for example, only females are radio-collared and males are unmarked and spatially separated from females. In such situations making inferences on male resighting rates from marked females may be unjustified. Further, the general statistical framework (i.e., numbers of marked and unmarked animals observed during a survey) of this approach is relatively intuitive and may be easier to communicate to the public than more complex quantitative methods. Mark-resight estimators (McClintock & White, in press) have recently been incorporated into the software MARK (White & Burnham, 1999), greatly enhancing the ability to model both abundance and resighting rates, including the specification of temporal, group-level, and individual covariates.

A final recommendation we offer relates to the use of terminology. We use the term dye-marks rather than paintballs when discussing this methodology. We feel this euphemism conveys a more professional attitude towards the survey approach. Indeed, in many areas biologists must be aware of and respect cultural sensitivities surrounding the impact of research or management activities on wildlife (e.g., Wilson & McMahon, 2006). Use of less technical terms such as "paintballs" may convey a message that this survey approach is less rigorous than it actually is, and that in fact marking animals with dye may be less invasive than marking with collars which requires capture and handling.

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