

# Summaries of Wildlife Research Findings 2013



Minnesota Department of Natural Resources  
Division of Fish and Wildlife  
Wildlife Populations and Research Unit



# **SUMMARIES OF WILDLIFE RESEARCH FINDINGS 2013**

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## EVALUATION OF LOCALIZED DEER MANAGEMENT TECHNIQUES FOR THE REDUCTION OF DAMAGE CAUSED BY WHITE-TAILED DEER IN MINNESOTA: A PILOT STUDY

Gino D'Angelo

### SUMMARY OF FINDINGS

Minimizing damage caused by white-tailed deer (*Odocoileus virginianus*) is an important consideration for managing deer densities in Minnesota. I conducted a pilot study during April-December 2013 in southeast Minnesota to begin assessing the effectiveness of localized management of deer (i.e., targeted removal of deer in a limited area) to reduce damage to agricultural crops. I developed an efficient methodology for assessing crop damage caused by deer. I also used baited infrared camera surveys to estimate the abundance of deer on individual properties. Preliminary data on the number of deer harvested relative to the number of deer utilizing properties was important for planning the expansion of localized management in Minnesota. The pilot study provided the basis for a broader study which was initiated in spring 2014. A more comprehensive examination of crop losses relative to deer abundance will be conducted as part of the broader study.

### INTRODUCTION

Damage caused by white-tailed deer can be severe in the United States with  $\geq$ \$100 million lost annually by agricultural producers (Conover 1997). Researchers have used multiple methods to evaluate crops depredated by deer (Krueger and McAninch 1990, Wywiałowski 1996, Tzilkowski et al. 2002, Stewart et al. 2007), but there are no standard protocols. Regional factors such as growing regime (e.g., timing of planting, fertilizer and herbicide applications, harvest), conservation practices (e.g., contour farming, tillage, irrigation), and local conditions mean that specific techniques to evaluate crop depredation may not be applicable for all settings. Wywiałowski (1996) found that producers generally predicted losses to wildlife well. However, Tzilkowski et al. (2002) compared on-the-ground estimates of damage to corn with estimates by farmers. They found that estimates by farmers were not reliable for assessing an individual farmer's level of damage, but may be more useful to generate damage estimates for a larger sample of farms. Certainly, reliance on estimates of crop losses by producers is not ideal when seeking objective assessments to guide regulations for managing deer. Stewart et al. (2007) recommended that crop damage be assessed on a field-by-field basis using actual evaluation of the plants and the damage plants sustain from deer.

Results from previous studies have demonstrated through anecdotal evidence that population reduction of deer can reduce damage to agriculture (McShea et al. 1993, Frost et al. 1997, Conover 2001). In some situations, localized management has effectively reduced the abundance of deer to maintain lowered deer densities over time (McNulty et al. 1997). As a result, damage to resources targeted for protection should be reduced because fewer deer are available to cause damage. However, conditions including high deer densities in surrounding areas (Miller et al. 2010), seasonal migratory behavior of deer (Vercauteren and Hygnstrom 1998), and colonization by deer from adjacent populations (Comer et al. 2007) may inhibit the creation of sufficient temporal periods of low deer densities to provide resource protection. Studies of the effectiveness of localized management to reduce damage to specific properties in agricultural settings are lacking.

In many deer permit areas (DPAs) in Minnesota, deer are managed at or near population goals annually. However, complaints of deer damage from agricultural producers are common. During 2010, wildlife managers received 614 complaints about damage caused by wildlife, and 23% (n = 142) of those complaints involved deer (Reindl and Benson 2011). Complaints of depredation by deer in Minnesota include consumption of forage stored for livestock, damage to specialty crops (e.g., produce, Christmas trees, nursery stock), row crops (corn [*Zea mays*] and soybeans [*Glycine max*]), alfalfa (*Medicago sativa*), and forest stands.

Deer damage is reported throughout Minnesota, but distinct clusters of complaints occur in the southeastern and southwestern regions of the state (Reindl and Benson 2011).

In Minnesota, most deer damage occurs during the growing season in standing row crops and alfalfa in the field or during winter when deer consume forage stored for livestock. By excluding deer from stored forage, the damage can be effectively eliminated. Farmers who enter into a Cooperative Damage Management Agreement (CDMA) with Minnesota Department of Natural Resources (MNDNR) are eligible for a material assistance program to aid in the installation of exclusion fencing. However, funds for deer damage assistance are limited and fencing is only practical for protecting areas that are relatively small (i.e., stored forage and specialty crops). Sound and visual deterrents and taste and smell repellents have proven ineffective for reducing deer damage in agricultural fields (Belant et al. 1996, Belant et al. 1998, Gilsdorf et al. 2004). Therefore, most attempts to reduce damage to standing crops in Minnesota involve the use of localized deer damage management techniques such as shooting permits and depredation permits.

MNDNR Regional Offices have issued shooting permits to agricultural producers experiencing extreme damage caused by deer for use outside of hunting seasons. Shooting permits allow landowners to shoot deer at any time of day or night and with a high-powered rifle. For years 2004 through 2010, an average of 55 shooting permits for nuisance deer were issued annually for use during summer and winter (Reindl and Benson 2011). In southeast Minnesota, landowners with support from local legislators requested shooting permits to be issued during the regular hunting seasons to reduce depredation to standing row crops. As an alternative to their request, a pilot program using depredation permits allocated to specific properties (herein, focal properties) was instituted in 2012 in southeast Minnesota (Luedtke 2013). Depredation permits were to be used by private sport-hunters during regular hunting seasons. Additionally, a temporary MNDNR position, the Landowner Assistance Specialist, was created to administer the program in Fillmore, Goodhue, Houston, Olmsted, Wabasha and Winona counties.

Depredation permits allowed up to 15 hunters per focal property to harvest up to 5 antlerless deer in addition to established bag limits during regular hunting seasons. Consequently, an additional 75 deer could be harvested on an individual property using this program. To be eligible, applicants had to demonstrate: 1) crop losses verified by MNDNR personnel, 2) enrollment in a CDMA with MNDNR including a plan for deer hunting management, and 3) hunting was allowed on the property during the previous hunting season. Depredation permits were issued for 8 properties encompassing 971 hectares during 2012. Seventy five hunters were awarded permits and 166 deer were harvested on depredation permits.

The program was deemed successful because deer were harvested on depredation permits (Luedtke 2013). Anecdotally, landowners and hunters participating in the program felt that the program was beneficial. Several landowners suggested that private properties adjacent to their parcels were providing sanctuary to deer, which inhibited adequate harvest (C. Luedtke, personal communication). Undoubtedly, the service provided by MNDNR staff to facilitate positive interactions among landowners and hunters was important for improving localized management on focal properties. However, without objective measurement of relevant indices including deer abundance and crop depredation, the effects of deer harvest through localized management may not be fully understood. Documentation is lacking regarding the magnitude of damage caused by deer to various agricultural crops in Minnesota. With complaints of deer damage and localized overabundance of deer in other parts of the state, MNDNR Wildlife Managers would like the opportunity to utilize depredation permits throughout the state in future years (MNDNR Deer Management Committee, 28 January 2013, personal communication).

The goal of this pilot study was to develop methods to quantify damage caused by deer in an agricultural landscape which will improve understanding of the relationships between deer damage and localized deer abundance, and the efficacy of localized management.

## PILOT STUDY RESEARCH OBJECTIVES

1. To develop methods for evaluating depredation caused by white-tailed deer to agricultural crops in Minnesota.
2. To evaluate the intensity of deer management on focal properties where localized management of deer is utilized.
3. To provide a preliminary assessment of localized management in southeast Minnesota.

## STUDY AREA

The study was conducted in the Minnesota counties of Fillmore, Houston, and Winona. Southeast Minnesota is characterized by a mosaic of rolling limestone uplands dominated by agriculture (Mossler 1999). Typical crops include corn, soybeans, and alfalfa. Steep ravines cut by narrow streams are interspersed throughout the uplands. Ravines are rocky and primarily forested by mature hardwoods (Omernik and Gallant 1988).

DPA 345, 346, and 349 were included in the study area. Pre-fawn deer densities in these DPAs averaged 6 deer per km<sup>2</sup> (Grund and Walberg 2012), which represents the highest deer densities found in the farmland zone of Minnesota. An average of 2.3 deer per km<sup>2</sup> was harvested in these DPAs during 2012, which was nearly twice the statewide average (McInenly 2013).

## METHODS

### Experimental Design

*Crop Evaluations*—During the pilot study, my intent was to develop methods to evaluate agricultural crops, which would minimize: 1) visits to properties, 2) disturbance to the sites, and 3) disruption to normal agricultural processes. Therefore, to achieve these objectives I worked with landowners, who were willing to cooperate closely with MNDNR, but may not necessarily use localized management of deer on their properties. Refining methods during the pilot study improved my ability to work efficiently on properties where landowners may be less tolerant of my research. For the pilot study, I selected 3 corn fields and 3 soybean fields on properties where landowners were willing to communicate with me about their farming practices and afforded regular access for research purposes.

*Management Intensity*—Another important aspect of the pilot study was to estimate the intensity of localized deer management. I estimated deer abundance and surveyed landowners about deer harvest on 3 individual properties. Crop evaluations and estimates of management intensity were not conducted on the same properties.

### Data Collection

*Corn Evaluations*—I delineated 6 plots within each field, which were stratified into interior (beginning 25 m from the field edge) and edge (beginning on the field edge). Each plot included 2 paired 5-m X 5-m subplots separated by 5 m. One subplot of each pair was fenced to exclude deer and the other subplot was unfenced. I assigned the fencing randomly within each pair. Square enclosures were constructed with 2-m high heavy-duty plastic mesh attached to 4 2.4-m u-posts. Enclosures surrounding subplots were approximately 6 m X 6 m to reduce the effects of fencing on plants within the subplot. Enclosures were installed immediately following planting. When necessary, I temporarily ( $\leq 72$  hours) removed enclosures during herbicide application. I evaluated corn crops near the estimated date of plant maturity before

senescence. I recorded the number of rows, number of plants, plant height, level of herbivory per plant, and I classified the quality of each ear of corn relative to damage caused by deer for each subplot. I estimated grain yield (bushels per hectare) for fenced and unfenced subplots using a standard yield estimator. I consulted with the agricultural producer to determine the variety of corn planted in each field.

*Soybean Evaluations*—I established plots and installed exclosures in soybean fields immediately after planting. I used the aforementioned protocols for placement and number of plots as outlined for evaluations in corn fields. Exclosures surrounding subplots were approximately 5 m X 5 m. The square subplots were smaller (2 X 2 m) than those used for the corn evaluations. Soybeans are planted at a higher density than corn and the plants are shorter in height. The smaller subplots used for soybeans included a sufficient number of individual plants for measurement, but were located far enough to the interior to reduce the effects of fencing on plant growth. I temporarily ( $\leq 72$  hours) removed exclosures during herbicide application when necessary. I evaluated soybean crops near the estimated date of plant maturity before senescence. I recorded the number of rows and the number of plants within each subplot. I then measured plant height, number of bean pods per plant and classified the level of browsing of trifoliolate leaves per plant on 30 randomly selected plants in the subplot. I estimated grain yield (bushels per hectare) for fenced and unfenced subplots using a standard yield estimator. I consulted with the agricultural producer to determine the variety of soybeans planted in each field.

*Deer Abundance Estimates*—I used baited infrared camera surveys to estimate the abundance of deer in the area of crop fields to estimate harvest rates of deer on focal properties. This method of survey was conducted according to previous research by Jacobson et al. (1997). These researchers demonstrated that the abundance of deer in an area could be determined using baited surveys, where bucks could be uniquely identified by antler characteristics and their number used to infer the number of does and fawns visiting repeatedly a bait site. Cameras were placed at a density of 1 camera per 65 hectares in wooded or brushy habitat immediately adjacent to crop fields. This relatively high density of cameras was intended to reduce bias associated with capturing adult bucks at a higher rate at lower camera densities because males have larger home ranges (Jacobson et al. 1997). A bait site was established at each camera location during a 7-day pre-baiting period. During pre-baiting, whole kernel corn and trace mineral salts were placed at each bait site in a quantity sufficient to maintain consistent access by deer 24 hours per day. Following this acclimatization period, an infrared camera was set to record still photographs of deer 24 hours a day at 10-minute intervals during a 14-day survey period. As in the pre-baiting period, bait was provided ad libitum. I generated deer abundance estimates using data pooled from all cameras on a property according to the methods of Jacobson et al. (1997). Deer abundance estimates were conducted during August. This timing increased the likelihood that: 1) fawns were mobile with their dams and available for survey, 2) antler growth of bucks was sufficient to uniquely identify individuals, 3) deer photographed near crop fields were likely those that caused damage during the growing season and should have been available for harvest in the same area, and 4) harvest mortality and disturbance of deer by hunting activities was minimized since the survey preceded deer hunting seasons.

## **RESULTS AND DISCUSSION**

I conducted the pilot study from April-December 2013. My methods to evaluate crops were acceptable to agricultural producers, did not interfere with normal farming practices, and generated estimates of crop yields and crop losses attributed to deer that may be compared among properties. On the properties where I evaluated crops for the pilot study, agricultural producers did not have complaints about damage caused by deer. Correspondingly, my estimates of crop losses due to deer were low for corn (<10%, Table 1) and negligible for soybeans (Table 2). However, I speculate that focal properties where producers complain of deer damage would have higher deer densities and greater crop losses.

Preliminary results from the pilot study suggested that localized management increased harvest levels of deer on properties where agricultural damage was occurring (Table 3). Relative deer densities on focal properties were a minimum of two times higher than estimated deer densities for their respective DPAs (MNDNR, unpublished data). Likewise, the number of deer harvested per km<sup>2</sup> on focal properties was a minimum of two times higher than that reported for the respective DPAs where focal properties were located (McInenly 2013). An average of 26% of the deer estimated to be utilizing focal properties were harvested. These findings suggest that properties where deer damage is apparent to producers likely have an abundance of deer, which is elevated above management goals. Extra opportunities to harvest deer on these properties (i.e., localized management) should be afforded to elevate harvest levels concomitant with localized overabundance of deer.

This pilot study aided in the development of an efficient methodology for assessing damage caused by white-tailed deer to agricultural crops. Data related to management intensity on focal properties was important for planning the expansion of localized management in Minnesota. The pilot study provided the basis for a broader study which was initiated in spring 2014. A more comprehensive examination of crop losses relative to deer abundance will be conducted as part of the broader study.

## ACKNOWLEDGEMENTS

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**Table 1.** Yield estimates for field corn and estimated crop loss attributed to damage caused by white-tailed deer on 3 privately owned properties in southeast Minnesota, 2013.

Property	Bushels per ha		Mean crop loss
	Fenced	Unfenced	
A	572	523	8.5%
B	370	362	2.0%
C	531	478	9.9%

**Table 2.** Yield estimates for soybeans and estimated crop loss attributed to damage caused by white-tailed deer on 3 privately owned properties in southeast Minnesota, 2013.

Property	Bushels per ha		Mean crop loss <sup>1</sup>
	Fenced	Unfenced	
A	100	101	-1.0%
B	167	171	-2.7%
C	182	176	3.0%

<sup>1</sup>Negative values indicate higher average yield estimates in unfenced subplots versus subplots fenced to exclude deer.

**Table 3.** Estimates of the abundance of white-tailed deer and management intensity of deer on 4 privately owned properties in southeast Minnesota, 2013.

Property	Area (km <sup>2</sup> )	Estimated deer abundance	Relative deer density (deer per km <sup>2</sup> )	Total deer removed	Deer removed per km <sup>2</sup>	Management intensity <sup>1</sup>
D	0.9	80	89	12	13.3	15%
E	1.0	37	37	10	10.0	27%
F	2.5	106	42	43	17.2	41%
G	3.8	107	28	21	5.5	20%

<sup>1</sup>Proportion (%) of the number of deer estimated to be using a property that were harvested.



## EVALUATING PREFERENCES OF HUNTERS AND LANDOWNERS FOR MANAGING WHITE-TAILED DEER IN SOUTHWEST MINNESOTA<sup>1</sup>

Gino D'Angelo and Marrett Grund

### SUMMARY OF FINDINGS

During 2012, Minnesota Department of Natural Resources (MNDNR) conducted goal-setting process to gather public input to aid in setting white-tailed deer (*Odocoileus virginianus*) population goals for southwest Minnesota. The goal-setting process included development of recommendations for deer population goals by stakeholder teams and an online questionnaire of voluntary participants. The results of the goal-setting process were not clear, with 46% of respondents indicating that deer numbers were about right and 50% of respondents indicating that deer numbers were too low. With no major opinion about deer population levels in southwest Minnesota, the results of the goal-setting process were difficult to apply to management. In addition, only 36% of online respondents were satisfied with the goal-setting process. Thus, the purpose of our study was to obtain detailed public input data to aid in setting deer population goals for southwest Minnesota.

We mailed surveys to 3,600 hunters and 4,604 landowners in southwest Minnesota to evaluate their experiences and attitudes regarding white-tailed deer densities, hunting opportunities, and potential regulations for deer hunting (Figure 1). We received a total of 2,063 completed surveys from hunters for a response rate of 59.3%. We received a total of 2,105 completed surveys from landowners for a response rate of 47.8%.

Most respondents (98%) to the hunter survey participated in the 2012 deer hunting season. Fifty-two percent of landowners either deer hunted during 2012 (26%) or hunted in Minnesota in the past (26%). Hunters had an average of 23 years of experience hunting in Minnesota, whereas landowners hunted an average of 33 years in Minnesota. Twelve percent of hunters archery hunted, 98% firearm hunted, and 17% muzzleloader hunted. Almost nineteen percent of landowners who hunt participated in archery season, 90% firearm hunted, and 25% muzzleloader hunted. Most hunters (57%) and landowners who hunt (62%) would prefer to kill a mature buck versus female deer or smaller bucks.

One-third of respondents who hunted harvested a buck in southwest Minnesota in 2012. Most hunters ( $\geq 57\%$ ) were satisfied with the number of antlerless deer and the total number of deer seen while hunting. Likewise, most hunters ( $\geq 76\%$ ) felt that the number of either-sex permits provided was either too low or about right. Only about one-third of all hunters were satisfied with the number and quality of bucks.

The landowners we surveyed controlled relatively large landholdings, which were primarily used for row-crop agriculture. They represented approximately 50% of landowners with  $\geq 160$  acres in southwest Minnesota. Two-thirds of respondents had knowledge about wildlife damage to crops on their properties. Therefore, the opinions of landowners included in this survey should accurately describe those of agricultural producers in southwest Minnesota. Although 73% of landowners reported at least some damage due to deer, average total crop damage due to deer was \$4885. Considering the large-scale production by respondents, crop losses to deer damage were relatively minor. Also, most landowners recognized that other species caused damage to crops, and attributed an average of 35% of damage to deer. Sixty-nine percent of landowners believed that deer damage was less than or the same as 5 years ago, which suggests that perceptions about crop damage may not be influenced by increased commodity prices.

<sup>1</sup>For full report, please see: D'Angelo, G. J., and M. D. Grund. 2014. Evaluating preferences of hunters and landowners for managing white-tailed deer in southwest Minnesota. Division of Fish and Wildlife, Minnesota Department of Natural Resources, St. Paul, Minnesota. 101 pp.

Ninety percent of landowners allow family members to hunt on their property and 19% of landowners allow hunting by non-family. Other than family, most landowners will allow friends or neighbors to hunt (80%). Eighteen percent of landowners will allow strangers who ask permission to hunt, and few allow specific hunting groups (5%). The average number of people landowners allow to hunt their land was 5 individuals, which equates to about 1 hunter per 123 acres. Less than 1% of landowners stated that they do not allow people to hunt because they do not believe in hunting.

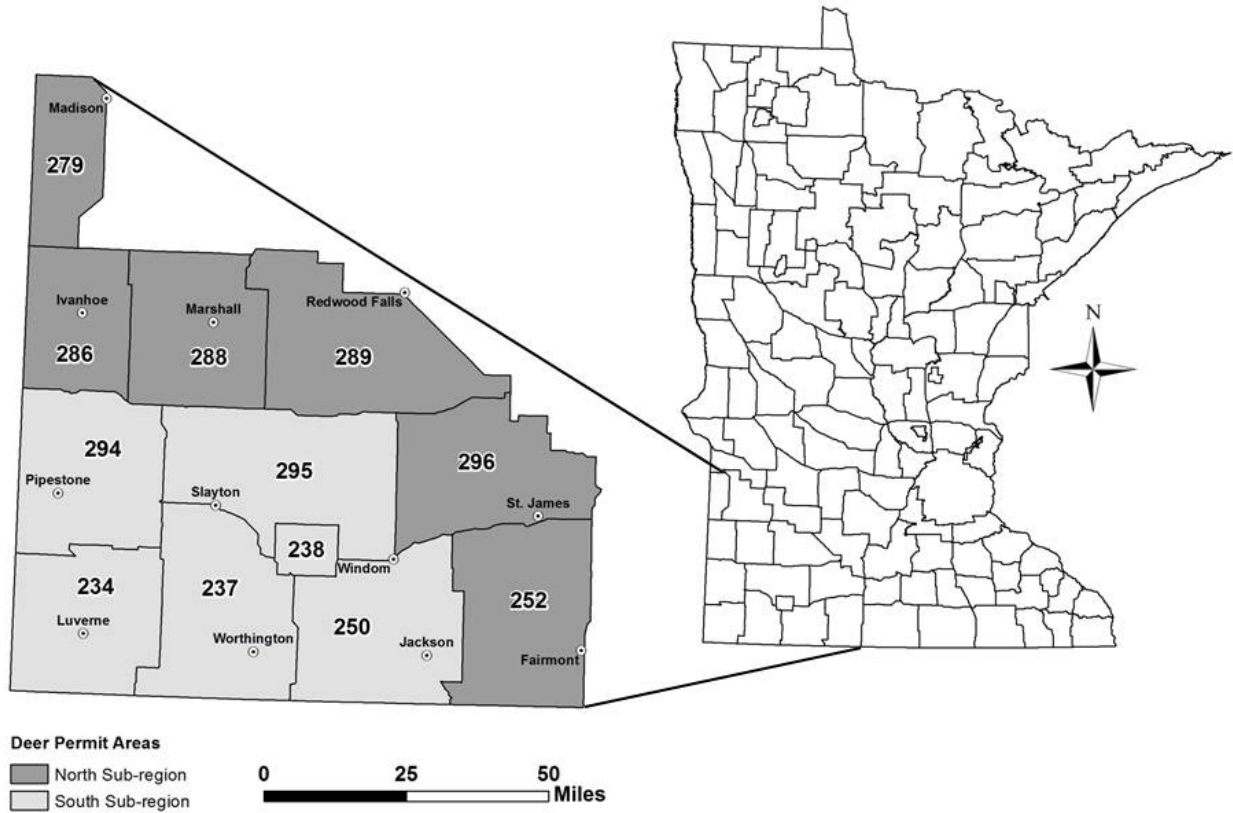
Only 36% of hunters and 18% of landowners felt that there were fewer deer in 2012 than 5 years ago in their area. Similar proportions of hunters (28%) and landowners (30%) believed there should be no change in the level of the deer population. However, the opinions of hunters trended toward increasing the deer population, while landowners tended to want the deer population decreased. Our results indicated that most hunters and landowners believed that there was an adequate number of deer in the population.

Landowners were less concerned about the specifics of deer harvest regulations than hunters. This is evident in their response rates, and answers which were distributed among categories of support or opposition with no clear majority opinion. Most hunters (59%) support a regulation to increase the proportion of antlered bucks in the population (Figure 2). Hunters were asked to rate their support for three potential regulations to reduce harvest pressure on bucks—buck permit lottery, antler point restriction, and a prohibition on cross-tagging of bucks. There was little support among hunters for a buck permit lottery (28%) or a prohibition on the cross-tagging of bucks (28%), however 50% of hunters supported an antler point restriction and 50% of hunters supported a youth-only deer season.

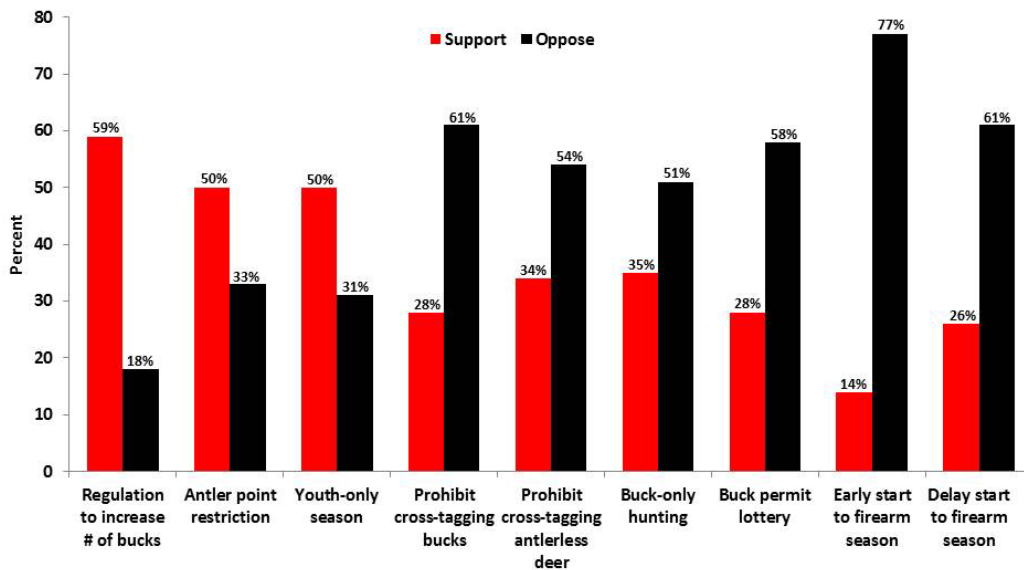
Only one-third of hunters supported buck-only hunting or a prohibition of cross-tagging antlerless deer. It is unlikely that these regulatory changes will be necessary since deer population levels are at or near goal levels and most hunters and landowners are satisfied with current deer numbers. Despite claims voiced by some hunters, there is little support for starting firearm season earlier (14%) or later (26%).

Our results indicated that that most hunters and landowners were satisfied with current deer numbers and believed the number of either-sex permits issued by the MNDNR has been appropriate. Although reports of damage due to deer were relatively minor, nearly one-half of landowners wanted deer densities reduced. Although hunters tend to prefer higher deer densities to maximize recreational opportunities, 20% of hunters wanted deer densities reduced. Thus, current deer densities should be considered to be near maximum levels to be acceptable to all stakeholders. Special opportunities to harvest deer should be afforded to landowners when non-lethal measures are not sufficient to minimize damage.

About two-thirds of the hunters we surveyed were not satisfied with the number or quality of bucks in the southwest Minnesota deer population. As demonstrated in southeast Minnesota and in other states, an antler-point restriction regulation reduces harvest mortality rates of young bucks thereby allowing bucks to reach older-age classes and grow larger racks. Previous hunter surveys conducted in Minnesota suggested that buck harvest mortality would slightly decrease if hunters were not able to cross-tag bucks with their hunting licenses. Our results suggest that 50% of hunters support an antler-point restriction regulation but there was strong opposition from hunters about prohibiting the cross-tagging of deer. Based on these findings, we believe wildlife managers should consider implementing an antler-point restriction to address satisfaction levels associated with the quantity and quality of bucks in southwest Minnesota deer populations.



**Figure 1.** Location of deer permit areas in southwest Minnesota where Minnesota Department of Natural Resources surveyed hunters and landowners during 2013 to evaluate their preferences for managing white-tailed deer in the region.



**Figure 2.** Support of hunters for regulatory changes to white-tailed deer hunting in southwest Minnesota, 2012.



## **ESTABLISHMENT OF FORBS IN EXISTING GRASS STANDS DOMINATED BY WARM-SEASON GRASSES**

Nicole Davros, Molly Tranel Nelson, Kurt Haroldson, and Véronique St-Louis

### **SUMMARY OF FINDINGS**

Interseeding native forbs into reconstructed grasslands dominated by warm-season grasses could restore plant species diversity and improve wildlife habitat, yet many managers report having limited experience with interseeding and poor success with a few early attempts. Survival of forbs interseeded directly into existing vegetation may be enhanced by management treatments that reduce competition from established warm-season grasses. In 2009, we initiated a field experiment to investigate the effects of two mowing and two herbicide treatments on diversity and abundance of forbs interseeded into established grasslands on 15 sites across southern Minnesota. Each site was burned and interseeded in fall 2009 (n=8) or spring 2010 (n=7), and two mowing treatments (Mow 1, Mow 2) and two grass-selective herbicide treatments (Low Herbicide, High Herbicide) were applied during the 2010 growing season. Sites were surveyed during summer 2011, burned during spring 2013, and surveyed again during summer 2013. We also completed a cost analysis to determine the cost per acre of 3 management options (interseeding + mowing twice, interseeding + herbicide spraying at higher rate, cropping + new planting). We observed 24 (83%) of the 29 native seeded forbs in study plots each year. Species richness of seeded forbs was marginally greater in the high herbicide treatment than the control in 2011. Additionally, total species richness and total plant community diversity were greater in the high herbicide and mow 2 treatments than the control in 2011. However, these treatment differences disappeared by 2013. Overall, seeded forb species richness, total species richness, and total plant community diversity were lower in 2013 than 2011. None of the treatments were more effective than the control in helping to increase the percent canopy cover of native forbs over time, and warm season grasses continued to dominate canopy cover 3 years post-treatment. Our cost analysis indicated that interseeding plus mowing (\$296/acre) or herbicide spraying at the higher rate (\$342/acre) were cheaper options than eliminating existing vegetation and planting entirely new seed (\$450/acre). However, the results from our field experiment indicate that neither mowing nor spraying a grass-selective herbicide during the first growing season post-interseeding are effective ways to establish forbs in grasslands dominated by warm-season grasses. Natural resource managers may obtain improved vegetation structure and diversity by spending the additional money to completely eliminate the existing vegetation and then re-planting a higher diversity seed mix into bare ground.

### **INTRODUCTION**

Minnesota Department of Natural Resources (MNDNR) wildlife managers indicated a need for more information on establishing and maintaining an abundance and diversity of forbs in reconstructed grasslands (Tranel 2007). A diversity of forbs in grasslands provides the heterogeneous vegetation structure needed by many bird species for nesting and brood rearing

(Volkert 1992, Sample and Mossman 1997). Forbs also provide habitat for pollinators and other invertebrates, essential foods for breeding grassland birds and their broods (Buchanan et al. 2006).

The forb component in many restored grasslands has been lost or greatly reduced. Managers interested in increasing the diversity and quality of forb-deficient grasslands are faced with the costly option of completely eliminating the existing vegetation and planting into bare ground, or attempting to interseed forbs directly into existing vegetation. Management techniques that reduce competition from established grasses may provide an opportunity for forbs to become established in existing grasslands (Collins et al. 1998, McCain et al. 2010). Temporarily suppressing dominant grasses may increase light, moisture, and nutrient availability to seedling forbs, ultimately increasing forb abundance and diversity (Schmitt-McCain 2008, McCain et al. 2010). Williams et al. (2007) found that frequent mowing of grasslands in the first growing season after interseeding increased forb emergence and reduced forb mortality. Additionally, Hitchmough and Paraskevopoulou (2008) found that forb density, biomass, and richness were greater in meadows where a grass herbicide was used.

In this study, we examined the effects of two mowing and two herbicide treatments on diversity and abundance of forbs interseeded into established grasslands dominated by warm-season grasses in southern Minnesota. Further, we calculated costs associated with two management approaches for comparison with the cost of a re-planting option. Our results can help guide future management decisions made by wildlife managers.

## **METHODS**

### **Field Experiment**

*Study Site Selection.* – We selected study sites (n=15) throughout the southern portion of Minnesota's prairie/farmland region on state- and federally-owned wildlife areas. Each site was  $\geq 4$  ha and characterized by relatively uniform soils, hydrology, and vegetative composition. All sites were dominated by relatively uniform stands of warm-season grasses with few forbs, most of which were non-native species [e.g., sweet clover (*Melilotus alba*, *M. officinalis*)]. Dominant grasses included big bluestem (*Andropogon gerardii*), switchgrass (*Panicum virgatum*), and Canada wild rye (*Elymus canadensis*).

*Site Preparation and Interseeding.* – Eight sites were burned in October and November 2009 and frost interseeded during December 2009 and March 2010, whereas 7 sites were burned and interseeded during April and May 2010. The same 30-species mix of seed was broadcast seeded at all sites at a rate of 239 pure live seeds/m<sup>2</sup> (Table 1). Seed used on spring-burned sites was cold-moist stratified for 3-5 weeks in wet sand to stimulate germination prior to interseeding; seed used on fall-burned sites was not cold-moist stratified prior to interseeding.

*Treatments.* – We divided sites into 10 study plots of approximately equal size and randomly assigned each of 4 treatments and the control. Each site received all treatments to account for variability among sites, and the control and each treatment were replicated twice at each site. The following treatments, designed to suppress grass competition, were applied during the first growing season after interseeding (2010) while the forbs were becoming established:

- Mow 1: mowed once to a height of 10-15 cm when vegetation reached 25-35 cm in height.

- Mow 2: mowed twice to a height of 10-15 cm when vegetation reached 25-35 cm in height.
- Low Herbicide: applied grass herbicide Clethodim (Select Max®) at 108 mL/ha (9 oz/A) when vegetation reached 10-15 cm.
- High Herbicide: applied grass herbicide Clethodim (Select Max®) at 215 mL/ha (18 oz/A) when vegetation reached 10-15 cm.

*Sampling Methods.* – We visited all sites once annually between 25 July – 27 September in 2011 and between 22 July and 15 August 2013. Twenty points within each study plot were randomly chosen for sampling. We estimated presence of all plant species in a 76 x 31 cm<sup>2</sup> quadrat at each sampling point. In addition, we estimated litter depth and percent cover (Daubenmire 1959) of native grasses, exotic grasses, native forbs, exotic forbs, bare ground, and duff within each sampling quadrat. We estimated percent cover within 6 classes: 0-5%, 5-25%, 25-50%, 50-75%, 75-95%, and 95-100%. Finally, we recorded visual obstruction readings (VOR; Robel et al. 1970) in the 4 cardinal directions at the 5<sup>th</sup>, 10<sup>th</sup>, 15<sup>th</sup>, and 20<sup>th</sup> quadrats in each plot to determine vegetation vertical density.

Deviations from the field protocol occurred in 2012 in the following ways: 1) Only 10 of the 15 sites were visited; 2) Several flags and markers disappeared or fell down between seasons, and plot corners were not remarked or reflagged prior to the start of data collection. As a result, plot boundaries could not be reliably determined; 3) The start of data collection was delayed. All data was collected between 28 August – 23 September 2012, a period of drought in southern Minnesota that could have affected species detection rates; 4) Robel pole readings were only taken at 7 of the 10 sites. Due to these deviations from the original protocol, we have not included the 2012 data in our analyses.

*Post-Treatment Management.* – To aid forb establishment and persistence, managers conducted prescribed burns at 14 sites during April and May 2013. Due to time and weather limitations, 1 site was not burned.

*Statistical Analyses.* – We used mixed models to evaluate the effects of treatments, years, and the treatment x year interaction on our estimates of canopy cover, vegetation vertical density, species richness, and diversity. We nested plots within site as a random effect. We did not correct for detection probability in our analyses of richness and diversity measures. We defined species richness as the average number of species present in a sampling quadrat, and we estimated two measures of species richness: interseeded forbs only and total species richness. We calculated the Shannon-Wiener index of diversity,  $H = -\sum P_i(\ln P_i)$ , where  $P_i$  is the proportion of each species in the sample. This diversity index combines measures of species richness and evenness, and a greater value indicates a more diverse suite of species are present. We estimated two different measures of diversity – interseeded forb species diversity and overall plant community diversity. We report untransformed data throughout the text.

## **Cost Analysis**

During winter 2012, we obtained work summaries from the area wildlife offices at Talcot Lake and Slayton. The work summaries contained detailed notes on the number of personnel, the type(s) of fleet vehicles and equipment, and the number of man hours spent prepping, burning, and treating (i.e., mowing or herbicide spraying) the sites used for our study which allowed us to estimate the cost of 3 particular management activities: 1) interseeding + mowing



twice, 2) interseeding + herbicide spraying at higher rate, 3) cropping + new planting. Some variables (e.g., fleet costs, number of personnel needed for a prescribed burn) varied widely in cost depending on the distance to or the size of the site so we calculated average costs based on a range of site sizes and distances traveled. Additionally, managers provided estimates for activities and supplies not included in the work summaries (e.g., acres/hour of planting into recently cropped soil). We used seed prices from a current MNDNR state contract vendor (Shooting Star) to determine the cost of our 30-species interseed mix versus the typical mesic southwest prairie seed mix used by MNDNR wildlife managers when they seed into bare soil. Finally, managers often use cooperative farming agreements (CFAs) to accomplish certain activities (e.g., mowing, row cropping to prep soil) on state land. The CFAs are essentially a barter system between the state and a private cooperator in which work is done at no cost to the state. We accounted for CFAs where appropriate in our cost analysis.

## RESULTS

### Field Experiment

In 2011 (i.e., one year post-treatment), we observed 24 (82.7%) of the 29 interseeded species across all study plots and detected 1718 individual interseeded forb plants. Black-eyed Susan (*Rudbeckia hirta*) was the most common seeded forb species (forming 40.2% of all seeded forb observations), followed by wild bergamot (*Monarda fistulosa*, 16.5%), golden Alexander (*Zizia aurea*, 10.1%), common milkweed (*Asclepias syriaca*, 8.0%), and yellow coneflower (*Ratibida pinnata*, 7.2%). In 2013, we again observed 24 (82.7%) of the 29 seeded forb species across all study plots but had fewer overall detections (n = 520). Black-eyed Susan (21.5%), wild bergamot (18.5%), common milkweed (15.2%), Canada milk vetch (*Astragalus canadensis*, 8.1%), and golden Alexander (6.7%) were the most common seeded forb species in 2013.

Native grasses formed the greatest component of canopy cover (48.1%) across both years (Fig. 1). Canopy cover of native grasses was lowest in the Mow 2 treatment compared to the other treatments and control in 2011 (Table 2), but there were no overall treatment effects. Canopy cover of native forbs, including interseeded species, averaged 22.3% across all treatments and the control in 2011 but decreased to 9.5% by 2013 (Fig. 2). Canopy cover of native forbs was slightly greater in the High Herbicide treatment in both years (Table 2); however, none of the treatments significantly affected the cover of native forbs compared to the control. Exotic grasses and exotic forbs also showed reduced canopy cover over time (Table 2) but there were no differences among the treatments and control.

The control plots had slightly greater vegetation vertical density than the treatment plots in 2011 (Table 2; Fig. 3) but these differences were not significant. Vertical density was significantly less in 2013 compared to 2011.

Species richness of interseeded forbs was higher in the High Herbicide treatment plots compared to the controls in 2011, but these differences did not persist in 2013 (Table 2; Fig. 4). Total species richness was also greater in the High Herbicide treatment plots than the control in 2011 (Table 2; Fig. 5), but total species richness declined through time.

The diversity of interseeded forbs was greater in 2011 than 2013 but did not vary significantly among the treatments and controls (Fig. 6). Overall, interseeded forb diversity was slightly greater in the High Herbicide and Mow 2 treatments in 2011 (Table 2). High Herbicide

and Mow 2 treatments had greater overall plant community diversity than the control in 2011 but these differences were no longer evident in 2013 (Table 2; Fig. 7).

### **Cost Analysis**

“Interseeding + Mowing Twice” was the cheapest management option (\$296/acre) available to wildlife managers for attempting to increase diversity on a WMA followed by the “Interseeding + High Herbicide” option (\$342/acre)(Table 3). In both scenarios, the cost of the forb-only seed mix helped make these options more affordable. Although forb seeds are generally more expensive than grass seeds, fewer forb seeds are needed for an interseeding approach. The “Crop + Re-Plant” option was the most expensive option (\$450/acre), and an entirely new seed mix, including grasses and forbs, drove the price of this option (Table 3).

### **DISCUSSION**

Mowing and herbicide treatments were effective in suppressing grasses during the first growing season after application during a pilot study (Tranel 2009). Here, we also found that the High Herbicide treatment had some effect on increasing the percent canopy cover of native forbs, species richness, and plant diversity during the first growing season (2011) after treatment. The Mow 2 treatment helped reduce the percent canopy cover of native grasses and increase plant diversity in 2011. However, each of these treatment effects were weak and none persisted into 2013. Although we found that most of the seeded forb species (82.7%) were established in relatively low numbers in 2011, their numbers had dropped by 2013 whereas the percent canopy cover of native grasses remained dominant each year. Thus, the mowing and herbicide treatments were not effective at suppressing warm-season native grasses over the long term. Other studies have reported better success with persistence of forbs over time. Williams et al. (2007) observed similarly abundant seeded forbs in mowed and control treatments at the end of a second growing season, but seeded forbs were twice as abundant in mowed treatments by the beginning of year 5. Hitchmough and Paraskevopoulou (2008) found that, in treatments where grass was suppressed with a graminoid herbicide, sown forb density was higher in the second and third year after treatment and forb richness was greater 3 years after treatment.

The goal of our study was to test potential management options that would be applicable for grassland restoration across a broad spectrum of sites. Despite choosing study plots with relatively uniform vegetation (i.e., dominated by warm-season native grasses), our sites still had a high degree of among-site variability. Factors beyond vegetation type (e.g., soil moisture, nutrients, and microbes) undoubtedly play a role in determining plant competition and the success or failure of seed establishment (e.g., Grygiel et al. 2012, Rossiter 2013). Thus, management options that are suitable for one particular site may not work well at another site. As a result, broadly applicable management options for increasing diversity through interseeding may not be a viable option.

Our cost analysis indicated that starting over on a WMA planting (i.e., tilling the vegetation under, cropping, then re-planting with a new seed mix) is the most costly option for grassland restoration considered in this study. However, MNDNR wildlife managers report having good long-term success with increasing forb canopy cover and plant diversity when they use this option (J. Beech, personal communication). Managers may be able to increase the

effectiveness of interseeding by giving more attention to a site (e.g., mowing more than twice within a season, mowing across multiple seasons, spraying a grass-selective herbicide more often), but such activities would only add to the cost of interseeding-based restoration. Therefore, starting over may be the most costly option in the short-term but it is likely the cheapest option in the long-term.

## **MANAGEMENT IMPLICATIONS**

The use of the pre-emergent grass selective herbicide Clethodim (Select Max®) at 215 mL/ha (18 oz/A) had limited effectiveness at helping forbs establish in sites dominated warm-season native grasses during the first growing season post-application. Growth of grass was stunted but grass mortality was not observed even at the high application rate at any of the study sites. However, forbs did not persist over time and native grasses continued to dominate regardless of treatment. We suggest that starting a restoration over may be the more expensive option in the short-term, but it is likely to provide better results (i.e., increased forb cover and plant community diversity) over the longer term when compared to mowing or herbicide-spraying options.

## **ACKNOWLEDGMENTS**

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Table 1. Forb species mix and seeding rates used in this study. Fifteen study sites across southern Minnesota were prepped (mowed and burned) then interseeded during fall 2009 or early spring 2010.

Common Name	Scientific Name (Family)	Family	Oz/Acre	Seed/ft <sup>2</sup>	Seeds/Oz	Seeds/Acre	% of Mix
Leadplant	<i>Amorpha canescens</i>	Fabaceae	0.50	0.18	16,000	8,000	0.79
Black-eyed Susan	<i>Rudbeckia hirta</i>	Asteraceae	1.20	2.53	92,000	110,400	10.92
Maximilian Sunflower	<i>Helianthus maximilianii</i>	Asteraceae	0.50	0.15	13,000	6,500	0.64
Yellow Coneflower	<i>Ratibida pinnata</i>	Asteraceae	1.35	0.93	30,000	40,500	4.01
Golden Alexanders	<i>Zizia aurea</i>	Apiaceae	1.00	0.25	11,000	11,000	1.09
Sky Blue Aster	<i>Aster oolentangiensis</i>	Asteraceae	0.85	1.56	80,000	68,000	6.73
Canada Milk Vetch	<i>Astragalus canadensis</i>	Fabaceae	1.75	0.68	17,000	29,750	2.94
Prairie Cinquefoil	<i>Potentilla arguta</i>	Rosaceae	0.85	4.49	230,000	195,500	19.35
White Prairie Clover	<i>Dalea candida</i>	Fabaceae	1.50	0.65	19,000	28,500	2.82
Purple Prairie Clover	<i>Dalea purpurea</i>	Fabaceae	1.50	0.52	15,000	22,500	2.23
False Sunflower	<i>Heliopsis helianthoides</i>	Asteraceae	1.25	0.18	6,300	7,875	0.78
Alumroot	<i>Heuchera richardsonii</i>	Saxifragaceae	0.05	0.80	700,000	35,000	3.46
N. L. Purple Coneflower	<i>Echinacea angustifolia</i>	Asteraceae	0.85	0.14	7,000	5,950	0.59
Virginia Mountain Mint	<i>Pycnanthemum virginianum</i>	Lamiaceae	0.20	1.01	220,000	44,000	4.35
Common Milkweed	<i>Asclepias syriaca</i>	Asclepiadaeaceae	1.00	0.09	4,000	4,000	0.40
Blue Vervain	<i>Verbena hastata</i>	Verbenaceae	0.75	1.60	93,000	69,750	6.90
Rough Blazingstar	<i>Liatris aspera</i>	Asteraceae	0.15	0.06	16,000	2,400	0.24
New England Aster	<i>Aster novae-angliae</i>	Asteraceae	0.65	0.98	66,000	42,900	4.25
Prairie Onion	<i>Allium stellatum</i>	Liliaceae	0.70	0.18	11,000	7,700	0.76
Hoary Vervain	<i>Verbena stricta</i>	Verbenaceae	0.65	0.42	28,000	18,200	1.80
Heath Aster	<i>Aster ericoides</i>	Asteraceae	0.15	0.69	200,000	30,000	2.97
Stiff Goldenrod	<i>Oligoneuron rigidum</i>	Asteraceae	0.75	0.71	41,000	30,750	3.04
Culver's Root	<i>Veronicastrum virginicum</i>	Scrophulariaceae	0.10	1.84	800,000	80,000	7.92
Showy Tick Trefoil	<i>Desmodium canadense</i>	Fabaceae	0.85	0.11	5,500	4,675	0.46
Wild Bergamot	<i>Monarda fistulosa</i>	Lamiaceae	0.70	1.12	70,000	49,000	4.85
Prairie Coreopsis	<i>Coreopsis palmata</i>	Asteraceae	0.25	0.06	10,000	2,500	0.25
Partridge Pea	<i>Chamaechrista fasciculata</i>	Caesalpiniaceae	2.00	0.12	2,700	5,400	0.53
Closed Bottle Gentain	<i>Gentiana andrewsii</i>	Gentianaceae	0.08	0.51	280,000	22,400	2.22
Heart Leaf Golden Alexander	<i>Zizia aptera</i>	Apiaceae	0.20	0.06	12,000	2,400	0.24
Brown Fox Sedge	<i>Carex vulpinoidea</i>		0.25	0.57	100,000	25,000	2.47

Table 2. Comparison of estimated (means  $\pm$  SE) percent canopy cover (native grass, exotic grass, native forbs, exotic forbs), vegetation vertical density (visual obstruction reading), species richness (interseeded native forbs only, all species), and species diversity (interseeded native forbs only, all species) on 15 study sites across southern Minnesota during summer 2011 and 2013. Sites were prepped (mowed and burned) then interseeded during fall 2009 or early spring 2010 with a 30-species seed mix. Treatments were applied during the 2010 growing season.

	Control		High Herbicide		Low Herbicide		Mow 1		Mow 2	
	2011	2013	2011	2013	2011	2013	2011	2013	2011	2013
% Canopy cover										
Native grasses	48.7 $\pm$ 3.63	47.1 $\pm$ 1.89	48.1 $\pm$ 3.40	46.1 $\pm$ 2.69	47.6 $\pm$ 3.37	46.8 $\pm$ 2.39	50.5 $\pm$ 3.55	51.3 $\pm$ 2.27	45.6 $\pm$ 3.80	49.0 $\pm$ 2.86
Exotic grasses	31.2 $\pm$ 4.79	26.7 $\pm$ 2.66	31.1 $\pm$ 4.89	24.8 $\pm$ 3.09	36.4 $\pm$ 5.16	24.9 $\pm$ 2.97	33.2 $\pm$ 4.50	25.6 $\pm$ 2.50	39.4 $\pm$ 4.72	27.5 $\pm$ 3.58
Native forbs	21.6 $\pm$ 4.31	7.9 $\pm$ 1.38	25.0 $\pm$ 4.16	10.5 $\pm$ 2.04	22.4 $\pm$ 4.45	10.4 $\pm$ 2.29	21.5 $\pm$ 4.31	8.9 $\pm$ 1.51	21.3 $\pm$ 4.13	9.6 $\pm$ 1.79
Exotic forbs	21.2 $\pm$ 3.49	16.3 $\pm$ 2.43	18.2 $\pm$ 2.54	15.4 $\pm$ 2.64	18.4 $\pm$ 3.23	15.1 $\pm$ 2.54	19.3 $\pm$ 2.24	11.5 $\pm$ 1.93	20.8 $\pm$ 2.89	15.3 $\pm$ 2.21
Vertical density	6.0 $\pm$ 0.59	4.4 $\pm$ 0.33	5.7 $\pm$ 0.43	4.2 $\pm$ 0.28	5.4 $\pm$ 0.49	4.0 $\pm$ 0.29	5.5 $\pm$ 0.53	4.7 $\pm$ 0.44	5.5 $\pm$ 0.56	4.5 $\pm$ 0.39
Species richness										
Interseeded forbs	3.7 $\pm$ 0.37	3.6 $\pm$ 0.31	4.6 $\pm$ 0.42	3.5 $\pm$ 0.42	3.8 $\pm$ 0.33	3.6 $\pm$ 0.35	3.9 $\pm$ 0.36	3.2 $\pm$ 0.35	4.4 $\pm$ 0.36	3.5 $\pm$ 0.29
All species	19.4 $\pm$ 1.12	16.0 $\pm$ 0.63	22.0 $\pm$ 1.34	15.8 $\pm$ 0.57	21.0 $\pm$ 1.14	15.9 $\pm$ 0.69	21.4 $\pm$ 1.27	15.3 $\pm$ 0.72	22.0 $\pm$ 1.34	16.2 $\pm$ 0.65
Species diversity										
Interseeded forbs	0.9 $\pm$ 0.08	0.8 $\pm$ 0.08	1.1 $\pm$ 0.10	0.9 $\pm$ 0.10	0.9 $\pm$ 0.08	0.8 $\pm$ 0.08	0.9 $\pm$ 0.08	0.8 $\pm$ 0.08	1.0 $\pm$ 0.09	0.8 $\pm$ 0.06
Entire community	4.4 $\pm$ 0.27	3.5 $\pm$ 0.18	5.2 $\pm$ 0.36	3.6 $\pm$ 0.16	4.9 $\pm$ 0.29	3.6 $\pm$ 0.19	5.0 $\pm$ 0.33	3.5 $\pm$ 0.20	5.1 $\pm$ 0.19	3.6 $\pm$ 0.19

Table 3. Results of a cost analysis comparing 3 management options aimed at increasing forb diversity on Minnesota Department of Natural Resources (MNDNR) Wildlife Management Areas (WMAs). Costs are calculated as \$/acre. Cooperative farming agreements (CFA) are often used by wildlife managers for certain management activities and occur at no cost to the state. Not all activities need to occur under each management situation (denoted with a period).

Management Activity	Management Option		
	Interseed + Mow Twice	Interseed + High Herbicide	Crop + Re-Plant
Site preparation			
Spray with Round-up herbicide	.	.	\$10
Haying	CFA	CFA	.
Mow firebreaks and conduct fall prescribed burn	\$32	\$32	\$32
Frost interseed (broadcast spread) - labor only	\$30	\$30	\$30
Cost of seed mix	\$202	\$202	\$336
Management treatment			
Crop for 2 years	.	.	CFA
Mow first time	CFA	.	.
Mow second time	CFA	.	.
Spray grass-selective herbicide (labor and herbicide costs)	.	\$46	.
Spot spray for weeds	.	.	\$10
Future site management			
Mow firebreaks and conduct prescribed burn	\$32	\$32	\$32
Total cost/acre	\$296	\$342	\$450

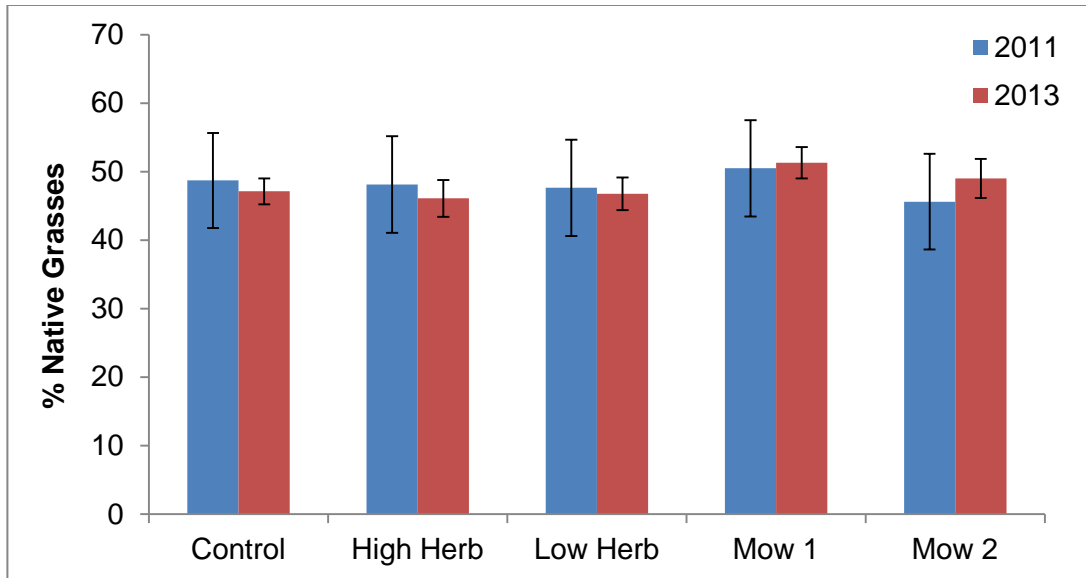


Figure 1. Estimated canopy cover of native grasses in treatment and control plots during summer 2011 and 2013. Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010, and treatments were applied during the 2010 growing season. Warm-season grasses, especially big bluestem (*Andropogon gerardii*), switchgrass (*Panicum virgatum*), and Canada wild rye (*Elymus canadensis*) dominated this category. Untransformed means and SEs are presented.

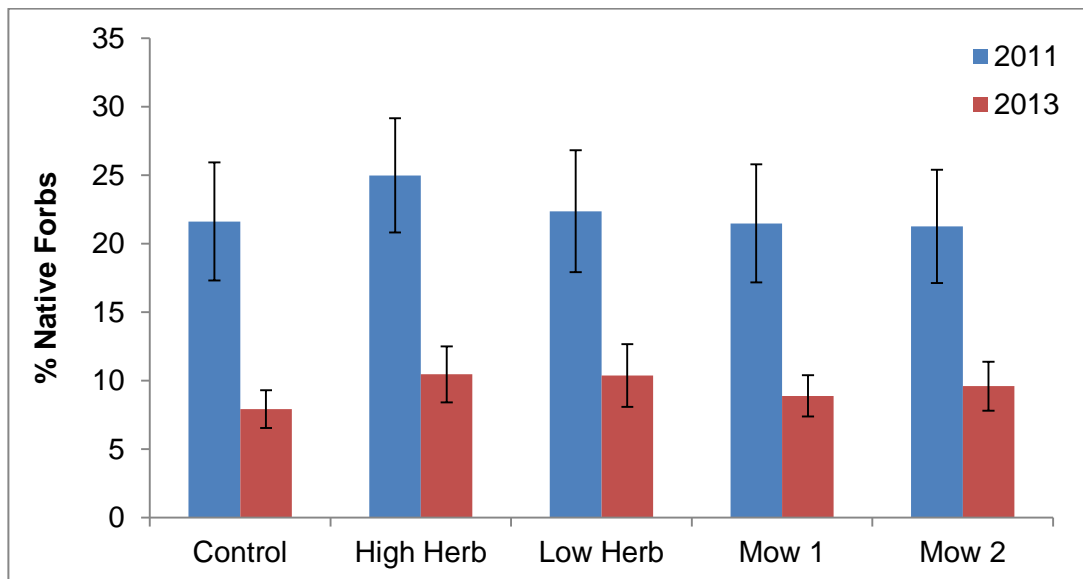


Figure 2. Estimated canopy cover of native forbs, including interseeded species, in treatment and control plots during summer 2011 and 2013. Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010 and treatments were applied during the 2010 growing season. Untransformed means and SEs are presented.



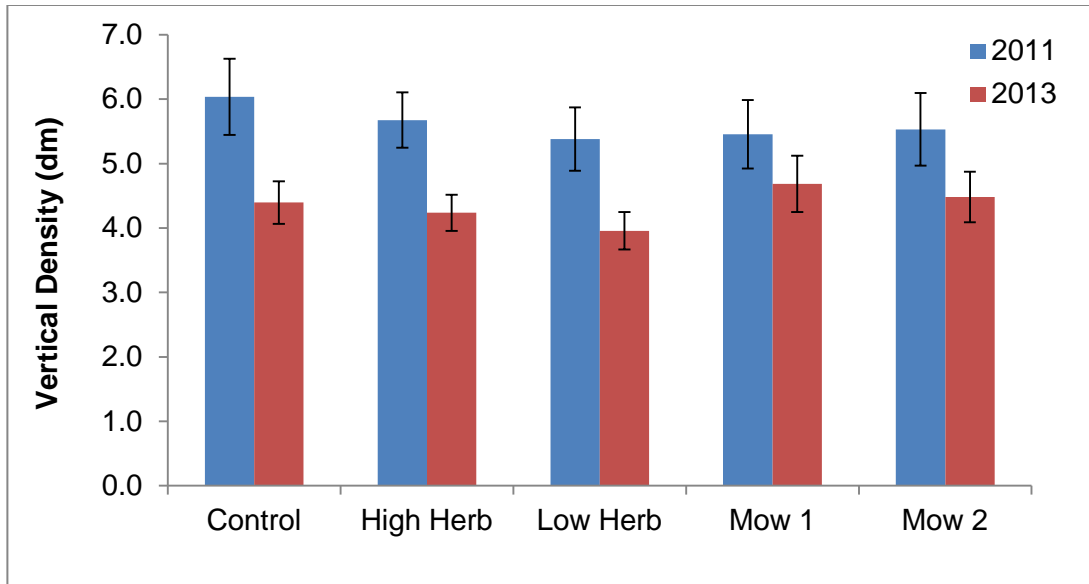


Figure 3. Vegetation vertical density (i.e., visual obstruction readings; Robel et al. 1970) in treatment and control plots during summer 2011 and 2013. Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010 and treatments were applied during the 2010 growing season. Untransformed means and SEs are presented.

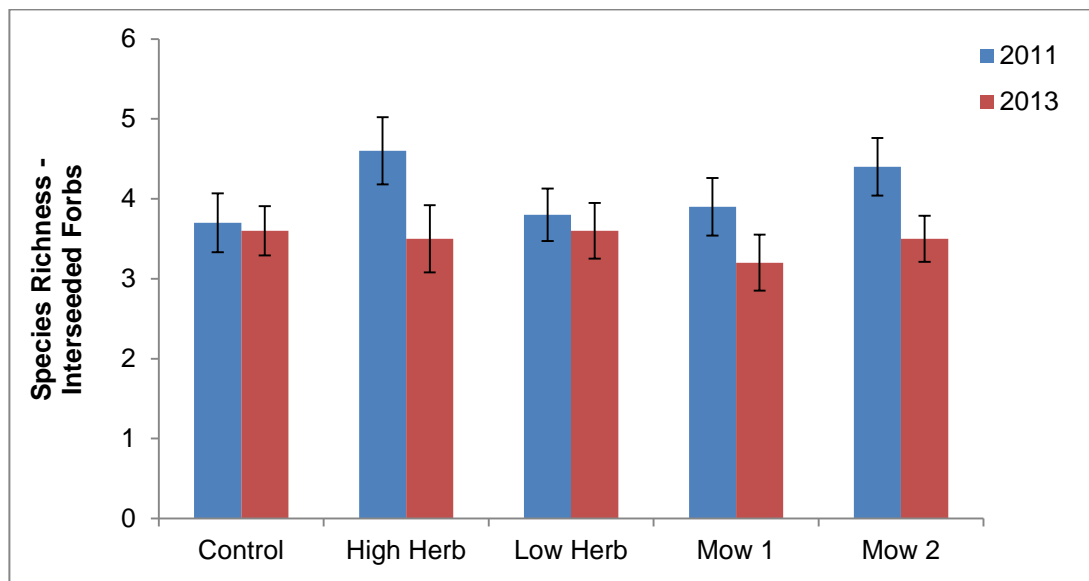


Figure 4. Species richness of interseeded native forbs in treatment and control plots during summer 2011 and 2013. Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010 and treatments were applied during the 2010 growing season. Untransformed means and SEs are presented.

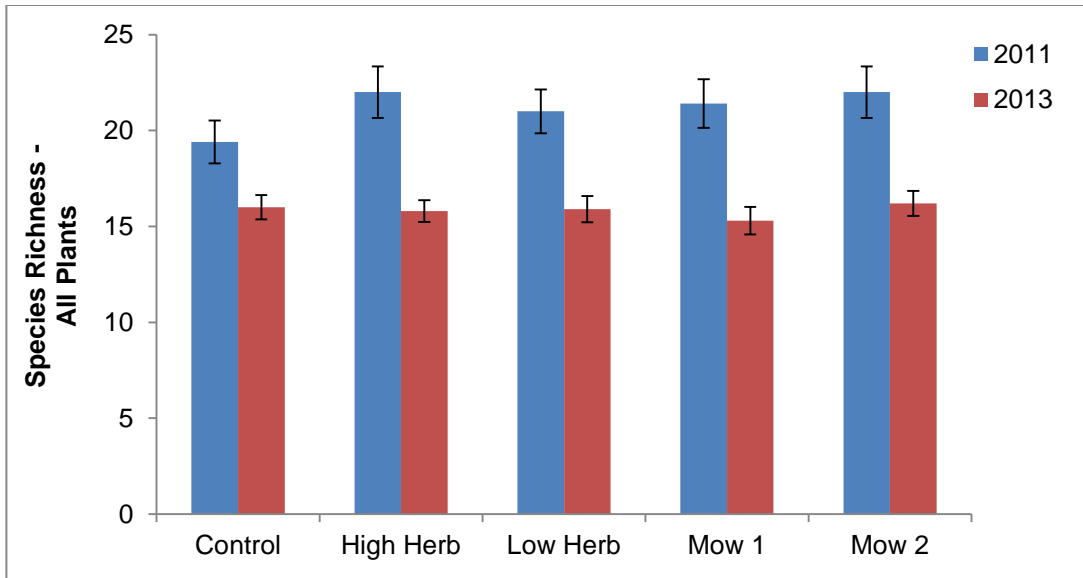


Figure 5. Total species richness (all species) in treatment and control plots during summer 2011 and 2013. Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010 and treatments were applied during the 2010 growing season. Untransformed means and SEs are presented.

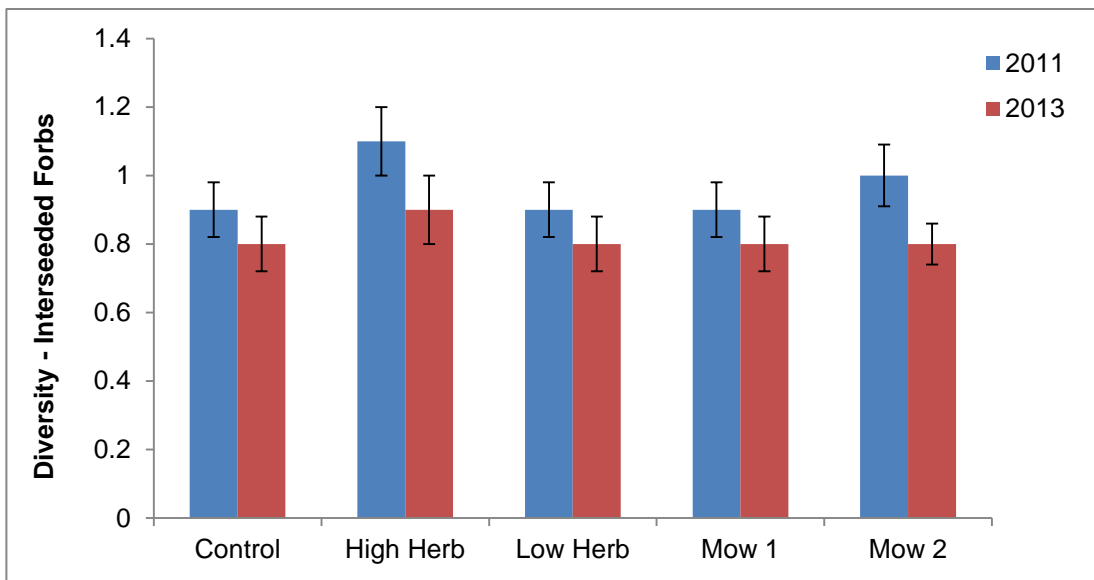


Figure 6. Diversity of interseeded native forbs in treatment and control plots during summer 2011 and 2013. The Shannon-Wiener Index was used to estimate diversity (see text for details). Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010 and treatments were applied during the 2010 growing season. Untransformed means and SEs are presented.

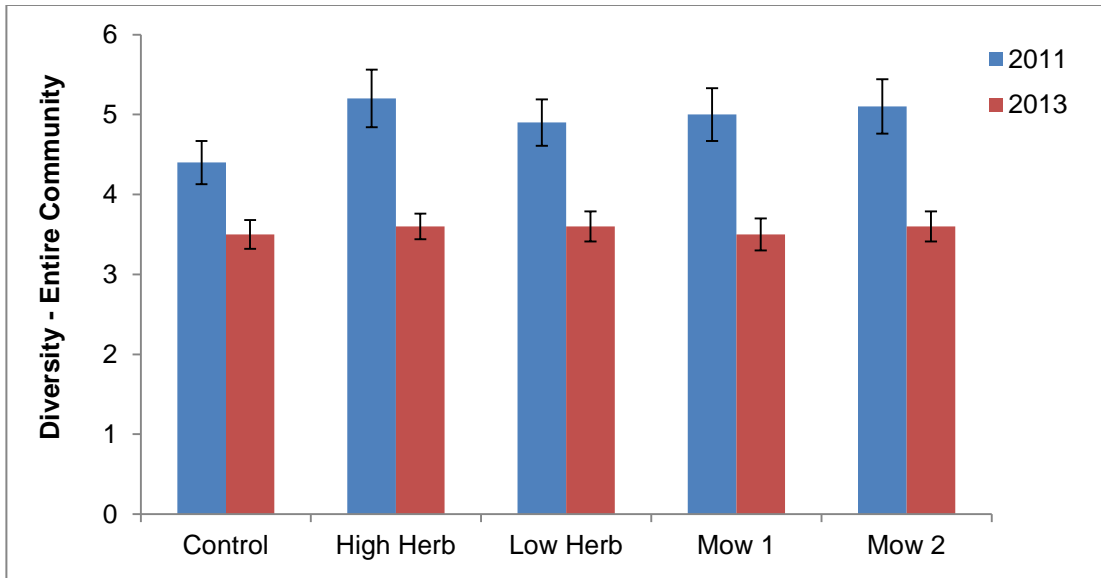


Figure 7. Diversity of the entire plant community in treatment and control plots during summer 2011 and 2013. The Shannon-Wiener Index was used to estimate diversity (see text for details). Plots were interseeded with a 30-species seed mix (29 forbs, 1 sedge) in 2009-2010 and treatments were applied during the 2010 growing season. Untransformed means and SEs are presented.

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## DETERMINING AN EFFECTIVE APPROACH FOR CAPTURING NEWBORN MOOSE CALVES AND MINIMIZING CAPTURE-RELATED ABANDONMENT IN NORTHEASTERN MINNESOTA

Glenn D. DelGiudice, William J. Severud<sup>1</sup>, Tyler R. Obermoller, Katie J. Foshay, and Robert G. Wright<sup>2</sup>

### SUMMARY OF FINDINGS

The goal of our recently initiated moose (*Alces alces*) calf research in northeastern Minnesota is to enhance our understanding of calf survival and specific causes of mortality and to assess their quantitative impact on the population's performance. The instantaneous probability of death (hazard) for northern ungulates is highest at birth, and although it declines sharply during the first 12 months, it is markedly higher than during the subsequent prime years of life. Fulfilling the primary goal of the calf study requires 3 things, the ability to: 1) capture and Global Positioning System (GPS)-collar a sample of newborn moose calves representative of the population, 2) closely monitor their movements and survival, and 3) conduct rapid investigative responses to calf mortalities. Unexpectedly, 2013's capture operations resulted in a relatively high number of neonates being abandoned within 48 hours post-capture (DelGiudice et al. 2013). This was unacceptable and prompted our team to examine closely all of the associated data collected to better understand capture-related abandonment and adopt a new approach for this year (2014) which would limit this undesirable side-effect of capture operations. We expected hourly fixes from our GPS-collared neonates and their dams to facilitate our accomplishment of this task. Our objective was to further evaluate the helicopter-assisted captures of moose neonates in 2013 and present preliminary analyses comparing the 2 all-ground calf capture approaches used in spring 2014. During 8-17 May 2013 we captured 49 newborn calves of 31 dams. The adult fled with the approach of the helicopter in most cases. Ultimately, 7 of 31 (23%) dams abandoned 9 of 49 (18%) calves, and twins (8 versus 1 singleton) appeared to be predisposed to abandonment. Hourly location data of neonates and dams indicated that capture-related abandonment involves movement behavior which is highly variable and complex. During the 1-6-hour interval post-capture, abandoning and non-abandoning dams were similar distances from calf capture-sites and their calves, but by the 7-12-hour interval mean distances of the 2 groups were diverging. By the 13-18-hour interval post-capture the non-abandoning dams were returning to their calves, whereas the abandoning dams were moving farther away. Paradoxically, movements of most of the abandoning dams included periodic returns to their calves, just as had occurred for some of the non-abandoning dams within the 48 hours post-capture. While our intense monitoring of movement behavior has illuminated much about capture-related abandonment, additional study should provide a greater understanding of its effects on the survival of neonates. During 8-15 May 2014, our initial approach to ground captures resulted in 7 of 12 (58%) neonates ultimately being abandoned by 5 of 9 dams (56%) at least in partial response to capture operations. Adhering to our Abandonment Contingency Plan, we successfully recovered 6 of the 7 calves and transported them to the Minnesota Zoo. Beginning 21 May, we reduced our capture teams to 2 people and limited our handling of calves to fitting the GPS collar quickly over the head to the neck and determining sex. By 19 June, we captured an additional 13 (8 males, 5 females) calves from 10 dams with no capture-related abandonment. These captures included 3 sets of twins and 7 singletons. While it is difficult to ascertain that any one factor was most important, we think that reducing the capture team to 2 people and mean handling time to <60 seconds likely had the most positive impacts.

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## INTRODUCTION

The winter moose population in northeastern Minnesota has decreased 51% from an estimated 8,840 moose in 2006 to the current (2014) estimate of 4,350 (Lenarz et al. 2009, 2010; DelGiudice 2014). Climate change (i.e., warming temperatures) has been implicated as an underlying factor in the declines of the state's northeastern and northwestern populations (Murray et al. 2006; Lenarz et al. 2009, 2010). In the latter, malnutrition and pathogens additionally were identified as contributing factors; now recent aggressive study in the northeast is accumulating evidence of similar impacts of such factors (Butler et al. 2013). Mean annual natural mortality rates of adults were similarly high in the northwest and northeast (21%, Murray et al. 2006; Lenarz et al. 2009; Butler et al. 2013; R. A. Moen, Natural Resources Research Institute [NRRRI], personal communication), and have remained elevated in the northeast up until this year (Carstensen et al. this issue).

Adult survival has a greater impact on ungulate population dynamics than that of juveniles; however, high annual variability in juvenile survival also can have a pronounced influence on a population's growth rate (Gaillard et al. 1998, 2000). Across much of moose range in Ontario, Canada, declining moose numbers and winter calf:cow ratios have been a cause for concern since the 1990s. Overall, natural causes were the leading mortality forces on calves. In Algonquin Provincial Park it was primarily predation by black bears (*Ursus americanus*) and wolves (*Canis lupus*), whereas in a Wildlife Management Area where hunting was permitted and accounted for 16% of calf mortality it was malnutrition, exposure, and tick-related mortality (Patterson et al. 2013).

Average pregnancy rates have been relatively high (83%) in northeastern Minnesota, but annually it has been variable (range of 55-100%, M. S. Lenarz, Minnesota Department of Natural Resources [MNDNR], unpublished data; Butler et al. 2013). Recently, Lenarz et al. (2010) reported an average annual survival rate of 0.40 for calves in the northeastern population. These crude estimates were based on fixed-wing flights conducted during May-June to determine whether radiocollared cows had newborn calves present, and again in April-May of the following year to determine if calves were still present. Further, based on the MNDNR's annual aerial moose survey conducted in January, the calf:cow ratio has declined from 0.52 in 2005 to 0.36 in 2012, and was as low as 0.24 in 2011 (Lenarz 2012).

The average annual survival rate of northeastern Minnesota moose was consistent with estimates from moose populations elsewhere where black bears and wolves were common (Hauge and Keith 1981), yet black bear predation on moose calves can be highly variable across North America (see Ballard's 1992 review). Determination of cause-specific mortality of calves was not part of the Lenarz et al. (2009, 2010) study design, consequently very little is known about the specific mortality forces or potential contributing factors.

The goals of our recently initiated moose calf research in northeastern Minnesota, a companion study to the MNDNR's adult moose study, are to enhance our understanding of the seasonal and annual survival of calves, specific causes of mortality and associated factors, and to quantify their impacts on the population's performance. The hazard, or instantaneous probability of death, for northern ungulates is highest at birth, and although it declines sharply during the first 12 months, it is markedly higher than during the subsequent prime years of life (DelGiudice et al. 2002, 2006; Lenarz et al. 2010). Fulfilling the primary goal of the calf study requires the ability to: 1) capture and GPS-collar a sample of newborn moose calves representative of the population in northeastern Minnesota, 2) closely monitor the movements and survival of moose calves, and 3) mount a rapid investigative response to calf mortalities.

Currently, this study is funded to support 3 field seasons. To efficiently and cost-effectively obtain a sample size of 50 newborn calves during the first spring (2013), we opted for capture and handling by an experienced helicopter capture crew (Quicksilver Air, Inc., Fairbanks, Alaska, and Peyton, Colorado). An inherent goal of any wild animal capture operation is to minimize capture-related injury or mortality of the study subjects, and additionally, in the case of calves, to minimize capture-related abandonment. Livezey (1990:193) defined abandonment as

“the permanent separation of mother and young causing the death of the young” and tried to focus increased attention on this potentially serious side-effect of such field operations so that more might be done to minimize associated losses. What became evident from his literature review and communication with many ungulate researchers was that the reported capture-related abandonment rates were highly variable, sorely underestimated, and rarely the primary focus of wildlife research.

Having captured more than 600 newborn moose calves, as well as neonates of other ungulate species, the company awarded our capture contract is considered one of the leading helicopter capture companies. However, 2013’s capture operations resulted in an unexpected and unacceptable number of captured neonates being abandoned. We carefully reviewed the data generated from those operations to better understand capture-related abandonment and to develop a more acceptable protocol for this year (2014). Having both the neonates and their respective dams fitted with GPS collars would be a significant aid to accomplishing this.

## OBJECTIVE

1. To further evaluate the helicopter capture operations of newborn moose calves in 2013 and to compare the 2 all-ground calf capture approaches used in spring 2014 in northeastern Minnesota. In a companion research summary (please see Severud et al. this issue), we expand on our description and evaluation of monitoring the GPS-collared calves and dams associated with the new capture approaches and mortality investigations, and we present preliminary findings related to cause-specific mortality.

## STUDY AREA

The 6,068-km<sup>2</sup> study site for this calf research is the same as that of the Environmental and Natural Resources Trust Fund (ENRTF)-supported research addressing survival and cause-specific mortality of adult moose in northeastern Minnesota (Figure 1). This area has been classified as the Northern Superior Upland region (MNDNR 2007) and is characterized by a variety of wetlands, including bogs, swamps, lakes, and streams; lowland stands of northern white cedar (*Thuja occidentalis*), black spruce (*Picea mariana*), and tamarack (*Larix laricina*); and upland conifers of balsam fir (*Abies balsamea*) and jack (*Pinus banksiana*), white (*P. strobus*), and red pines (*P. resinosa*). Trembling aspen (*Populus tremuloides*) and white birch (*Betula papyrifera*) occur on the uplands, often intermixed with conifers. Open lands included lowland and upland deciduous shrub and sedge meadows.

Potential predators of adult moose and their calves include gray wolves and black bears (Fritts and Mech 1981, Erb 2008, Lenarz et al. 2009, Garshelis and Noyce 2011). White-tailed deer (*Odocoileus virginianus*) share most of the study area with moose; their pre-fawning densities are managed at  $\leq 10$  deer per square mile (MNDNR 2011). The State moose hunt in northeastern Minnesota has been restricted to adult bulls-only since 2007 and accounted for 1.1-1.9% of the overall population (Lenarz 2011). A total of 87 licenses were purchased in 2012 for the State moose hunt, and 46 adult bulls were harvested. Sixteen moose (11 bulls, 5 cows) were harvested during the 1854 Treat Authority’s hunt and 20 bulls were harvested by members of the Fond du Lac Indian Band. Due to rapidly declining numbers, the State moose hunting season was cancelled in 2013 until further notice.

## METHODS

Beginning 1 May 2013, we began monitoring closely the locations and movements of 52 GPS-collared (Iridium GPS collars, Vectronic Aerospace, Berlin, Germany) adult female moose, which were determined to be pregnant during the previous winter by serum progesterone concentrations. Additionally, we similarly monitored 7 collared adult females not blood-sampled

during winter capture; their pregnancy status was unknown (see DelGiudice et al. 2013 and Severud et al. 2013 for additional details). Our primary monitoring objective was to record when and where pregnant females made their “pre-calving move” (Bowyer et al. 1999, Severud et al. 2013, McGraw et al. 2014). This is an atypical, long distance move that often occurs just prior to localization and calving (Figure 2). During early May 2014, we began similar monitoring of 16 known pregnant GPS-collared females and 50 collared females of unknown pregnancy status (see Severud et al. this issue); 1 of the pregnant and 6 of the “unknowns” abandoned calves in 2013, consequently their calves were not selected for capture this year.

The Iridium collars of the adults were programmed to record hourly fixes during May 2013 and May-June 2014 when most of the calving occurred. Adult location fixes, and subsequently calf fixes, were transmitted 4 and 8 times per day, respectively, to our base station located about 59 km north of the Twin Cities. We had continuous computer access to the base station. Additional details of our monitoring process are presented by Severud et al. (2013, and this issue).

We assumed that once cows made their pre-calving move then localized, they calved within 12 hours (R. A. Moen, NRRI, personal communication). We then allowed an additional 24 hours for bonding between the dam and her calf or calves for an estimated minimum total bonding time of 24-36 hours. Once monitored females had calved and were allowed this minimum bonding time, the calves were identified as “eligible” for capture and handling. Typically, by capture, actual bonding times were markedly longer than 24-36 hours. Each morning during the May 2013 operations our team provided the commercial capture crew with a list of females (identification numbers, VHF frequency) and their most recent GPS coordinates. Typically, in May 2014, we similarly identified cows eligible to be approached from the ground for calf capture.

In May 2013, the helicopter capture crew located the target dam from the air and then landed some distance away to allow the handler(s) to approach calves on foot. The handling protocol included fitting an expandable Globalstar GPS Calf Collar (440 g, Vectronic Aerospace, Berlin, Germany) over the head; fixing ear-tags; collecting blood by syringe for various analyses; weighing the calf; recording morphological measurements and a rectal temperature; and a physical examination to record any noteworthy injuries or abnormalities. We planned the complete handling protocol to require about 4-6 minutes per calf to limit separation from the dam (Keech et al. 2011), and in the case of twins, an attempt was made to handle both calves. Capturing and collaring both members of a twin set limited the risk of the dam abandoning the twin being handled with the one not being handled (M. A. Keech, Quicksilver Air, Inc., personal communication) and would provide a more thorough understanding of calf survival. Further, an important field objective was to capture, handle, and release twins at the same time and in close proximity (Keech et al. 2011). When twins were identified at captures in 2013 and 2014, handling crews achieved this objective with 100% success.

Initially, in May 2014, our plan was to have 3-person capture teams use the 2013 calf-handling protocol during ground captures. However, apparent capture-related abandonments of calves during 8-15 May prompted us to reconsider and revise the protocol. We trimmed our capture teams to 2 people and limited our handling to simply fitting the GPS collar and sexing each neonate, while simultaneously noting any injuries or abnormalities. Twins were collared simultaneously, 1 team member per calf. An additional capture would not be attempted until our monitoring documented that the dam had returned to its calf or calves post-capture.

The Globalstar GPS calf collars fitted in 2014 were the same as in 2013, except the collar band material was modified to minimize the risk of potential abrasions to the back of the calves' necks. As part of the revised capture protocol we employed during 21 May-19 June 2014, we removed a plastic sleeve fitted around the top of the expandable calf collars in an attempt to make them appear less obtrusive and conspicuous to the dam. Finally, as a precautionary measure, we used a commercial scent-blocking product on the handlers' clothes, gloves, and on the collars.



For our May 2014 operations we had developed an Abandonment Contingency Plan in collaboration with our consulting veterinarian and veterinary staff at the Minnesota Zoo. If our computer-monitoring showed that a dam was away from its newly captured calf/calves overnight or had reunited and then left them again, it was flagged as a potential abandonment. If a calf had been alone for 24 hours, we deployed a retrieval team. Precipitation or below normal May temperatures would hasten our response to recover abandoned calves. Recovered calves were brought to the Minnesota Zoo, which had agreed to act as a staging area before distributing calves to other zoos in the U. S. which had agreed to accept them. The Minnesota Zoo had agreed to accept 4-5 calves for its exhibits. All captures and handling protocols followed requirements of the Institutional Animal Care and Use Committee for the University of Minnesota (Protocol 1302-30328A).

## RESULTS AND DISCUSSION

### 2013 Moose Calf Captures

During 8-17 May 2013 we captured 49 (25 females, 24 males) newborn calves of 31 dams (Figure 3). Our process for monitoring and determining when GPS-collared dams had calved and met our minimum threshold of bonding time with their calves was highly successful (Severud et al. 2013). As reported last year, most of these dams were non-aggressive during the capture and handling of their calves (DelGiudice et al. 2013), particularly compared to dams of captured neonates in Alaska and Ontario (Keech et al. 2011, Patterson et al. 2013).

The twinning rate of the 2013 study cohort was unusually high at 58% (18 of 31 dams); 11, 4, and 3 were female/male, female/female, and male/male sets of twins, respectively. Thirteen adult females had singletons (6 females, 7 males). The high twinning rate likely had much to do with restricting our capture of newborn calves to a period early in the calving season when the birthing of twins is most likely to occur (see DelGiudice et al. 2013). Twinning rates can be highly variable. Patterson et al. (2013) reported an overall twinning rate of 16.7% in a 4-year study of moose calves in central Ontario. Keech et al. (2011) observed an overall average twinning rate of 42% (24-52%) for collared cows  $\geq 3$  years old during their 7-year study. The long-term average annual twinning rate in northeastern Minnesota may be about 29% (M. Schrage, Fond du Lac Band, unpublished data), whereas in northwestern Minnesota, Murray et al. (2006) reported an average twinning rate of 19%.

During 2013 we unexpectedly documented a high level of capture-related abandonment of calves by dams, but they were distributed at a low frequency throughout our 10-day operation (Figure 3). In almost all captures the adult fled at the approach of the helicopter or handler(s) and did not return to its calf or calves for varying lengths of time. Specifically, 7 of 31 (23%) dams abandoned 9 of 49 (18%) calves, apparently prompted by capture-related activities. Over the decades, rates of capture-induced abandonment of moose neonates have ranged up to 28% of calves captured, and up to 40% for caribou (*Rangifer tarandus*) (see the review of Livezey 1990). In a recent Alaskan study, researchers similarly used helicopters to capture 422 moose neonates and experienced 32 (8%) capture-related abandonments or mortalities (Keech et al. 2011). Because ours was the first study of free-ranging moose neonates fitted with GPS collars, it permitted nearly continuous monitoring of the calves and their GPS-collared dams. Indeed, unlike in other studies employing VHF telemetry, there was almost no way abandonment could be underestimated unless the collars malfunctioned. In a 4-year study involving all ground captures of moose neonates, capture-related abandonment of 5% of the calves was reported (Patterson et al. 2013). As with many of the dams in that study, VHF collars were fitted to the calves, but a high proportion of the dams were not collared. Consequently, the estimate of capture-related abandonment may have been biased.

In our study abandonment behavior was highly variable from day to day, and discernible patterns associated with various independent variables generally were not evident. On only 1 day did more than 1 dam abandon its calf, and on several days no dams abandoned calves,

despite a relatively high number of calves being captured (Figure 3, 16 May). Six of 25 (24%) females and 3 of 24 (13%) males were abandoned by their dams following capture and handling; however, according to a 2-sided Fisher's Exact Test the difference was not significant ( $P = 0.464$ ). As previously reported, there was no difference in mean birth-date, capture-date, minimum bonding time, body mass, hind leg length, or rectal temperature between calves abandoned versus those not abandoned (DelGiudice et al. 2013). However, whether 1 or both twins of a set were abandoned, a 1-sided Fisher's Exact Test showed an apparent ( $P = 0.104$ ) predisposition of twins to abandonment compared to singletons (Figure 4). Prompted by capture and handling, 33% of dams of twins abandoned their calves, whereas only 7% of dams of singletons responded to calf capture by abandoning neonates.

Hourly location data accumulated by GPS collars on the neonates and dams post-capture indicated that capture-related abandonment involves movement behavior which is highly variable and more complex than a dam simply taking flight in response to disturbances associated with the capture operation. Our first hourly locations indicated that on average the dams which did not ultimately abandon their calves actually fled farther than the those that did abandon (257 versus 183 m), although not significantly so (Figure 5). During the 1-6-hour interval post-capture, abandoning and non-abandoning dams were similar distances from the calf capture-sites and their calves, but by the 7-12-hour-interval mean distances of the 2 groups were diverging (Figure 5). By the 8-13-hour interval the non-abandoning dams clearly were returning to their calves, whereas the abandoning dams were steadily moving farther away from the capture-sites and their calves (Figure 5).

Paradoxically, abandoning movements included periodic returns by some of the dams to their calves, just as had occurred for some of the non-abandoning dams within 48 hours post-capture. We used the mean distance non-abandoning dams were away from their calves during the first 48 hours post-capture (256 m) as a threshold distance to indicate that dams had essentially reunited with their calves at varying points. Five abandoning dams and 5 non-abandoning dams returned a mean 1.5 (SE = 0.2, range = 1-2) and 1.3 times (SE = 0.3, range = 1-3), respectively. Interestingly, though they shared similar aspects of this post-capture movement behavior, the abandoning dams were much farther from their calves just before returning (mean = 1,531, SE = 210, range = 1,105-2,223 m) than the non-abandoning dams (mean = 582, SE = 80, range = 402-812 m), and they did not stay with their calves as long (mean = 1.5, SE = 0.2, range = 1-2 hours versus mean = 4.7, SE = 1.1, range = 1-7 hours) once they returned. Additionally, 5 of the abandoning dams stayed with their calves immediately after capture for 1-11 hours before leaving. While our intense monitoring of movement behavior has illuminated much about capture-related abandonment, additional analyses should provide a greater understanding of its direct and indirect effects on the survival of these neonates.

## **2014 Moose Calf Captures**

During 8-15 May 2014, our initial approach to ground captures resulted in 7 of 12 (58%) neonates ultimately being abandoned by 5 of 9 dams (56%) at least in partial response to capture operations (Figure 6). Capture-related abandonments began on the first day of captures with a set of twins, after which we stopped operations for several days to reconsider our protocol. The first day's capture had required a long trek ( $\geq 6.4$  km) under challenging field conditions (e.g., occasional deep snow), and we concluded that perhaps we had disturbed the dam well in advance of our capture of the twins. Consequently, we decided to employ a less disturbing ground approach, which we anticipated would be facilitated by rapidly improving field conditions. We also removed the more invasive blood-sampling from our handling protocol, which also would reduce handling time. Overall, from a field perspective, we viewed our captures as progressing smoothly during this initial interval without any unforeseen problems which would lead to abandonment. Our dam monitoring approach (see Severud et al. this issue), targeting GPS coordinates of the center of the dam's calving localization/cluster, and using a hand-held GPS unit in the field allowed us to locate calving sites and calves efficiently.

Search times for capturing calves typically required a mean of 5 minutes (SE = 3.0, range = 0-17,  $n = 6$ ). In 5 cases we did not find the calves we sought to capture (Severud et al. this issue). Handling required a mean 8.9 minutes (SE = 1.8, range = 5-23,  $n = 9$ ); this included twins, and there was no difference for calves abandoned versus those not abandoned. Having removed the disturbance of the helicopter from our capture and handling protocol this year, we initially were quite sure that a capture-related abandonment effect would be minimal, if not eliminated. However, the capture-related abandonments continued to occur during the 8-15 May interval (Figure 6).

Although our sample size was limited to a total of 12 calves by 15 May, twinning appeared to be less of a factor in predisposing calves to capture-related abandonment than during the 2013 operation, but in both cases of dams abandoning twins during this phase, they abandoned both neonates (Figure 4). Additionally, similar to 2013, there was no apparent potential influence of sex of the calf on capture-related abandonment (3 males, 4 females). Overall, mean bonding time (or calf age) at capture during this interval was 49 hours (SE = 4.7, range = 24-76,  $n = 9$ ); there was no difference between calves which were abandoned (mean = 47, SE = 8.4, range = 24-76 hours,  $n = 5$ ) versus those that were not (mean = 52, SE = 3.3, range = 43-58 hours,  $n = 4$ ). These bonding times were similar to those of the 2013 capture operations (mean = 54, SE = 2.7, range = 31-116 hours,  $n = 49$ ) when there also was no difference associated with calves abandoned versus those that were not (DelGiudice et al. 2013).

Adhering to our Abandonment Contingency Plan, we successfully recovered in good condition 6 of the 7 calves which had been abandoned and transported them to the Minnesota Zoo. There, the veterinary staff and zookeepers examined them thoroughly and have since been caring for them. Mean recovery time was 42 hours (SE = 5.8, range = 24-49,  $n = 4$ ) post-capture. The dam of the calf not recovered alive had been with it for 8 hours immediately post-capture, moved away, and then returned to within 200-300 m. Our intense monitoring had shown that the calf was moving about, and so we had hoped that periodic nursing bouts were occurring during the 48 hours post-capture, but a necropsy had concluded that this was unlikely.

With our seventh capture-related abandonment on 15 May, we considered not resuming captures during this calving season. However, during a week of contemplating our mode of operation, we decided to attempt a final new capture approach as described in the Methods section. Beginning 21 May, employing a 2-person team, we began approaching 1 calving dam daily once the minimum bonding time had elapsed. During 21 May-19 June, we captured an additional 13 (8 males, 5 females) calves from 10 dams with no capture-related abandonments. These captures included 3 sets of twins and 7 singletons. Our current success at eliminating capture-related abandonment may be attributable to several factors. While it is difficult to ascertain that any one factor was most important, reducing mean handling time to <60 seconds (0.9, SE = 0.2, range = 0-2 minutes,  $n = 10$ ), for a singleton or twins, may have been most beneficial. Slipping the expandable collar over the head and sexing the calf constituted "handling," but we also carried the calf or calves back to the nearby calving-site for release. The smaller capture team may have contributed to a quieter approach and an apparently briefer search time (mean = 1.5, SE = 1.0, range = 0-11 minutes,  $n = 10$ ), although during both capture phases search times were rather variable. Our allowed bonding time at capture (mean = 74.6, SE = 5.7, range = 53-109 hours,  $n = 10$ ) tended to be longer than during the initial phase and compared to our 2013 operations. Bonding times have been highly variable among studies. Patterson et al. (2013) reported bonding times before capture of 9.5-58 hours (median = 19 hours) on their WMU49 site and <48 hours (48%) and 48-120 hours (52%) at Algonquin Provincial Park. In Interior Alaska, Keech et al. (2011) estimated mean bonding times of 2.6 days (62 hours) and a range of 0.5-11 days (12-264 hours). Finally, the absence of conspicuous ear-tags on calves and collar modifications during the second phase of 2014 captures may have contributed to limiting potential recognition problems between dams and calves, but this is difficult to quantify.

## Considerations for Future Capture Operations

Currently, capture-related abandonment of newborns of moose and other ungulates is poorly understood and likely has been sorely underestimated due to the limitations of conventional VHF telemetry. However, our deployment of recently developed expandable GPS collars on moose neonates of GPS-collared dams has demonstrated enhanced potential for increasing our understanding of those factors which have the greatest influence on abandonment behavior and how to minimize that influence. Our hourly location data also indicated that the operative definition of capture-related abandonment and how to respond to suspected abandonment in the field as it occurs warrants re-examination. As challenging as capture-related abandonment has made our calf capture operations during 2013 and 2014 of this survival and cause-specific mortality study, what we have learned and our ability to adapt reflect a significant success with respect to our calf capture protocol. We now will be able to apply what we have learned towards next year's capture operations with the promise of obtaining a more rigorous seasonal sample size of collared calves to add to the 44 calves we have been able to study thus far for survival and natural causes of mortality (Severud et al. this issue).

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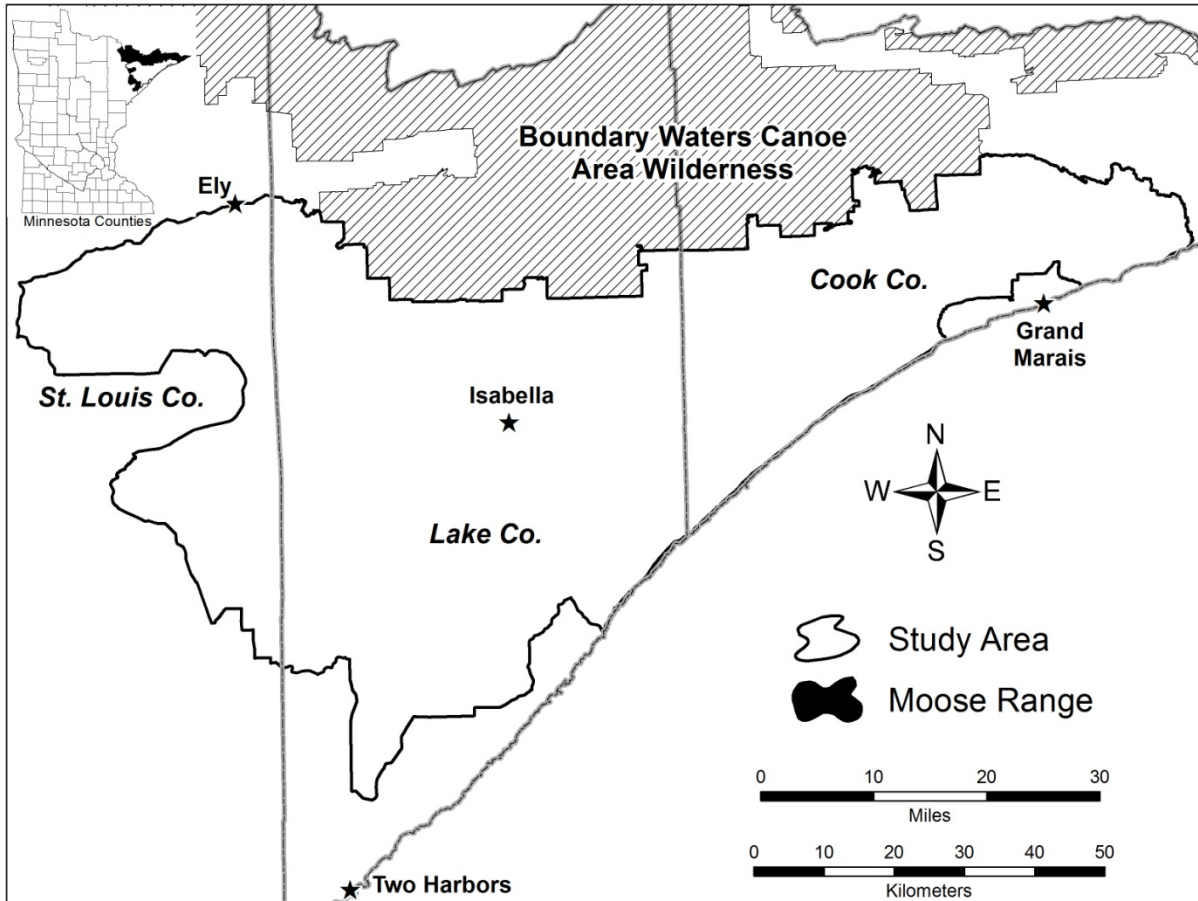


Figure 1. Study area for the study of moose calf survival and cause-specific mortality, northeastern Minnesota, 2013-2017.

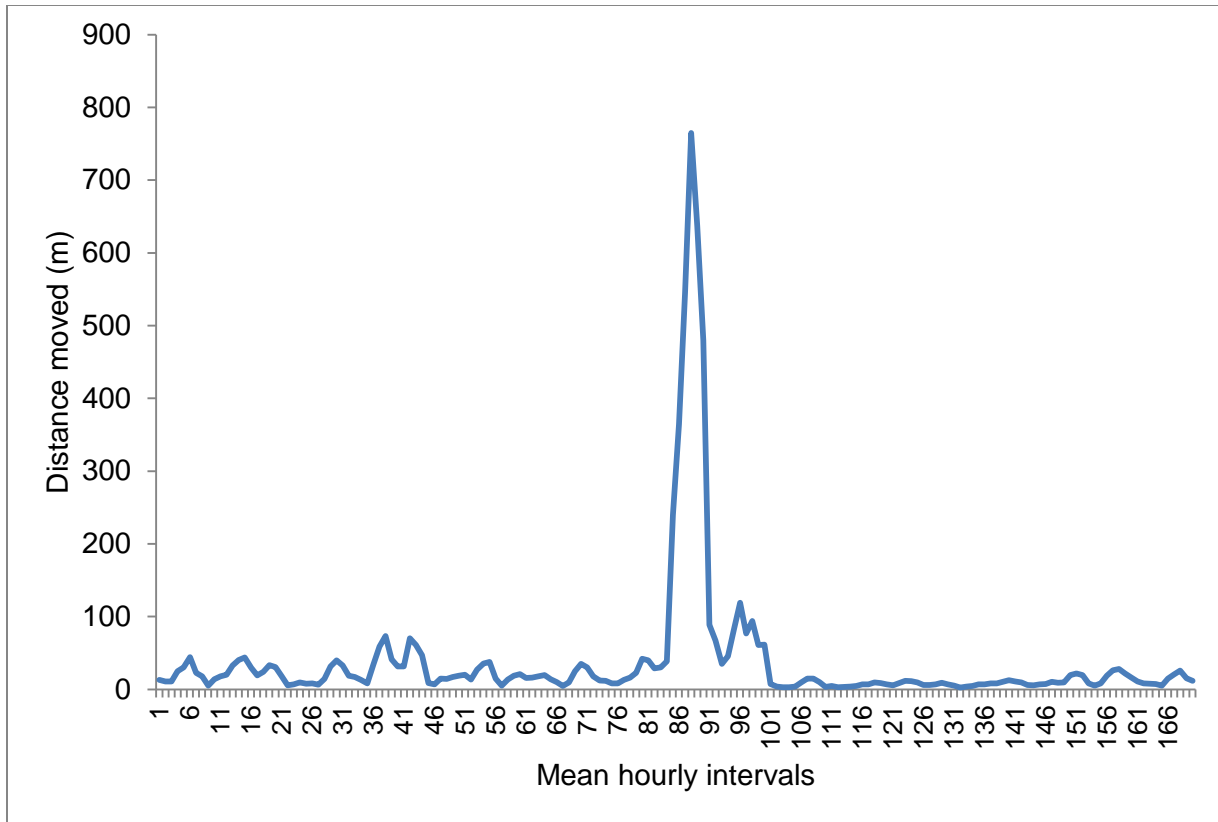


Figure 2. An example of calculated mean hourly distances moved by a pregnant, adult female moose (No. 12500) from 12:04 am, 1 May to 1:42 pm, 8 May 2013. The elevated peak at hour 88 represents the dam’s primary “pre-calving move” (about 800 m), but she didn’t localize completely until after hour 97. We used the latter as indicative of calving so as not to over-estimate bonding time, which was measured during the interval between then and capture time (hour 172). The pre-calving move was used for monitoring calving activity in northeastern Minnesota and for helicopter-assisted capturing of neonates during 8-17 May 2013 and for all ground captures during 8 May-19 June 2014.

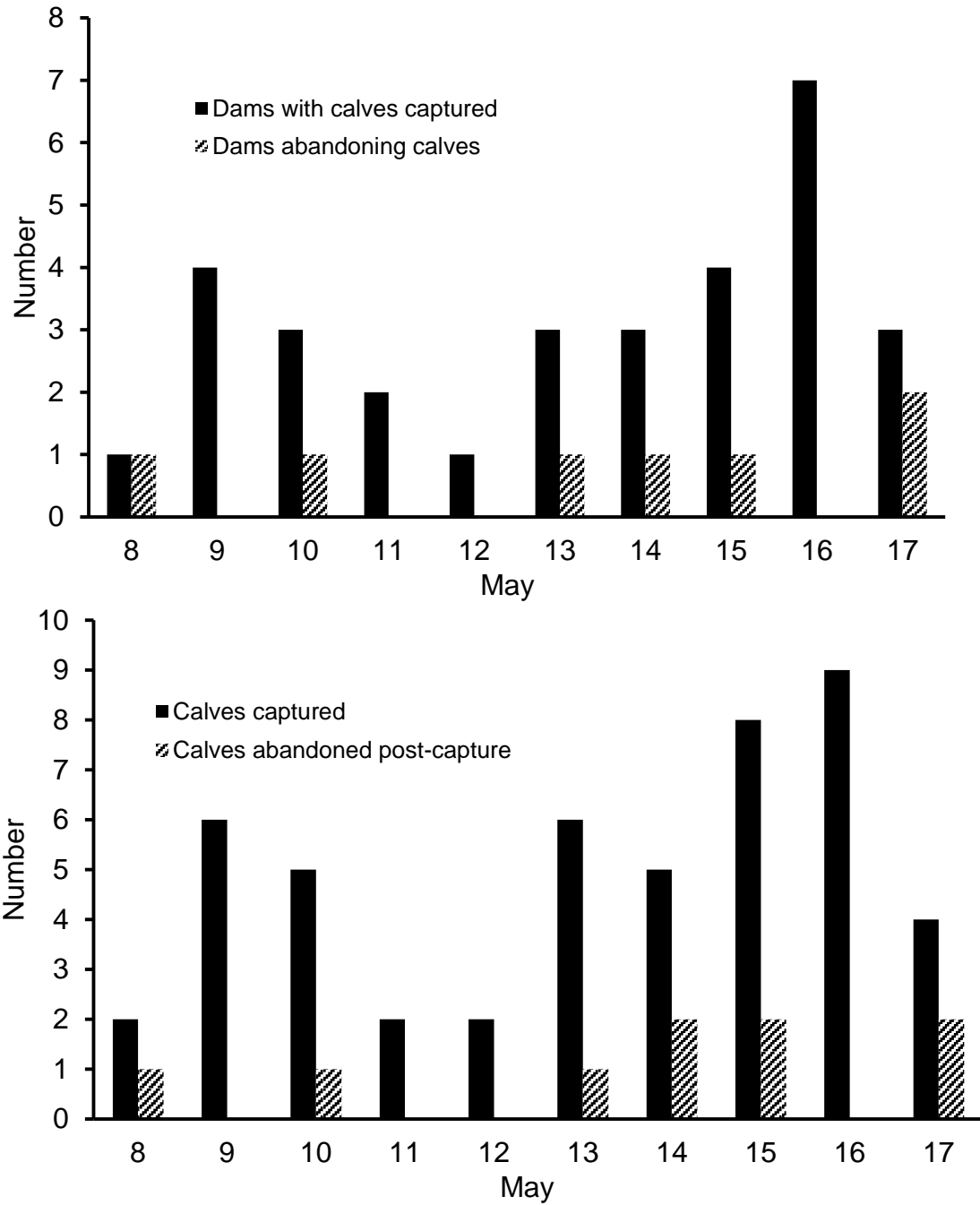


Figure 3. Temporal distributions of the total number of moose dams whose calves were captured, handled, and released compared to those that abandoned their calf or calves post-capture (top), and the total number of moose calves captured, handled, and released compared to those that were abandoned (bottom) apparently in response to capture operations, northeastern Minnesota, 8-17 May 2013.



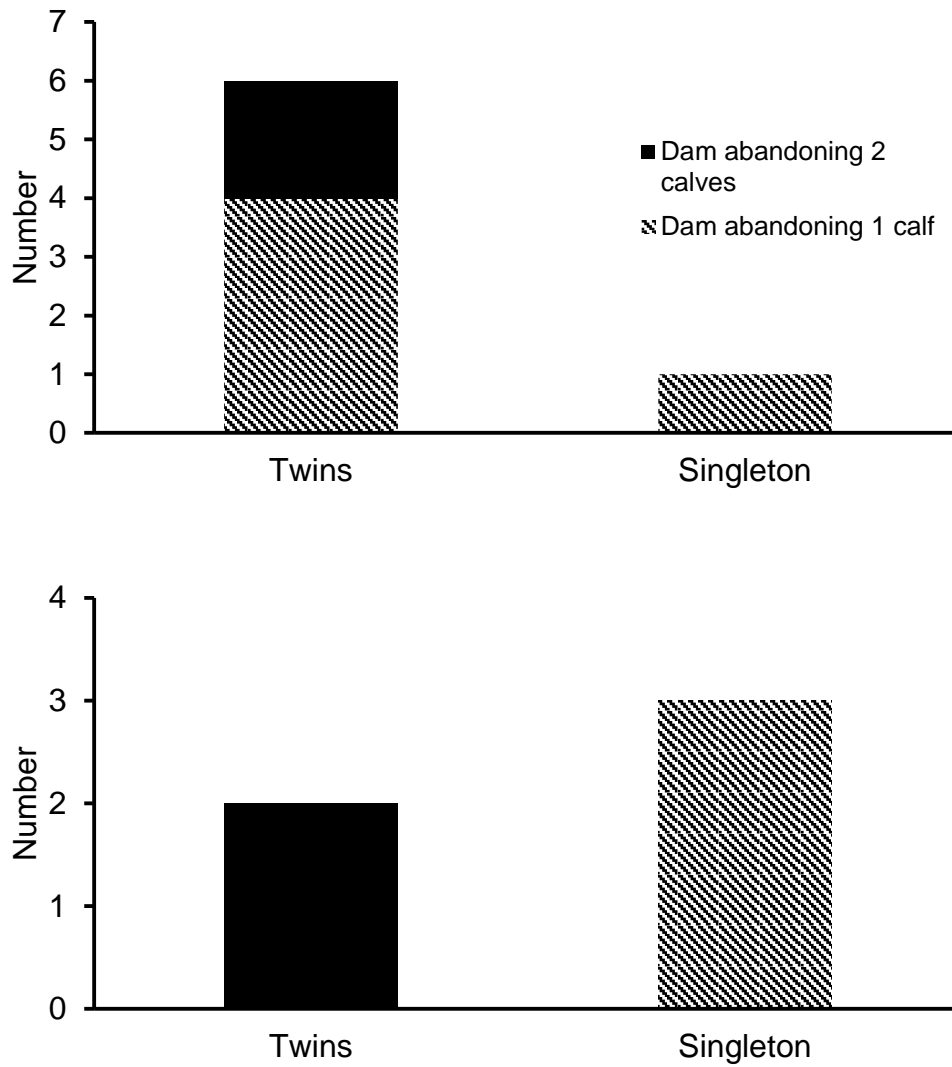


Figure 4. Comparison of dams abandoning twins versus singletons in response to helicopter-assisted captures (8-17 May 2013, top) versus ground captures not assisted by helicopters (8 May-19 June 2014, bottom), northeastern Minnesota.

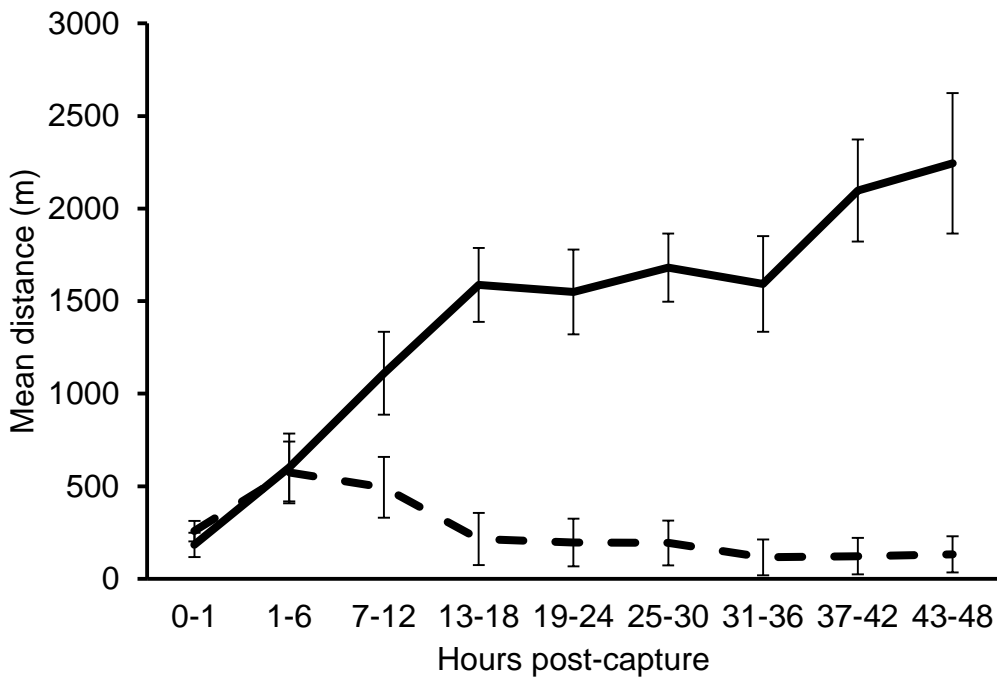
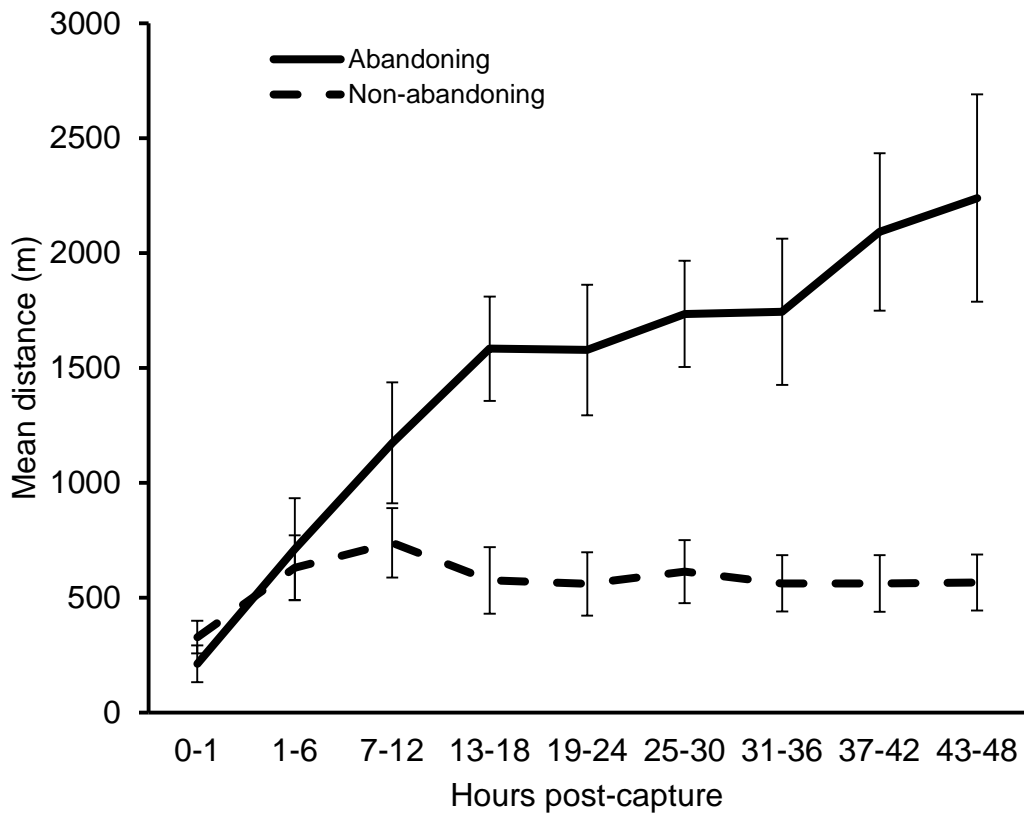


Figure 5. Mean ( $\pm$  SE) distance (m) of abandoning and non-abandoning moose dams from the capture site of their newborn calves (top) and from the calves (bottom, 2-4 days old) during 6-hour intervals up to 48 hours post-capture, northeastern Minnesota, 8-17 May 2013.

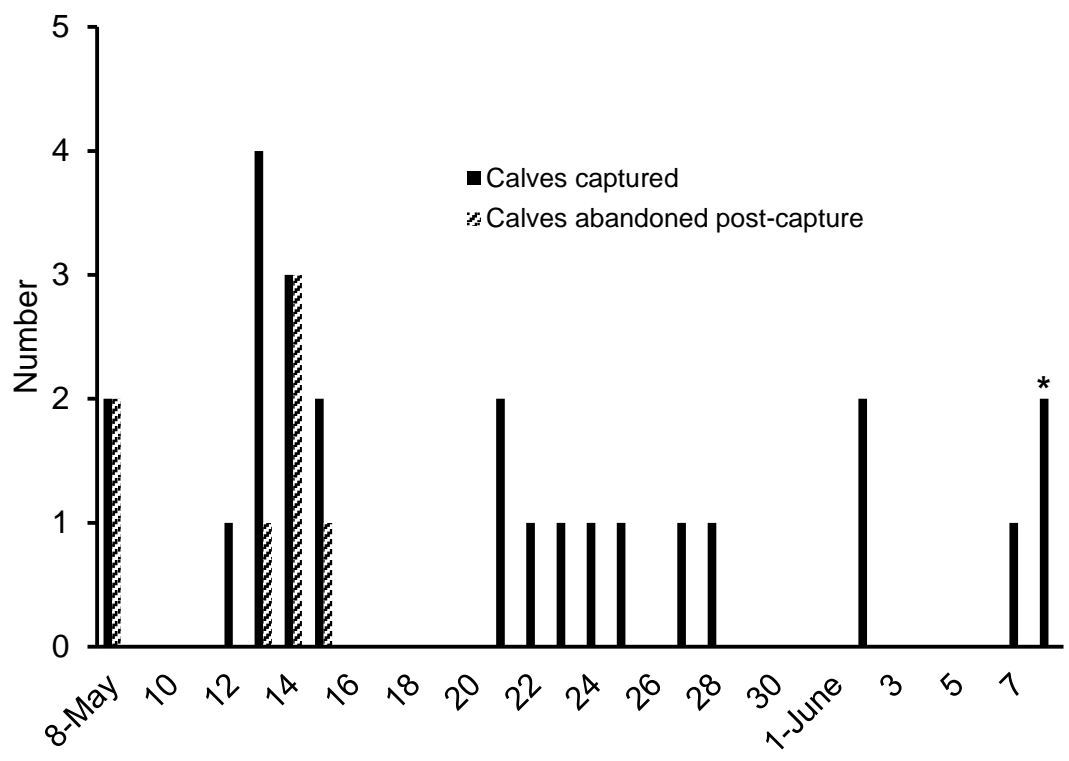
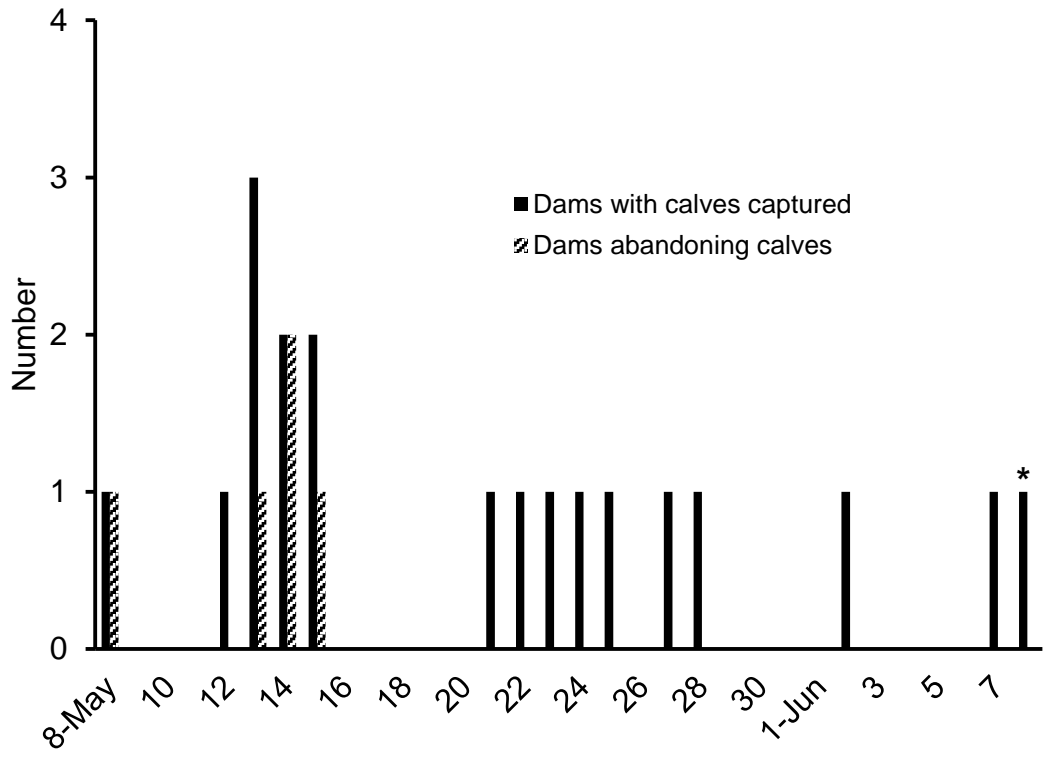


Figure 6. (top), and the total number of moose calves captured, handled, and released compared to those that were abandoned (bottom) apparently in response to capture operations, northeastern Minnesota, 8 May-19 (\*) June 2014.



## USING GPS COLLARS TO DETERMINE MOOSE CALVING AND CAUSE-SPECIFIC MORTALITY OF CALVES IN NORTHEASTERN MINNESOTA: PROGRESS REPORT ON SECOND FIELD SEASON

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### SUMMARY OF FINDINGS

Adult survival is an important driver of large herbivore population dynamics; however, low and variable recruitment also can have a strong influence on population trajectory. The northeastern Minnesota moose (*Alces alces*) population has been exhibiting a downward trend since 2006. Our research was initiated because neonatal and seasonal survival rates and specific causes of mortality (e.g., predation, undernutrition, disease) of calves are largely unknown. Beginning 1 May 2014 we monitored 70 adult female moose fitted with global positioning system (GPS) collars (16 confirmed pregnant at capture during February 2014 by progesterone concentrations, 50 unknown, 4 not pregnant) for long-distance pre-calving movements followed by localization. Our ground crew captured calves from 19 of the 56 cows which had made the pre-calving move then localized. Of these 56 cows, 13 were of the 16 confirmed pregnant and 43 collared in 2013 were of unknown pregnancy status. Twenty-five neonates from 19 dams (32% twinning rate) were fitted with expandable GPS collars during May and June 2014 for subsequent monitoring during their first year. We are retrieving collars from calf mortalities and estimating proximate causes of mortality on site. Mean elapsed time between estimated time of death and mortality investigation ranged from 21.5 to 62 hours; accessibility and functionality of individual collars were influential factors. Seven mortalities (6 natural, 1 capture-related) have occurred, and 7 collars were slipped during 16 May-22 June; 5 calves remained “on air” as of the latter date. After censoring the 7 slipped collars and 7 capture-related abandonments (6 retrieved and placed in zoos, 1 mortality), 6 of 11 calves have died (55%) naturally compared to 17 of 34 calves (50%) during this time period in 2013. A natural abandonment possibly due to umbilical infection, 1 predation by black bear (*Ursus americanus*) and 4 by wolves (*Canis lupus*) are preliminary causes of death. Identifying specific causes of calf mortality and understanding their relations to various landscape and other extrinsic factors should yield insight into mechanisms contributing to the declining moose population in northeastern Minnesota and serve as a basis for an ecologically-sound management response.

### INTRODUCTION

The moose is an iconic species of northern Minnesota, which has afforded valuable hunting and viewing opportunities (Minnesota Department of Natural Resources [MNDNR] 2012). The MNDNR has listed moose as a Species of Special Concern ([http://files.dnr.state.mn.us/natural\\_resources/ets/endlist.pdf](http://files.dnr.state.mn.us/natural_resources/ets/endlist.pdf)). Recently, the northwestern population declined precipitously to less than 100 moose due to a variety of natural factors (Murray et al. 2006). The northeastern population is in decline and is experiencing adult mortality rates similar to those of the northwestern population as it decreased (Lenarz et al. 2009, 2010; Butler et al. 2013; Carstensen et al. this issue).

Large herbivore population growth ( $\lambda$ ) is most sensitive to variation in adult survival (Gaillard et al. 1998, 2000; Lenarz et al. 2010). Juvenile survival has less of an impact on overall

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population growth, but differences in temporal variation of juvenile survival may be important in accounting for between-year variation in  $\lambda$  (Gaillard et al. 2000). Fecundity and calf survival ultimately determine recruitment rates which are important to more fully understanding population dynamics (Van Ballenberghe and Ballard 2007). When viable populations of predators are present, predation can be a primary cause of mortality of temperate ungulate neonates (Linnell et al. 1995, Carstensen et al. 2009). Less is known about other specific ultimate or proximate sources of moose calf mortality or factors which may influence predation and other sources of mortality. It also is unclear when predation is compensatory or additive to other sources of mortality (Franzmann et al. 1980, Linnell et al. 1995), although a recent study documented additive effects of predation on moose calves in Alaska (Keech et al. 2011). The degree of predation's impact on calf survival depends on the extant predator guild and relative densities of predator and prey (Eriksen et al. 2011, Patterson et al. 2013).

Particularly after the calves' first summer, the magnitude of mortality from wolves (*Canis lupus*) is variable (Patterson et al. 2013). Wolves are more adept at killing calves in deep snow (DelGiudice et al. 2009), but wolves in an Alaskan study also were responsible for calf mortalities in fall (Keech et al. 2011). Typically, bear-caused (*Ursus* spp.) mortality of calves is greatest closer to their parturition, more immediately following emergence of bears from winter dens (Bastille-Rousseau et al. 2011). Cows in poor nutritional condition may defend calves less vigorously (Patterson et al. 2013). Further, risk of predation is not independent of maternal care and experience (Ozoga and Verme 1986). The importance of natural non-predatory causes of calf mortality likely varies during different times of the year, such as malnutrition and exposure in spring, or malnutrition and tick-related deaths in winter (Patterson et al. 2013). The extent to which diseases drive calf mortality is not well understood, although diseases have led to poor recruitment in moose (O'Hara et al. 2001, Murray et al. 2006). Juvenile animals are more predisposed to parasites than adults, and pathology related to parasite infection may be an important source of mortality for moose calves (Jenkins et al. 2001, Murray et al. 2006). Further, small calves may not be tall enough to efficiently nurse, leading to malnutrition (Murray et al. 2006). Drowning and climate have been known to affect moose calves more than predation in some regions (Crête and Courtois 2009). In winter, temperature and snow depth can be more important causes of mortality than predation (Keech et al. 2011).

Pregnant cow moose tend to move long distances (mean = 6 km) prior to localizing to give birth (McGraw et al. 2014). These distances are typically much longer than movements between foraging and bedding sites. Following a long movement, calving localizations as measured by GPS collars resemble mortality localizations. A cow with calves may stay within a 1.7-ha area for up to 7 days (range = 1-18 days; McGraw et al. 2014).

Until the current study, expandable GPS collars have not been fitted to moose neonates and have only recently been used on other wild ungulate neonates in small numbers (fallow deer [*Dama dama*],  $n = 3$ , Kjellander et al. 2012) or in captivity (domestic horse [*Equus caballus*],  $n = 4$ , Hampson et al. 2010). Observable fine-scale movement and habitat use patterns of moose calves, made possible by GPS collars, are facilitating examination of landscape features important for their survival and rapid investigation of mortality events. Having dams and calves fitted with GPS collars also allows us to study the importance of their proximity to juvenile survival.

## OBJECTIVES

1. Evaluate monitoring of movement behavior of GPS-collared adult female moose to determine timing and location of calving; and
2. Evaluate remote tracking of GPS-collared calves and dams to determine and investigate calf mortalities and assign cause.

## METHODS

Our study area is the same as that of the Environmental and Natural Resources Trust Fund (ENRTF)-supported study focused on survival and cause-specific mortality of adult moose in northeastern Minnesota (see Figure 1, DelGiudice et al. this issue). As part of the adult moose mortality study, 111 (84 females, 27 males) and 37 (25 females, 12 males) moose were captured and fitted with Iridium GPS collars (Vectronic Aerospace, Berlin, Germany) during January 2013 and February 2014, respectively (Butler et al. 2013, Carstensen et al. this issue). Blood was collected and analyzed for pregnancy;  $\geq 2.0$  ng/mL was the progesterone concentration threshold indicative of pregnancy. We monitored cow movements during pre-parturition and calving, with particular attention afforded to pregnant cows. We looked for movement patterns indicative of calving, including a long-distance movement followed by localization (Bowyer et al. 1999, McGraw et al. 2014).

We began monitoring 70 collared adult female moose (16 confirmed pregnant at capture by progesterone concentrations, 50 unknown [captured and collared in 2013], 4 not pregnant) on 1 May 2014. Based on a 77% pregnancy rate of total tested cows in 2014, we assumed ~55 (39 unknown + 16 known) cows to be pregnant in 2014. Cow collars were programmed to collect hourly locations during May and early-June and to transmit these locations 4 times per day. An automated R program (J. D. Forester, University of Minnesota, unpublished data) generated emailed reports 2 times daily (0500, 1700 hours). Reports contained a file (pdf) displaying various movement and location metrics for each collared cow, and table (csv format) and map (kml format) files with all recent locations of each animal. The pdf reports contained a rough map of northeastern Minnesota with all current cow locations displayed and a summary table of all animal locations and distances moved in the last 24 and 48 hours. The metrics for each cow included the date and time of the last location, movement path of the last 5 days, movement path of the last 24 hours overlaid on Google Earth imagery, a plot showing 3-hour average distances moved, and each cow's data on a single page (Figure 1). The distance plot showed peaks in movements that we then monitored for possible dampening of movements (localization). If the cow moved  $< 100$  m during 36 hours after making a long-distance movement (dam-calf bonding time), the program flagged that cow as "localized," and that cow's calf was determined eligible for capture. Additionally, a blue line representing predictions from a regression based on 2013 calving movements showed if a calving move may have occurred in the last 12 hours. Larger spikes indicate higher likelihood of a calving event. A gray line shows relatively large or small movements over the past 12 hours. When a cow's calf was eligible for capture, we also checked her movement path on the Vectronic Aerospace website (<https://www.vectronic-wildlife.com>; Figure 2). After capture, dams and calves were paired for the automated reports, and an additional metric was added to the movement plot (proximity between dam and calf, Figure 3). This plot was monitored for possible abandonments. Calves also were added to the report and had a page similar to that of the cows displaying their location and movement metrics.

Once a cow localized for  $> 36$  hours, a ground crew searched the potential calving site for the cow and calf or calves (see DelGiudice et al. this issue). Each captured calf was fitted with an expandable Globalstar GPS Calf Collar (520 g; Vectronic Aerospace, Berlin, Germany) and 2 ear-tags, and was weighed ( $\pm 0.5$  kg). Collars were programmed to take a fix hourly and transmit every third successful fix. All fixes are stored on board the collar. After observing several calves with abrasions on the dorsal side of their necks in winter 2013-2014, we worked with Vectronic Aerospace to redesign the collar belting to be wider, softer, longer, and to include a sleeve to preclude abrasion. Twins each received a collar and ear-tags. As feasible relative to the dam's behavior, the crew also took morphometric measurements (neck circumference, girth, total body length, hind leg length), collected blood and hair, and measured a rectal temperature. As captures progressed, handling was reduced to simply fitting the collar and determining sex (DelGiudice et al. this issue). All captures and handling protocols followed requirements of the Institutional Animal Care and Use Committee for the University of

Minnesota (Protocol 1302-30328A) and were consistent with guidelines recommended by the American Society of Mammalogists (Sikes et al. 2011).

We monitored each collared calf daily until mortality or until its collar drops off (designed to be ~400 days). We relied upon the collars to send mortality alert notifications to cell phones via text message (i.e., SMS) when mortalities occurred, but after several mortalities went unnoticed last season (Severud et al. 2013), we began using the Vectronic Aerospace website and GPS Plus X software to check if calf collars were far from dam collars or in mortality mode. Each morning all dam and calf groups are checked and monitored closely throughout the day if separated by >500 m.

When we receive a mortality alert or determine a mortality may have occurred, we dispatch an investigative team (goal of within 24 hours of death) to collect the collar and carcass remains and to determine the cause of death (Ballard et al. 1979). To avoid possible investigation-induced abandonment, investigations are delayed if the dam is still in the area, especially if she is with a twin. Our primary field objective is to recover the entire carcass and deliver it to the University of Minnesota's Veterinary Diagnostics Laboratory (VDL) for necropsy. If the carcass cannot be extracted and transported, we perform a detailed field necropsy. If scavenged or fed upon, fresh organ and tissue samples are collected and shipped or transported to the VDL as feasible (Butler et al. 2011). Care is taken to haze off predators and scavengers when approaching a potential mortality site; bear repellent spray and firearms are available as a last resort for protection, but their use is not necessarily anticipated (Smith et al. 2008, 2012). We postpone the investigation when predators are sighted on the carcass; return is dependent on the age and size of the carcass as an indication of how long the predator or scavenger may feed.

Once we begin a thorough investigation of the site, we are careful not to disturb potential evidence. We photograph tracks and scat and collect scat when identification is uncertain. We note the presence of puncture wounds on the neck, skull, or hind quarters and claw marks across the body and take photographs of all wounds. When the hide is present, we note if it is inverted, which may indicate a bear was feeding on the carcass. We document the consumption of viscera, the rumen or its contents. Wolves may chew on ribs and ends of long bones, whereas bears are more likely to cache pieces of the carcass. To determine if injuries occurred ante mortem, we look for subcutaneous hemorrhaging or sprays of blood on the collar or surrounding vegetation. Signs of a struggle (broken or matted vegetation, disturbed ground) also are indications of predation. We take note of the position of the carcass (lateral or sternal), and the distribution of body parts (scattered or near the carcass). An odor of decomposition or many fecal pellets in the area may indicate scavenging versus predation.

If we find a GPS collar without a carcass or other evidence of predation, we backtrack to the last known locations of the calf and its dam to examine a larger area in an expanded search. The adult Iridium collars are more accurate than the calf Globalstar collars (W. J. Severud, University of Minnesota, unpublished data), so we use the cow's locations from the approximate time of death of the calf to look for a kill-site or evidence of the cause of mortality. We conclude a collar has been slipped rather than indicating a mortality if the breakaway section is frayed or the bolts holding the breakaway section are loose, coupled with both an absence of blood on the collar and lack of evidence within a 30-m radius of the collar. Additionally, the dam's behavior, large movements away or localization around the estimated time of death, can be used as an indication predation or a slipped collar, respectively.

## RESULTS

We observed 56 of 70 cows (80%) display calving movement behavior (pre-calving movement followed by localization for >36 hours). Mean localization date was 19 May 2014 (median = 18 May, range = 5 May-16 June; Figure 4), with 75% of the localizations occurring during 11-22 May 2014.

We deployed 25 expandable GPS collars on neonates (11 females, 14 males) captured from 19 dams (32% twinning rate) during 8 May-19 June 2014 (Figure 5; see DelGiudice et al.

this issue for additional details). Twin sex composition was 3 male/female, 2 male/male, and 1 female/female. Of the 19 dams, 4 were confirmed pregnant by progesterone and 15 were unknown. Once we deployed 2 collars on a set of twins, we ceased capture operations due to abandonment concerns possibly resulting from late deep snow cover (DelGiudice et al. this issue). Once conditions improved, we began collaring again, but ceased operations when abandonments continued. We then decided to reduce handling to solely fitting the collar and determining sex. We observed no capture-related abandonment after this final modification of our methods and collars (DelGiudice et al. this issue).

We weighed, measured hind leg length, neck circumferences (upper and lower), and took temperature for 12 calves (6 F, 6 M, 6 singletons, 3 sets of twins) during the first phase of captures (8-15 May 2014). We measured chest girth for 2 calves (1 F, 1 M, 1 singleton, 1 twin), and measured total body length of 1 calf (M, twin; Table 1). Mean rectal temperature was 101.4 °F (SE = 0.7, range = 97.7-107,  $n = 12$ ).

We visited 5 cows (2 pregnant, 3 unknown) which exhibited movement patterns indicative of calving, yet no calves were found. We often found evidence of a calving site (bare ground, many pellets and beds) or calves (pellets, tracks), but could not locate the calves. We limited search time to ~5 minutes to lessen disturbance. Often the cows returned to these sites after we departed, indicating a calf or calves may have been present, but undetected by our capture team.

As of 16 June 2014, we have documented 6 natural mortalities (Figure 6) and 7 slipped collars; 5 collared calves remain “on air.” Capture-related activities accounted for 6 abandonments and 1 mortality associated with capture-related abandonment (DelGiudice et al. this issue). Of the remaining 6 mortalities, there was 1 natural abandonment associated with an umbilical infection, 1 bear-kill, and 4 wolf-kills. Histological and disease-screening results from the VDL are pending. After censoring the capture-related abandonments and slipped collars, 6 of 11 calves have died (55%) as of 22 June 2014, with 5 of those preyed upon by wolves or bears (Figure 7).

Of the 6 mortalities we investigated on site, 1 of the collars failed to send a mortality alert text message. We investigated the site because the collar was not transmitting and the dam had departed from the last known calf location. This collar was found in a shallow puddle and never transmitted a mortality message to the satellite base station (and stopped collecting and sending GPS fixes).

Mean elapsed time between estimated time of death (when the collar entered mortality mode) and mortality investigation was 46 hours (range = 0-239 hours,  $n = 17$ ). A collar that was submerged in a shallow puddle did not collect locations or transmit for 7 days and took 158 hours to investigate. The mean response-time was 62 hours (range = 9.5-239 hours,  $n = 10$ ) when we received a mortality alert text message, although in 7 of these cases the collar was slipped (mean = 85, range = 13-239 hours). For natural mortalities (omitting slipped collars), mean response time was 21.5 hours (range = 9.5-52.5,  $n = 6$ ).

All collared dams were tracked for calving movements from 1 May to 20 June. We found 56 cows made calving movements and localizations indicative of calving. We documented several mortalities of uncollared calves which were incidental to mortalities of collared adult moose (Carstensen et al. this issue). One adult was killed by wolves and we found 2 partial calf skulls. Calving movements suggested that this cow calved and then she and her twins were killed by wolves. Another cow died of malnutrition and its calf subsequently died within 30 m. In another case, a cow was investigated due to collar malfunction. The investigation revealed cow and calf beds and tracks, but no evidence of predation or scavenging.

## DISCUSSION

Tracking GPS-collared cow movements was again this year a highly reliable way to estimate whether or not they had calved. Of the 24 dams suspected of calving and subsequently visited, we collared calves from 19 (79% success rate). We do not know for certain whether the 5 dams visited without collared calves had given birth, but site evidence and dam behavior often



suggested that dams had calved. In one case we visited a dam that was later determined to not have localized once more GPS fixes were transmitted. Our study objective was to fit GPS collars to 50 newborns over the entire calving period (May to early-June); we fit 25 collars over 13 days of captures (several periods of inactivity due to method refinement; DelGiudice et al. this issue). We decided to track cows during May and June to look for movement patterns indicative of calving rather than fitting vaginal implant transmitters (VITs) to pregnant cows for several reasons. Fitting VITs would have required determining pregnancy status during winter captures, which would have added significant expense and time to the handling of the captured adult females, and most of the cows we monitored for calving were collared the previous year. Monitoring known pregnant cows for a “calving move” did not limit us to only those 16 pregnant females which would have been fitted with a VIT; the latter also would have required the expense of monitoring from a fixed-wing aircraft. Monitoring calving movements has been invaluable as we plan to capture calves from collared cows that we will not need to recapture during winter to determine pregnancy.

We observed and handled 6 sets of twins throughout calving, as opposed to last year when most twins were observed and captured at the beginning of the calving window (Figure 5). To more accurately represent the northeastern Minnesota population we attempted to spread our capture efforts throughout the calving season. In a previous study in northeastern Minnesota, the mean calving date was 14 May 2011 (range = 3-27 May), with 70% of births occurring during 9-20 May (McGraw et al. 2014). In 2013, we observed the mean date a cow localized (assumed to have calved within 12 hours) was 12 May (median = 13 May, range = 5-16 May), while in 2014 mean localization date was 19 May (median = 18 May, range = 5 May-16 June). The later calving dates in 2014 may be a result of the severe winter of 2013-2014 or of more intense monitoring into June 2014.

We have documented 7 mortalities to date, 6 of them due to natural causes (55% natural mortality rate). One resulted from capture-related abandonment (DelGiudice et al. this issue). Another calf died of a severe umbilical infection. Its dam was spending time between the sick calf and its healthy twin, but ultimately left the sick calf and remained with the healthy calf. Wolf predation claimed 4 calves, and a bear killed 1 calf. During 5 May to 8 June 2013, we documented 17 natural mortalities (50% mortality rate); causes included drowning, natural abandonment, and predator-kills. During May 2013-February 2014, we documented 25 natural mortalities of 34 collared calves (74% mortality, Figure 8). Predation accounted for 22 of these mortalities (88%, 4 bear-kills, 18 wolf-kills).

To date we have had 7 collars slip off in 2014. In each instance the collar was stretched and the sewn expansion loops broke prematurely. There was no tearing or blood on the collars or sign of a struggle at the collar location. Additionally, the dam did not flee the site at the time the collar entered mortality mode. This is likely a design flaw that will need to be addressed before next year’s captures. We are currently testing the resilience of the collar banding.

Body mass measurements from calves in northeastern Minnesota (mean = 16.2 kg, SE = 0.5) agree with data from healthy moose calves throughout North American moose range (13-18 kg; Jensen et al. 2013). Measurements taken in 2013 and 2014 are comparable, although the 2014 sample size was limited ( $n = 43$  versus 12). We did not detect any sexual dimorphism in calf measurements, but singletons tended to be heavier than twin calves, as reported elsewhere. However, northeastern Minnesota calves tended to be heavier than Alaskan calves (Schwartz and Hundertmark 1993).

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Table 1. Morphometric measurements ( $\pm$ SE) from neonate moose calves captured and collared in northeastern Minnesota, May 2013 and 2014.

2013	Singleton		Twin		Female		Male		Total	
	Mean	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>
Morphometric										
Mass (kg)	17.2 $\pm$ 0.6	13	15.6 $\pm$ 0.3	30	16.2 $\pm$ 0.4	22	16.2 $\pm$ 0.5	21	16.0 $\pm$ 0.3	43
Hind leg length (cm)	47.2 $\pm$ 0.4	13	45.8 $\pm$ 0.3	36	46.0 $\pm$ 0.3	25	46.4 $\pm$ 0.3	24	46.2 $\pm$ 0.2	49
Chest girth (cm)	61.4 $\pm$ 1.7	5	59.3 $\pm$ 0.9	6	60.3 $\pm$ 1.3	6	60.2 $\pm$ 0.4	5	60.3 $\pm$ 0.9	11
Lower neck circumference (cm)	32.3 $\pm$ 0.5	5	28.8 $\pm$ 0.6	6	30.1 $\pm$ 0.9	6	30.7 $\pm$ 1.0	5	30.4 $\pm$ 0.7	11
Upper neck circumference (cm)	31.0 $\pm$ 0.9	4	28.3 $\pm$ 1.2	6	29.5 $\pm$ 1.3	6	29.3 $\pm$ 1.3	4	29.4 $\pm$ 0.9	10
Body length (cm)	106.7 $\pm$ 1.4	3	-	-	105.5 $\pm$ 1.5	2	109	1	106.7 $\pm$ 1.4	3

2014	Singleton		Twin		Female		Male		Total	
	Mean	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>
Morphometric										
Mass (kg)	16.8 $\pm$ 0.9	6	15.7 $\pm$ 0.4	6	16.0 $\pm$ 0.8	6	16.4 $\pm$ 0.7	6	16.2 $\pm$ 0.5	12
Hind leg length (cm)	43.6 $\pm$ 0.5	6	43.2 $\pm$ 1.0	5	42.8 $\pm$ 0.7	6	44.2 $\pm$ 0.6	5	43.4 $\pm$ 0.5	11
Chest girth (cm)	51.5	1	65	1	51.5	1	65	1	58.3 $\pm$ 6.8	2
Lower neck circumference (cm)	33.6 $\pm$ 0.8	6	34.5 $\pm$ 0.9	6	33.1 $\pm$ 0.8	6	35.0 $\pm$ 0.7	6	34.0 $\pm$ 0.6	12
Upper neck circumference (cm)	29.0 $\pm$ 0.5	6	30.8 $\pm$ 0.8	6	28.7 $\pm$ 0.5	6	31.1 $\pm$ 0.6	6	29.9 $\pm$ 0.5	12
Body length (cm)	-	-	101.0	1	-	-	101.0	1	101.0	1

57 Collar 12629 U // Localized

Last location: 2014-05-25 02:51:04  
 Max 3-h Avg. Speed (24h) = 11.7 m/h  
 Collar Temp = 20°C // Mortality Status =  
 UTM.X = 650291, UTM.Y = 5291498  
 Longitude = -90.99453, Latitude = 47.75932

Stat	T2h	T24h	T48h	Total
Disp.	9	12	4	3407
Path	36	156	314	436741
Speed	9	8	8	62

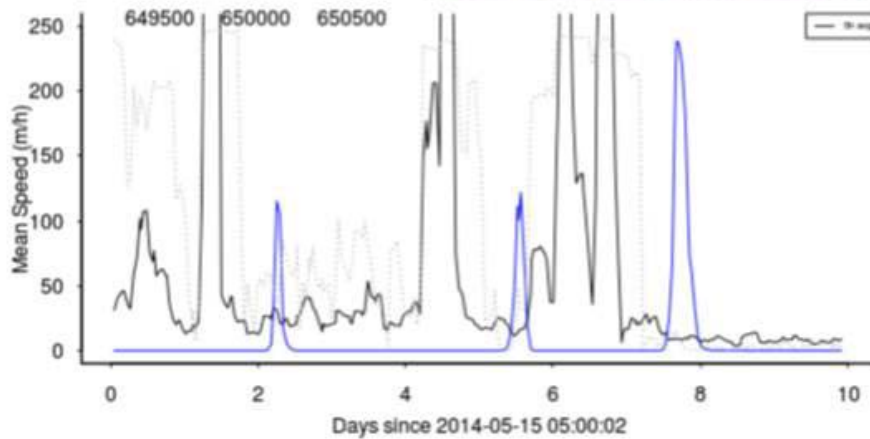


Figure 1. Example report for adult female moose number 12629 from 0500 hours, northeastern Minnesota, 25 May 2014, showing movement paths for the last 5 days and 24 hours, and 3-hour average hourly distances moved. Green circle represents the start of the 5-day period, green triangle the start of the 24-hour period, and red triangle the most recent location. Red dots indicate location when the collar was “localized.” The blue line represents predictions from a regression based on 2013 calving movement data; larger spikes suggest a higher likelihood a calving movement occurred in the past 12 hours. The light gray lines show relatively large or small movements in the past 12 hours. We visited this cow at 10 days since 15 May (25 May).

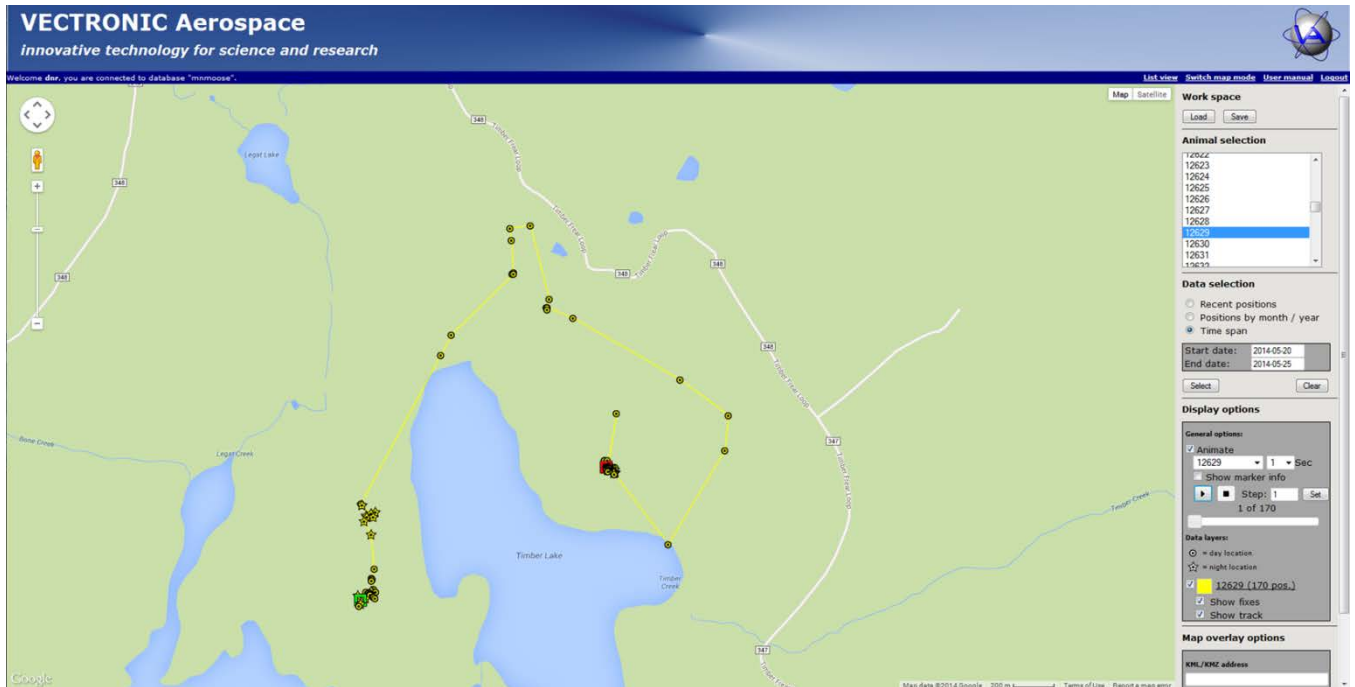


Figure 2. Vectronic Aerospace website (<https://www.vectronic-wildlife.com>) map interface showing the path of adult female moose number 12629, northeastern Minnesota, 20–25 May 2014. The green and red squares represent the start and end of the interval. The cow's movement pattern in the southwestern corner of the map indicates typical bedding and foraging, whereas the cluster in the middle of the map indicates a tight localization which followed a long-distance movement. This cluster is the calving ground and where calf 13253 was collared.

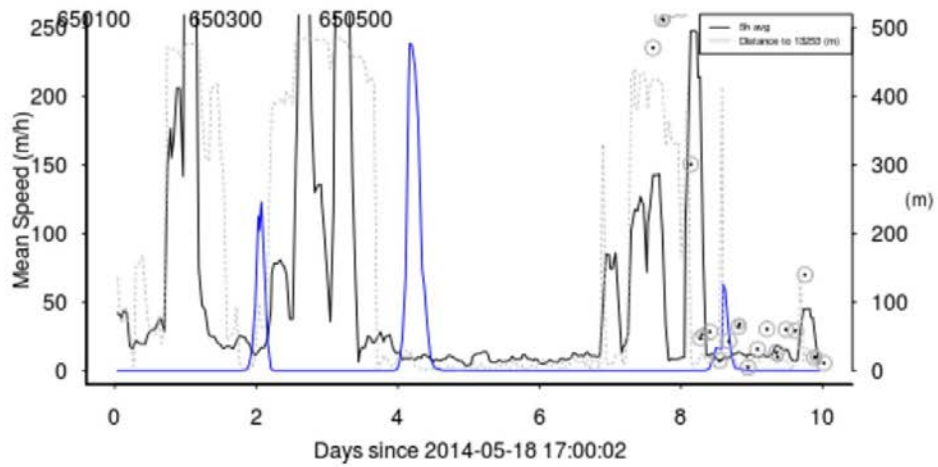


Figure 3. Distance plot displaying both 3-hour average distance moved and proximity of adult female moose number 12629 to calf number 13253, northeastern Minnesota. Black solid line displays the distance the dam has moved; circles with centered dots represent the distance between the dam and calf collar.

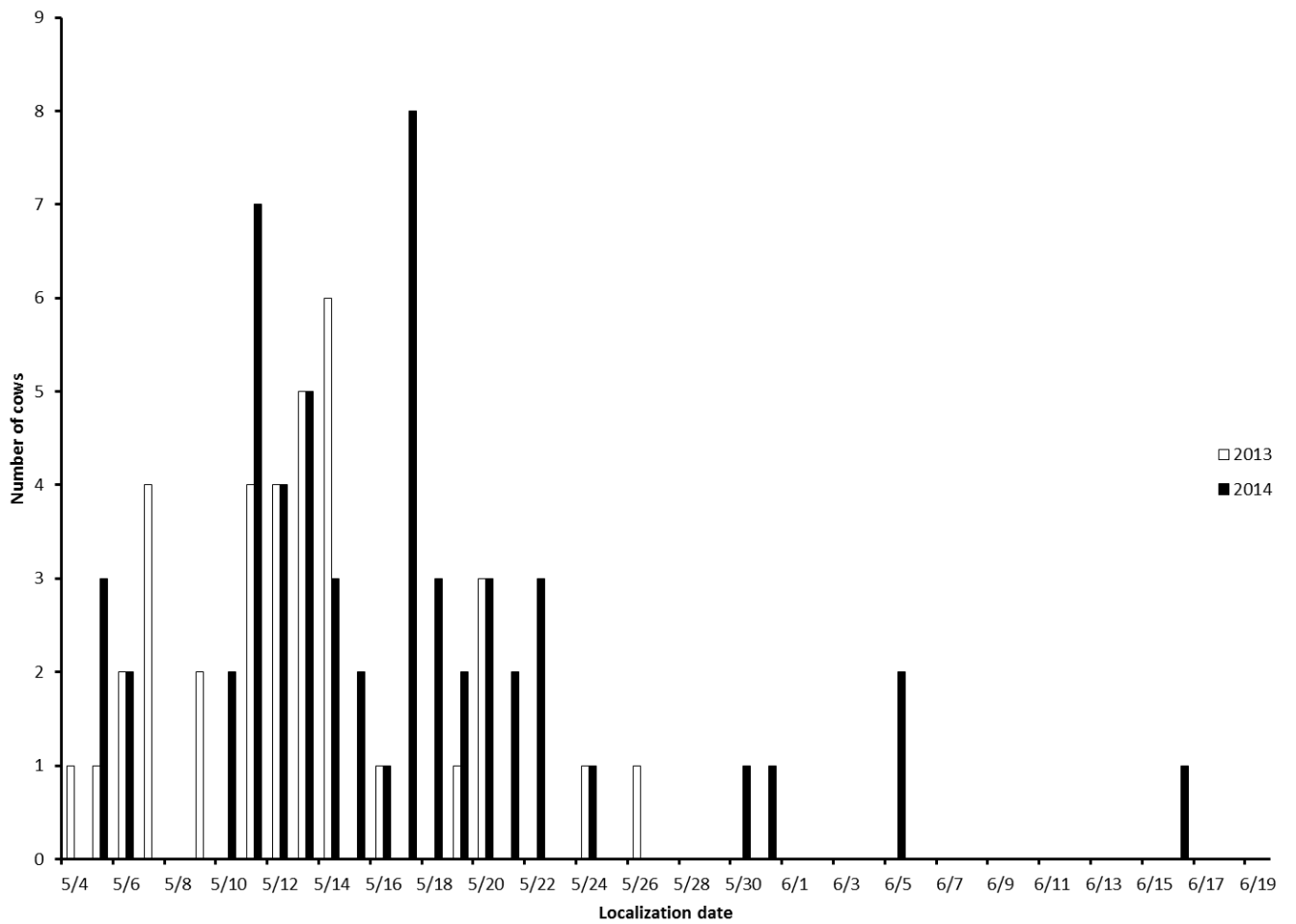


Figure 4. Temporal distribution of calving localizations of collared adult female moose, northeastern Minnesota, May-June 2013 and 2014.



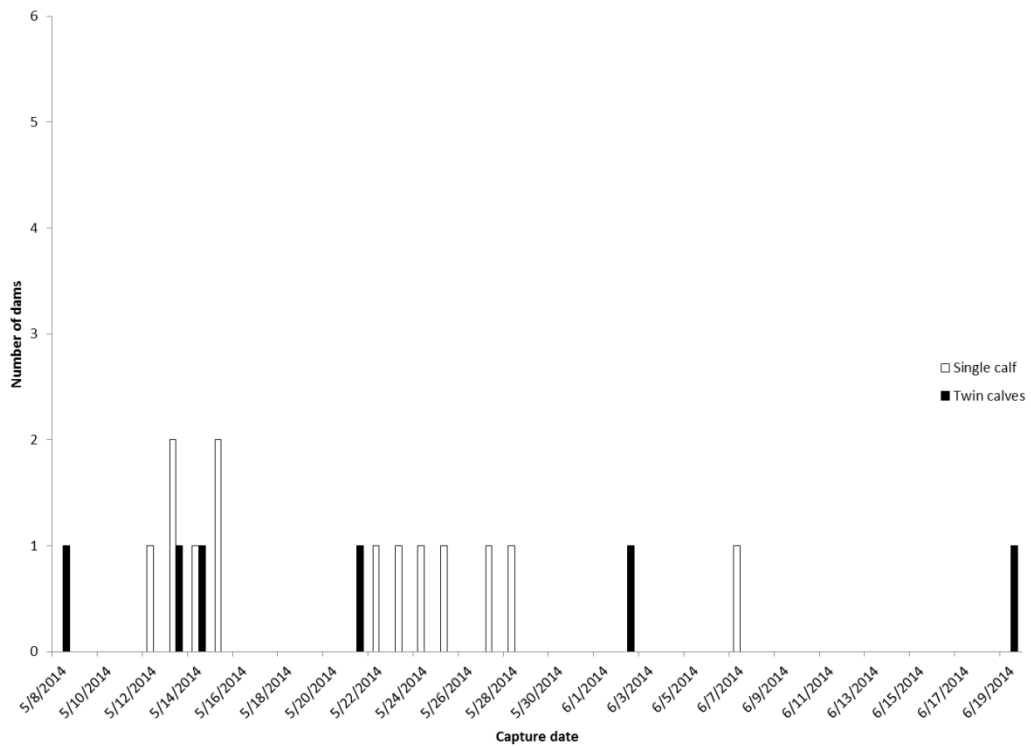
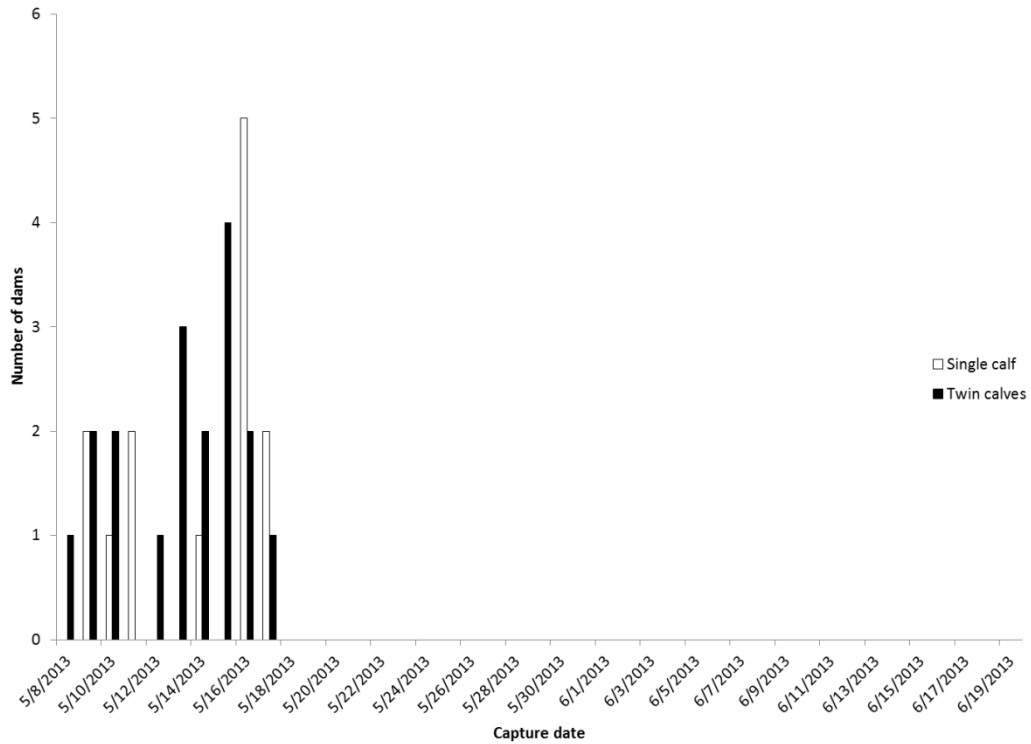


Figure 5. Number of moose dams with single and twin calves captured and handled, northeastern MN, 8-17 May, 2013 (top) and 8 May-19 June 2014 (bottom).

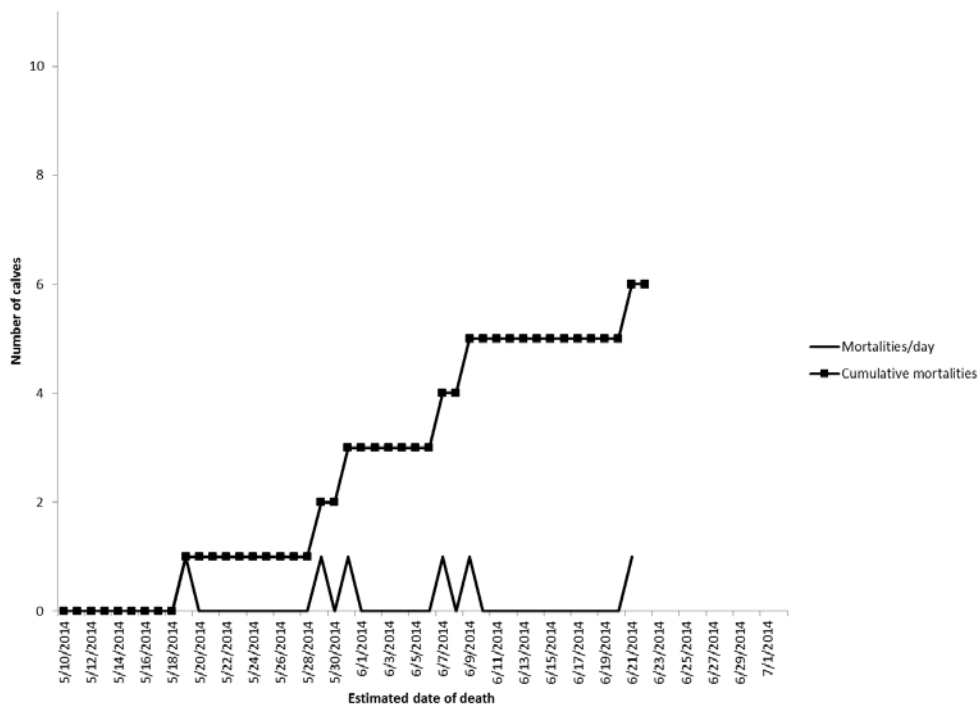
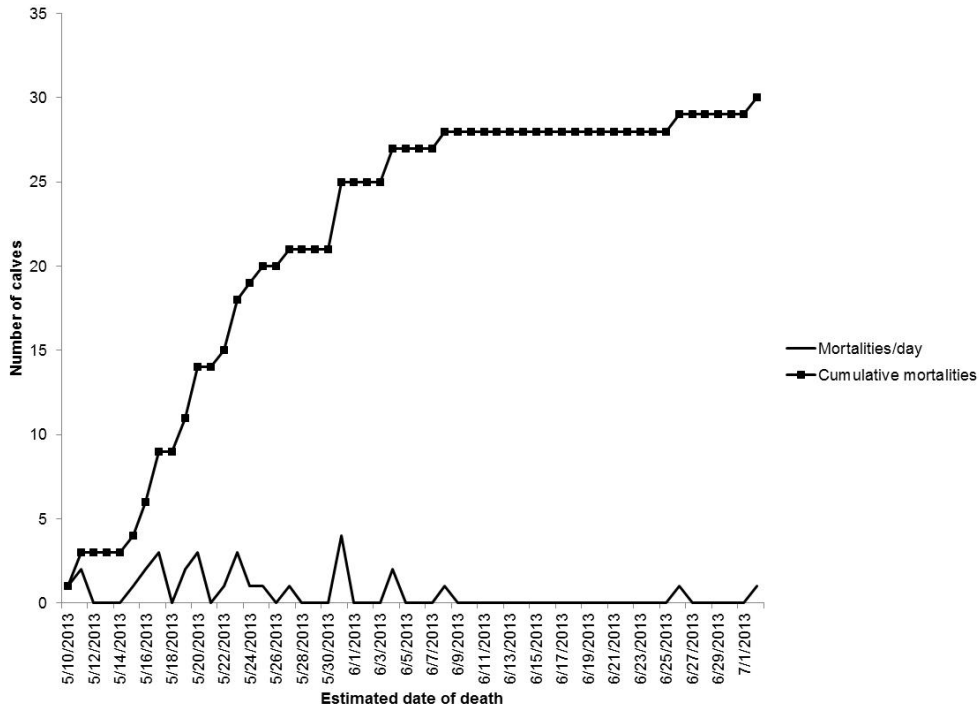


Figure 6. Number of natural mortalities of moose calves by day and cumulative mortality by day, northeastern MN, 10 May – 2 July 2013 (top) and 10 May-9 June 2014 (bottom).

### Kaplan-Meier

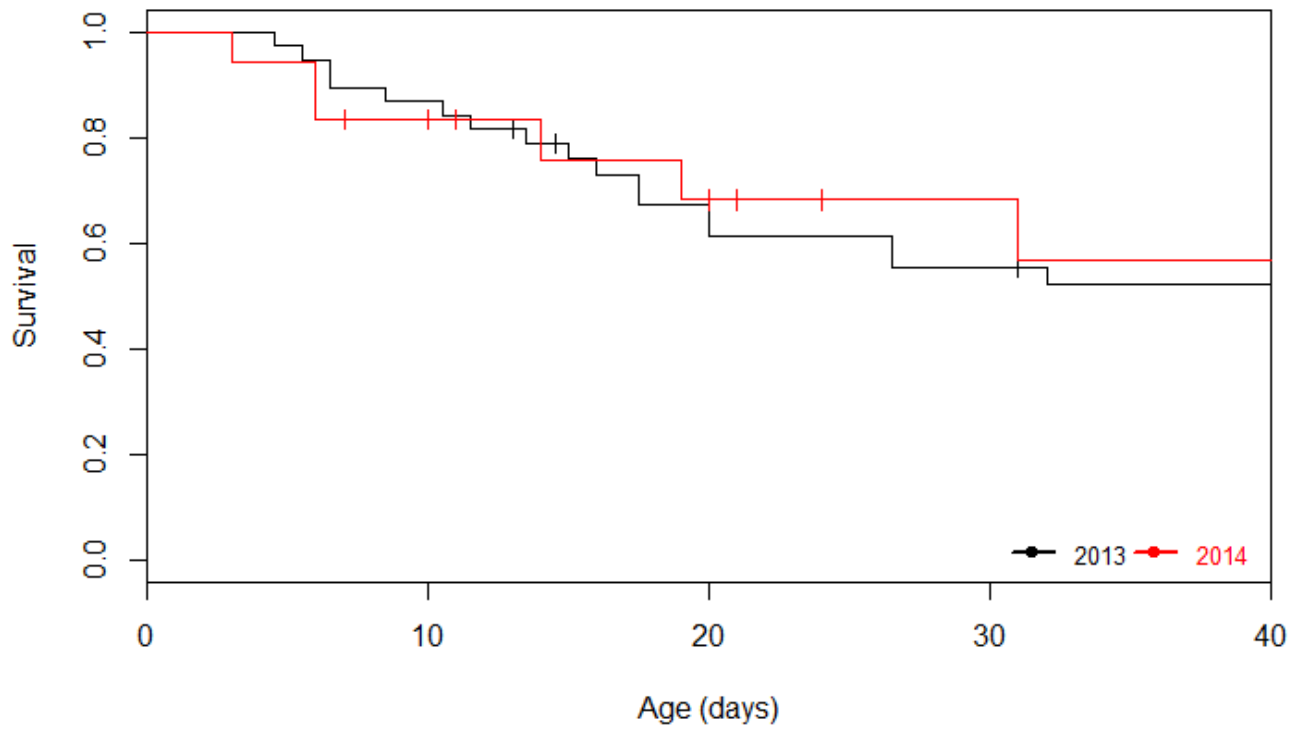


Figure 7. Kaplan-Meier survival curves plotting natural mortality for moose neonates (0-40 days) in northeastern Minnesota, 2013 ( $n = 34$ ) and 2014 ( $n = 11$ ). Plus symbols (+) indicate slipped (censored) collars.

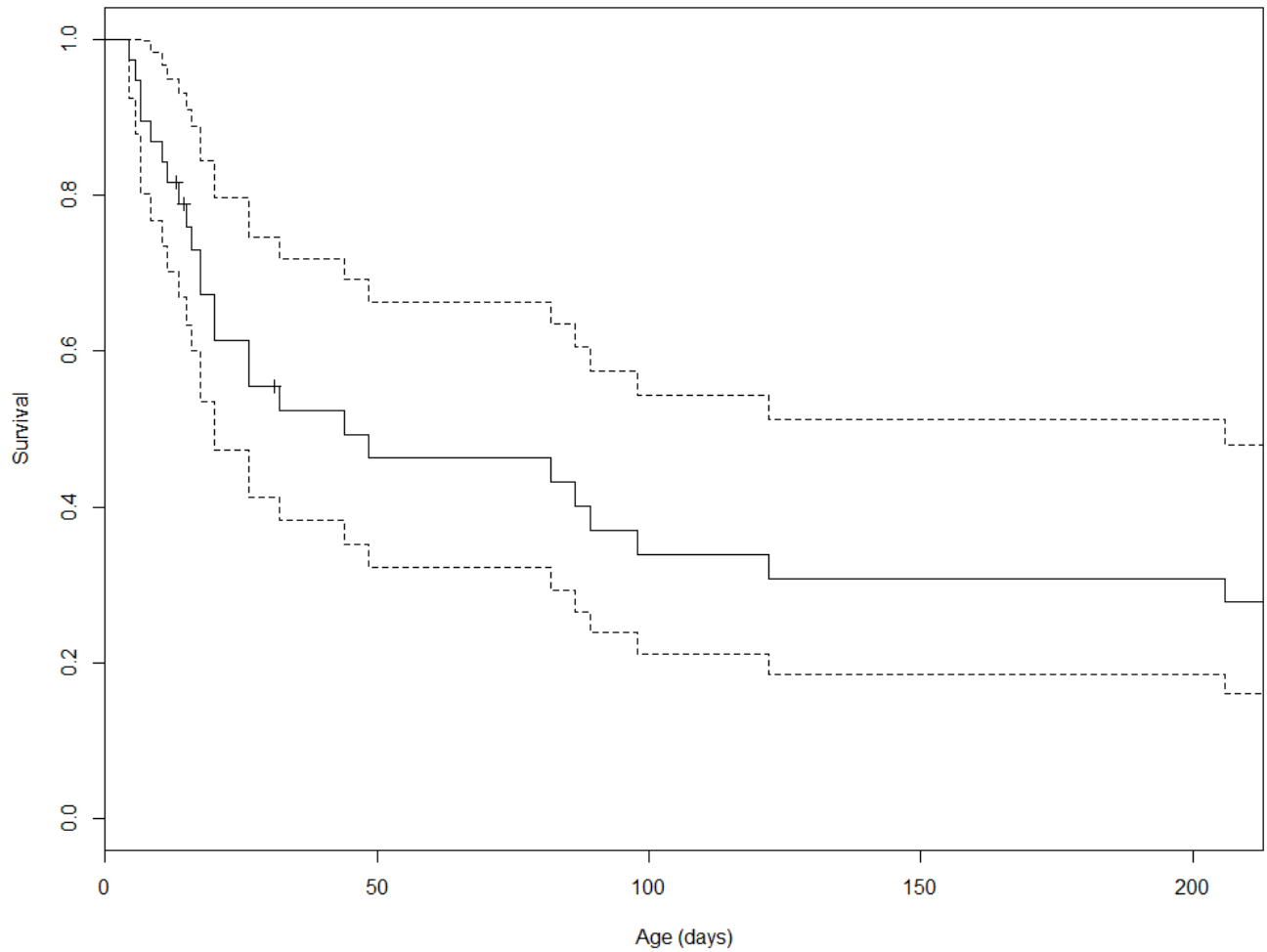


Figure 8. Kaplan-Meier survival curve ( $\pm 95\%$  confidence intervals) plotting natural mortality for moose calves (0-230 days) in northeastern Minnesota, 2013 ( $n = 34$ ). Plus symbols (+) indicate slipped (censored) collars.

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## SUMMARY OF FINDINGS

During April 2013–March 2014, we monitored 24 radiocollared black bears (*Ursus americanus*) at 4 study sites representing contrasting portions of the bear's geographic range in Minnesota: Voyageurs National Park (VNP, northern extreme, poorest food), Chippewa National Forest (CNF; central), Camp Ripley (southern fringe), and a site at the northwestern (NW) edge of the range. Most of the focus of this study has been in the NW site in recent years. This area had the highest reproductive rate of our 4 study sites, due to an abundance of both agricultural crops and natural foods. Our longest span of reproductive data (1981–2014) is in the CNF, where we observed some evidence that litter size may have increased, with a concomitant increase in cub mortality; however, this inference is limited by small sample size in recent years. Hunting has been the primary source of mortality in all areas; but with a concerted effort to discourage hunters from shooting collared bears, and by clearly marking bears with large ear tags, no collared bears that we knew of were killed by hunters in fall 2012 or 2013 (although 2 collared bears could not be found). The oldest bear in this study, which was the oldest known wild bear of any species, died naturally at 39 years old.

## INTRODUCTION

Telemetry-based research on black bears was initiated by the Minnesota Department of Natural Resources (MNDNR) in 1981, and has been ongoing since then. Objectives shifted over the years, and study areas were added to encompass the range of habitats and food productivity across the bear range. For the first 10 years, the bear study was limited to the Chippewa National Forest (CNF), near the geographic center of the Minnesota bear range (Figure 1). The CNF is one of the most heavily hunted areas of the state, with large, easily-accessible tracts of public (national, state, and county) forests dominated by aspen (*Populus tremuloides*, *P. grandidentata*) of varying ages. Camp Ripley Military Reserve, at the southern periphery of the bear range, was added as a second study site in 1991. The reserve is unhunted, but bears may be killed by hunters when they range outside, which they often do in the fall. Oaks (*Quercus* sp.) are plentiful within the reserve, and cornfields border the reserve. Voyageurs National Park (VNP), at the northern edge of the Minnesota range (but bordering bear range in Canada) was added as a third study site in 1997. Soils are shallow and rocky in this area, and foods are generally less plentiful than in the other sites. Being a national park, it is unhunted, but like Camp Ripley, bears may be hunted when they range outside.

In 2007 we initiated work in a fourth study site at the northwestern edge of the Minnesota bear range (henceforth NW; Figure 1). This area differs from the other 3 areas in a number of respects: (1) it is largely agricultural (including crop fields, like corn and sunflowers, that bears consume), (2) most of the land, including various small woodlots, is privately-owned, with some larger blocks of forest contained within MDNR Wildlife Management Areas (WMAs) and a National Wildlife Refuge (NWR); (3) the bear range in this area appears to be expanding and bear numbers have been increasing, whereas most other parts of the bear range are stable or declining in bear numbers; and (4) hunting pressure in this area is unregulated (it is within the no-quota zone, so there is no restriction on numbers of hunting licenses).

## OBJECTIVES

1. Quantify temporal and spatial variation in cub production and survival;
2. Assess bear mortality in different parts of the bear range.

## METHODS

We previously attached radiocollars with breakaway and/or expandable devices to bears either when they were captured during the summer or when they were handled as yearlings in the den with their radiocollared mother. We used VHF collars in CNF, Camp Ripley, and VNP, and GPS collars in the NW study site. We used aerial telemetry to locate den sites. During December–March, we visited all radio-instrumented bears once or twice at their den site. We immobilized bears in dens with an intramuscular injection of Telazol, administered with a jab stick or dart gun. Bears were then removed from the den for processing. We measured and weighed them, assessed body condition, and took blood and hair samples. We changed or refit the collar, as necessary. All collared bears had brightly-colored, cattle-size ear tags (7x6 cm; Dalton Ltd., UK) that would be plainly visible to hunters.

Six bears were implanted in December dens with a new Insertable Cardiac Monitor developed for human heart patients (Reveal LINQ™, Medtronic Inc., Minneapolis, MN). The device is small enough (4.0 x 7.2 x 44.8 mm; 2.4 grams) to be injected subcutaneously in a left peristernal location. Surgical sutures were used to close the puncture site. The device provided wireless transmission of heart and activity data to an antenna buried under the nest material in the den, which was then relayed by cell phone to a base station. Besides providing physiological information, the heart rate and activity data also yielded birth dates of cubs, signaled by significantly increased heart rates and activity (just prior to birth), followed by a rapid decline to lower than pre-birth levels, which was maintained the rest of the denning period.

We assessed reproduction by observing cubs in March dens. We sexed and weighed cubs without drugging them. We evaluated cub mortality by examining dens of radiocollared mothers the following year: cubs that were not present as yearlings with their mother were presumed to have died.

We periodically monitored survival of bears during the summer. Mortalities also were reported to us when bears were shot as a nuisance, hit by a car, or killed by a hunter. Prior to the hunting season (1 September–mid-October), hunters were mailed a letter requesting that they not shoot collared bears with large ear tags.

## RESULTS AND DISCUSSION

### Radiocollaring and Monitoring

As of April 2013, the start of the current year's work, we were monitoring 24 radiocollared bears: 4 in the CNF, 8 at Camp Ripley, 3 in VNP, and 9 in the NW (Table 1). We did not trap any new bears this year, but we collared 2 yearlings in dens, and 1 new bear was found and collared in a den at Camp Ripley. Since 1981 we have handled >800 individual bears and radiocollared >500, 361 of which were followed until they died (Table 2).

### Reproduction

Six collared females produced cubs in 2014. All bears maintained a 2-year reproductive cycle. All 11 collared females that had cubs in 2013 had surviving yearlings in this winter's den. Based on heart rate data from 3 pregnant females with implanted wireless cardiac devices, the following birthdates were discerned: 2, 7, and 9 January 2014.

Since 1982, within the 4 study areas, we have checked 275 litters with 703 cubs ( $\bar{x} = 2.6$  cubs/litter), of which 52% were male (Tables 3–6). Mortality of cubs during their first year of life averaged 21%, with mortality of male cubs (25%) exceeding that of females (16%;  $\chi^2 = 5.73$ ,  $P < 0.02$ ). The timing and causes of cub mortality are unknown.

Reproductive rates (cubs/female 4+ years old: combining litter size, litter frequency, and age of first reproduction into a single parameter) were highest in the NW study area, and lowest

in VNP (Figure 2). This corresponds with food conditions. The NW site contains not only agricultural crops consumed by bears, but also an abundance of natural foods, especially along the edges of woodlots. Reproductive rate was higher for 7+ year-old bears than 4–6 year-old bears because many bears in this younger age group either had not yet reproduced or just had their first litter, which tended to be smaller. Reproductive rates for 7+ year-old bears in the CNF and Camp Ripley were similar, although Camp Ripley bears tended to mature earlier (Figure 2). Reproductive rates for older bears at Camp Ripley appeared to decline this year because all of them had cubs last year and were with yearlings this year.

No strong trends in reproduction were evident through time. In the CNF, where we have been monitoring reproduction since 1982 (Table 3), we have observed a general increase in litter size ( $\bar{x}$ : 1982–1992=2.5; 1993–2003=2.7; 2004–2014=3.0 cubs/litter; Figure 3). Concomitantly, cub mortality has increased ( $\bar{x}$ :1982–1992=16%; 1993–2003=18%; 2004–2014=30%). Higher cub mortality may, in part, be related to a higher production of male cubs (Figure 3); however, female cub mortality in the CNF has increased dramatically, averaging only 11% during 1982–2003 but jumping to 28% since 2004 (nearly equal to males'). Sample sizes have declined in recent years (Figure 3), however, hampering interpretations of these data.

Notably all litters that we monitored in the last 10 years in the CNF have had at least 3 cubs (Table 3). We are cautious about inferring too much from these data, though, due to a small number of collared bears in the CNF: since 2009, we have only been monitoring 2 mature females (14 and 19 years old in 2014), neither of which has ever produced a 2-cub litter, and both of which recently produced 4-cub litters (one in 2010 and one in 2011). Only 1 other bear in this study (none from study areas other than CNF) have produced 5 or more consecutive litters of 3 or more cubs (while they were radio-collared). This goes to show that individual variation among females is important in reproductive output. The 3 most prolific bears were not unusually heavy (160–210 pounds in March dens at 7+ years old).

## Mortality

Legal hunting has been the dominant cause of mortality among radiocollared bears from all study sites (Table 2). Recently, hunters seemed to have respected our request not to shoot collared bears (with large eartags): none were known to have been shot during either 2013 or 2014. However, we could not locate 2 collared bears when we searched for their den, leaving the possibility that they were killed and not reported. No bears were known to have died from any other human-related cause this year.

One particularly small adult female bear (143 pounds in March 2013 at 8 years old) in VNP lost its collar, which we found tangled in boughs of a fallen balsam fir tree (*Abies balsamea*) when we attempted to locate its den. It is unusual that a collar of an adult female could be pulled off, suggesting that the bear had been losing weight, and may have died. We could not search the site for remains because it was covered in deep snow.

The oldest bear on our study, a 39-year-old female in the CNF (as of January 2013) died this year (July 2013). This bear, identified as #56, was first trapped and radio-collared at 7 years old in 1981, the first summer of our research. From assessment of her cementum annuli, we concluded that the cubs with her at the time were from her second litter. We monitored her from then until her death, 31 years later.

Bear #56 was the only collared bear in our study that died of old age. She outlived the next longest survivor by 16 years. To our knowledge, she was older than any known-age bear of any species in the wild. Few wild bears reach even 30 years old; one Alaskan brown bear was reported to live to 34 years old (Schwartz et al. 2003). Among the >60,000 teeth that the MDNR has collected and aged from hunter-killed bears since 1975, only 3 (0.005%) lived past 30 years; the oldest appeared to be 33 when it was shot, however interpretation of cementum annuli in teeth of very old bears is imprecise.

The longevity of bear #56 is likely attributable to a combination of factors, including the

location of her home range in a forested area with relatively few people or major roads and a general wariness of people — she was never reported in a U.S. Forest Service campground within her home range, apparently did not visit hunters' baits, and was rarely caught during 10 years of our trapping efforts that included her home range. Additionally, her long life, and our chance to witness it, certainly entailed a bit of luck. Her last years of life were prolonged by having brightly-colored eartags and radiocollar, and our request to hunters not to shoot her.

The last time bear #56 was handled, in March 2010, her body condition was good, but her teeth showed excessive wear, with active gum disease (Figure 4), and both eyes were clouding. By that summer it was clear that her hearing was poor. Her eyesight and hearing continued to deteriorate in ensuing years. This enabled us to approach her and observe her occasionally (Figure 5), although she never became human-habituated (she would flee upon becoming aware of our presence). She was observed by local people with increasing frequency foraging along trails and traveling on forest roads, likely because travel was easier there. Despite her deteriorating mobility, in 2011, bear #56 travelled over 20 miles outside her home range to feed on acorns along the Prairie River for several weeks in late summer.

During her last few years, her notoriety helped keep her alive. Knowing of her advanced age and physical impairments, residents often reported her whereabouts and tolerated her odd behavior. One local resident reported her in their yard and inquired whether she was deaf, as she did not respond to their shouting and clapping. In at least one case a hunter passed up his chance to shoot a bear when he learned that it was bear #56 visiting his bait site.

Bear #56 was a key animal during the many years of our research in the CNF study area. She and her offspring provided an almost uninterrupted record of reproduction, survival, movements, and eventually, senescence within a single matriarchal lineage over a 32-year span. During 1981–1995, bear #56 produced 8 litters, totaling 22 cubs, and successfully reared a remarkable 21 of them to 1½ years, the age of family breakup. Signs of reproductive senescence started to appear when she uncharacteristically lost 2 of the 3 cubs born in 1997. In 1999, at age 25, she produced and raised only 1 cub. The last evidence of any reproductive activity occurred in 2001, when, despite being in excellent nutritional condition in her den, we found her lactating, but alone.

For the next 10 years, 2001 – 2010, bear #56 maintained body mass and condition, though by 2010 (weighing 192 pounds in March) she had declined somewhat from her peak weight (255 pounds in March). During her last 2 summers, we observed her on several occasions walking unsteadily, but she was nevertheless able to walk at a pace through the forest faster than we could keep up. Sometime in July, 2013, she moved several miles west of her normal summer home range and died, apparently non-violently of natural causes. When we found her remains, they were already well-decayed but mainly undisturbed (Figure 6).

We collected her skeleton as a reference specimen. It exhibited extensive bone loss in the skull and mandibles around the teeth and advanced osteoarthritis, particularly along the spine. The skull was scanned and a model recreated for posterity (Figure 7). One daughter, now 19 years old, remains collared in the CNF.

## **ACKNOWLEDGMENTS**

We thank the collaborators in this study: Brian Dirks (MDNR) at Camp Ripley; Paul Iazzo (University of Minnesota) and Tim Laske (Medtronic, Inc.) in the NW and CNF study sites; and Steve Windels and Bryce Olson (National Park Service) in Voyageurs National Park. Agassiz NWR kindly provided use of their bunkhouse and assistance during the winter fieldwork.



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Table 1. Fates of radiocollared black bears in 4 study sites (Chippewa National Forest, Camp Ripley, Voyageurs National Park, and northwestern Minnesota), April 2013–March 2014.

	CNF	Camp Ripley	VNP	NW
Collared sample April 2013	4	8	3	9
Killed as nuisance				
Killed in vehicle collision				
Killed by Minnesota hunter				
Natural mortality	1			
Dropped collar		1	1	
Failed radiocollar				
Lost contact <sup>a</sup>			1	1
Collared in den	1	1		1
<b>Collared sample April 2013</b>	<b>4</b>	<b>8</b>	<b>1</b>	<b>9</b>

<sup>a</sup> Due to radiocollar failure, unreported kill, or long-distance movement.

Table 2. Causes of mortality of radiocollared black bears  $\geq 1$  year old in 4 Minnesota study sites, 1981–2014. Bears did not necessarily die in the area where they usually lived (e.g., hunting was not permitted within Camp Ripley or VNP, but bears were killed by hunters when they traveled outside these areas).

	CNF	Camp Ripley	VNP	NW	All combined
Shot by hunter	223	11	15	12	261
Likely shot by hunter <sup>a</sup>	8	1	0	4	13
Shot as nuisance	22	2	1	3	28
Vehicle collision	12	8	1	3	24
Other human-caused death	9	1	0	0	10
Natural mortality	8 <sup>b</sup>	3	5	0	15
Died from unknown causes	4	2	0	3	9
<b>Total deaths</b>	<b>286</b>	<b>28</b>	<b>22</b>	<b>25</b>	<b>361</b>

<sup>a</sup> Lost track of during the bear hunting season, or collar seemingly removed by a hunter.

<sup>b</sup> Only 1 bear died of “old age”.

Table 3. Black bear cubs examined in dens of radiocollared mothers in or near the Chippewa National Forest during March, 1982–2014. High hunting mortality of radiocollared bears severely reduced the sample size in recent years.

Year	Litters checked	No. of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 yr <sup>a</sup>
1982	4	12	3.0	67%	25%
1983	7	17	2.4	65%	15%
1984	6	16	2.7	80%	0%
1985	9	22	2.4	38%	31%
1986	11	27	2.5	48%	17%
1987	5	15	3.0	40%	8%
1988	15	37	2.5	65%	10%
1989	9	22	2.4	59%	0%
1990	10	23	2.3	52%	20%
1991	8	20	2.5	45%	25%
1992	10	25	2.5	48%	25%
1993	9	23	2.6	57%	19%
1994	7	17	2.4	41%	29%
1995	13	38	2.9	47%	14%
1996	5	12	2.4	25%	25%
1997	9	27	3.0	48%	23%
1998	2	6	3.0	67%	0%
1999	7	15	2.1	47%	9%
2000	2	6	3.0	50%	17%
2001	5	17	3.4	76%	15%
2002	0	0	—	—	—
2003	4	9	2.3	22%	0%
2004	5	13	2.6	46%	33%
2005	6	18	3.0	33%	28%
2006	2	6	3.0	83%	33%
2007	2	6	3.0	67%	17%
2008	1	3	3.0	100%	33%
2009	1	3	3.0	33%	33%
2010	1	4	4.0	100%	50%
2011	1	4	4.0	25%	50%
2012	1	3	3.0	67%	33%
2013	1	3	3.0	67%	0%
2014	1	3	3.0	67%	0%
<b>Overall</b>	<b>179</b>	<b>472</b>	<b>2.6</b>	<b>53%</b>	<b>19%</b>

<sup>a</sup> Cubs that were absent from their mother's den as yearlings were considered dead.

Table 4. Black bear cubs examined in dens in northwestern Minnesota during March, 2007–2014.

Year	Litters checked	No. of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 yr
2007	2	6	3.0	33%	100%
2008	5	15	3.0	67%	22%
2009	1	3	3.0	33%	33%
2010	6	17	2.8	41%	13%
2011	2	4	2.0	75%	25%
2012	4	10	2.5	60%	10%
2013	3	9	3.0	67%	18%
2014	3	8	2.7	0%	0%
<b>Overall</b>	<b>26</b>	<b>72</b>	<b>2.8</b>	<b>48%</b>	<b>26%<sup>a</sup></b>

<sup>a</sup> Excludes the total loss of a 5-cub litter in 2007 (which was not within the designated study area).

Table 5. Black bear cubs examined in dens in or near Camp Ripley Military Reserve during March, 1992–2014.

Year	Litters checked	No. of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 yr <sup>a</sup>
1992	1	3	3.0	67%	0%
1993	3	7	2.3	57%	43%
1994	1	1	1.0	100%	—
1995	1	2	2.0	50%	0%
1996	0	0	—	—	—
1997	1	3	3.0	100%	33%
1998	0	0	—	—	—
1999	2	5	2.5	60%	20%
2000	1	2	2.0	0%	0%
2001	1	3	3.0	0%	33%
2002	0	0	—	—	—
2003	3	8	2.7	63%	33%
2004	1	2	2.0	50%	—
2005	3	6	2.0	33%	33%
2006	2	5	2.5	60%	—
2007	3	7	2.3	43%	0%
2008	2	5	2.5	60%	0%
2009	3	7	2.3	29%	29%
2010	2	4	2.0	75%	25%
2011	3	8	2.7	50%	25%
2012	1	2	2.0	100%	0%
2013	6	14	2.3	50%	21%
2014	1 <sup>b</sup>	— <sup>b</sup>	—	—	—
<b>Overall</b>	<b>40</b>	<b>94</b>	<b>2.4</b>	<b>52%</b>	<b>21%</b>

<sup>a</sup> Blanks indicate no cubs were born to collared females or collared mothers with cubs died before the subsequent den visit to assess cub survival.

<sup>b</sup> Cubs heard, litter not handled. This litter not included in total.

Table 6. Black bear cubs examined in dens in Voyageurs National Park during March, 1999–2014. All adult collared females were killed by hunters in fall 2007, so no reproductive data were obtained during 2008–2009.

Year	Litters checked	No. of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 yr <sup>a</sup>
1999	5	8	1.6	63%	20%
2000	2	5	2.5	60%	80%
2001	3	4	1.3	50%	75%
2002	0	—	—	—	—
2003	5	13	2.6	54%	8%
2004	0	—	—	—	—
2005	5	13	2.6	46%	20%
2006	1	2	2.0	50%	0%
2007	3	9	3.0	44%	—
2008	0	—	—	—	—
2009	0	—	—	—	—
2010	1	2	2.0	50%	0%
2011	1	2	2.0	0%	0%
2012	1	2	2.0	0%	50%
2013	1	2	2.0	50%	—
2014	1	3	3.0	33%	—
<b>Overall</b>	<b>29</b>	<b>65</b>	<b>2.2</b>	<b>48%</b>	<b>27%</b>

<sup>a</sup> Blanks indicate no cub mortality data because no cubs were born to collared females, or collared mothers were lost from study (died or lost collar) before denning with yearlings.

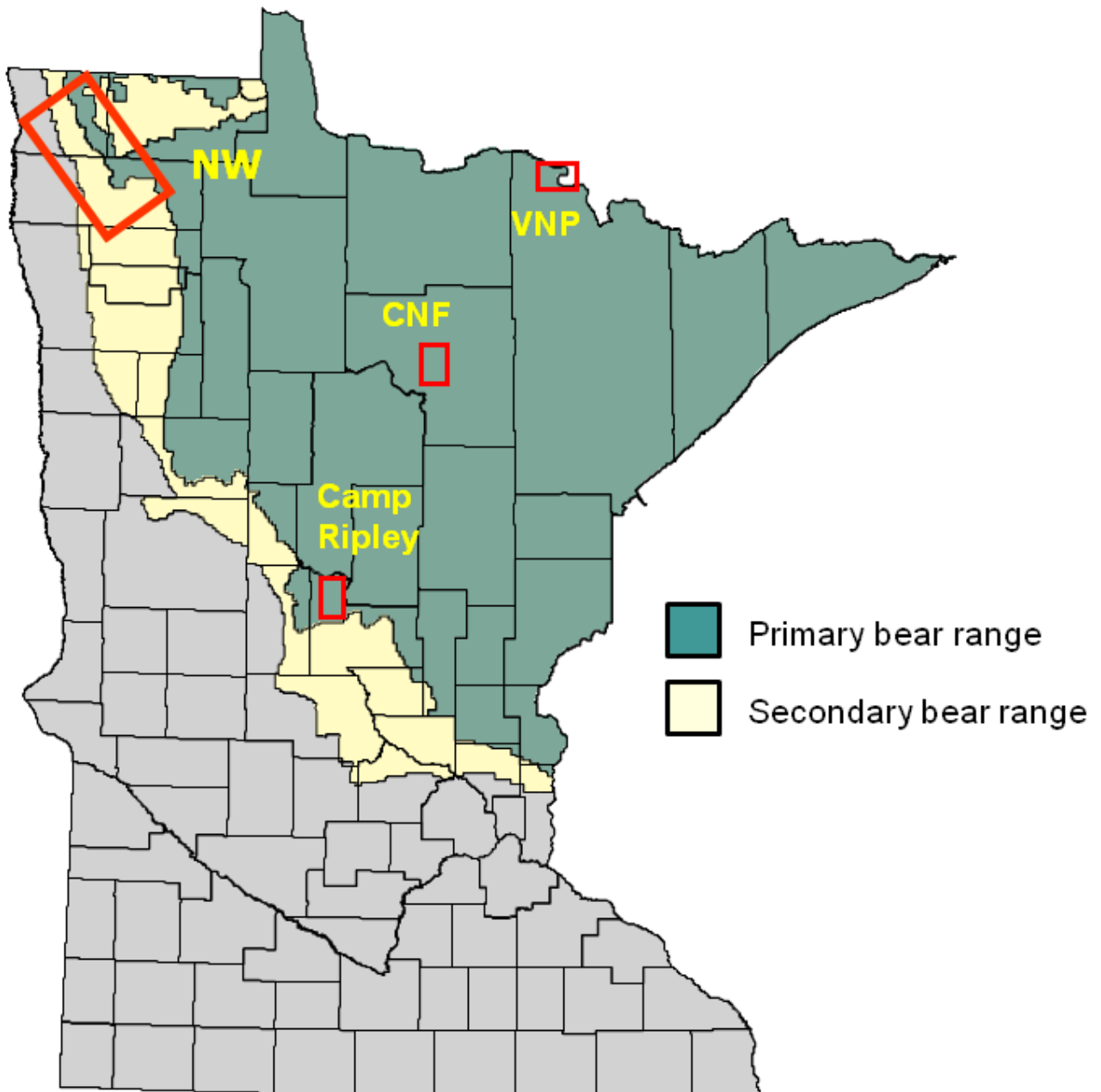


Figure 1. Location of 4 study sites within Minnesota's bear range: CNF (Chippewa National Forest, central bear range; 1981–2014); VNP (Voyageurs National Park, northern fringe of range; 1997–2014); Camp Ripley Military Reserve (near southern edge of range; 1991–2014); NW (northwestern fringe of range; 2007–2014).

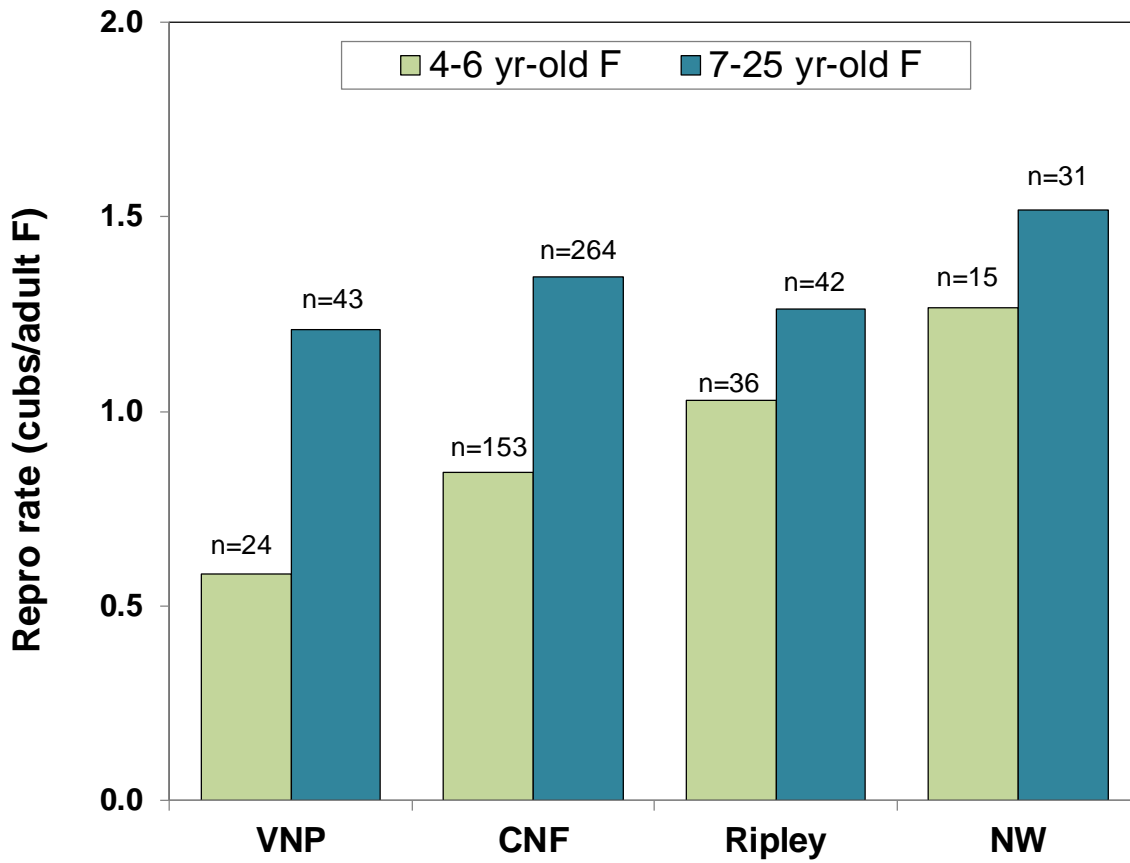


Figure 2. Reproductive rates of radiocollared bears within 4 study sites (see Figure 1) through March 2014. Sample sizes refer to the number of female bear-years of monitoring in each area for each age group. Data include only litters that survived 1 year (even if some cubs in the litter died). Some bears in CNF, Camp Ripley, and NW produced cubs at 3 years old, but are not included here.

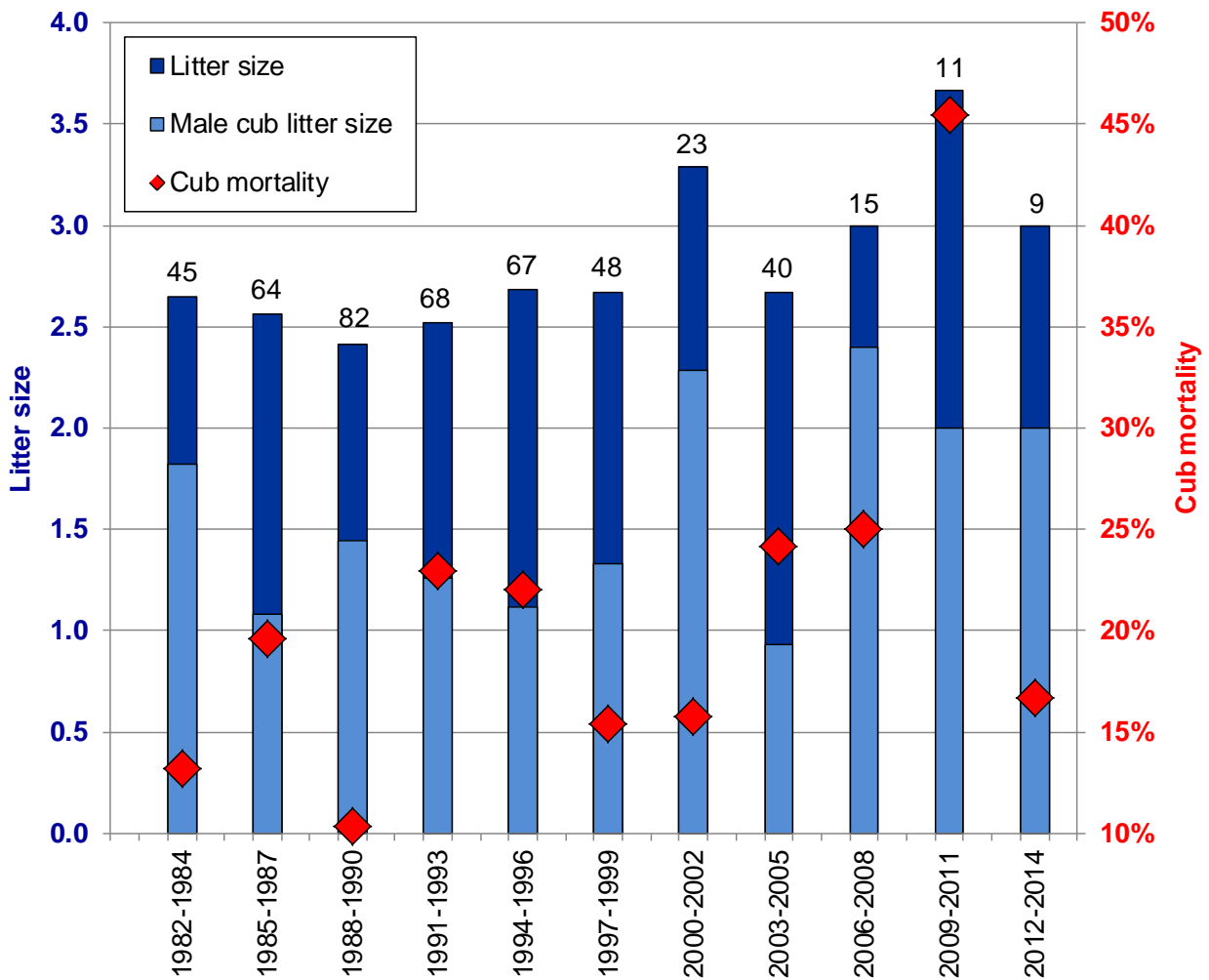


Figure 3. Cub litter sizes (examined in dens in March) in the CNF appeared to increase through time (1982–2014), with more males per litter. Correspondingly, cub mortality has also trended upward (assessed by examining yearlings in dens of radio-collared mothers that had cubs the year before). Each bar represents 3 years of combined data, with number of cubs indicated above bars. Note that secondary Y-axis begins at 10%.

Figure 4. Deteriorating teeth condition of bear #56 the last time she was handled (March 2010) at 36 years old. Note missing left lower canine and 2 missing lower incisors, as well as gum disease.

Figure 5. Despite poor teeth, bear #56 nevertheless foraged on natural foods throughout her life (left, spring 2011). However, her dulled senses made her more easily observable (right, within 1 month of death at age 39). When she did become aware of our presence, she quickly moved away (she was not human-habituated). The colored collar and eartag, made her more visible to hunters, who avoided shooting her at our request, enabling her to live to very old age.



Figure 6. Bear #56 was found dead in August, 2013, at 39 years old. The remains as found (shown unaltered in photo) indicate that she died naturally, and was not preyed upon by another bear or wolves.



Figure 7. The skull of bear #56, the oldest recorded wild bear of any species, was CT-scanned (top) and a model produced with a 3-D printer (bottom right) to preserve for posterity, as the original skull (bottom left) is quite fragile due to extreme wear and degeneration.

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### SUMMARY OF FINDINGS

During the spring and summer of 2012, we conducted genetic capture–mark–recapture (CMR) of black bears in the Chippewa National Forest (CNF) using hair traps in order to ascertain changes in bear abundance since our last estimates of this same population during the 1980s and 1990s. Because previously we captured bears in physical traps or by camera traps, one objective here was to examine potential sources of bias specifically related to hair-trapping, which could hinder comparisons with previous estimates. We set 121 2-stranded barbed wire hair traps in the same study area as used in the 1980s and 1990s. We checked sites during 6 sampling sessions at 10-day intervals during late-May to mid-July. Visitation by bears was high (55% of site-session checks), yielding 2784 hair samples in 1642 clusters of 1–11 adjacent barbs. We assumed that clusters of barbs with hair represented places where a bear went over or under a wire and rubbed across several barbs. We submitted a sample from each of 1113 clusters for genetic analysis, and 1019 (92%) were successfully genotyped. We examined data for evidence of capture heterogeneity due to annual molt and barbed wire position. In nearly two-thirds of the clusters, hair occurred only on the lower strand of barbed wire, suggesting that most hair was snagged when bears crawled under the lower wire. Hair occurred on both upper and lower wires in 22% of clusters and only on upper wires in 16% of clusters. Samples from both males and females were more common on lower wires, but male hair was relatively more common than female hair on upper wires, suggesting that 2-strand designs may better capture population diversity than a single strand of barbed wire, as used in many studies. The total number of sites visited by bears per sampling session increased from the first to second session, and was relatively consistent thereafter. The number of barbs and clusters with hair at each site, however, declined through the study, as did the relative frequency of clusters involving both wires, suggesting that hair was harder to snag during later sessions because the long winter coat had been molted and the new summer coat was harder to pluck out. Awareness of these trends will help inform modeling and interpretation of CMR population estimates. Further work with camera traps at hair snares is needed to ascertain whether cubs are sampled by hair snaring, an important consideration in comparing this estimate to previous estimates that excluded cubs.

### INTRODUCTION

During summer 2012 we conducted DNA-based capture–mark–recapture (CMR) of black bears to estimate bear population size in a 300-km<sup>2</sup> study area in northern Itasca County, Minnesota, mainly within the Chippewa National Forest. We specifically sought to examine population change since the 1980–1990s, when we last estimated abundance on the same study area using physical trapping and camera trapping. Multiple lines of evidence suggested that bear numbers had declined significantly since our last estimates.

DNA-based CMR, using hair collected from barbed-wire hair traps, has become the “gold standard” for estimating bear population size where logistics and budgets allow. Genetic CMR has many advantages over marking bears through physical captures and radiocollaring. Because bears are not handled, checking hair traps requires a lower level of skill and less equipment. Also, more traps can be set because they do not have to be checked daily, and bears likely have less aversion to the traps, so are more likely to be recaptured. Thus, sample sizes from hair trapping are apt to be large (improving precision) and less biased. However, hair trapping potentially introduces new sources of sampling heterogeneity that are unique to this method. Hence it was important to consider potential forms of bias that may have differed from our physical mark-recapture work.

Most hair trapping studies have employed a barbed wire enclosure constructed by

stretching a single strand of barbed wire around several trees at a height of about 50 cm above the ground. A scent lure, hung over the center of the enclosure but out of reach of bears, entices bears into the enclosure (Woods et al. 1999). This design was developed originally for grizzly bears (*Ursus arctos*) and was intended to snag hair from all sizes of bears – i.e., the barbed wire was high enough to sample large bears that climbed over the wire, but still low enough to sample smaller bears that crawled underneath, including cubs of the year (Woods et al. 1999, Kendall et al. 2008). Other protocols made use of hair left at trees (Kendall et al. 2008) or power poles (Karamanlidis 2007) where bears naturally rub, scratch, and mark. In this case, barbed wire is secured to the tree or pole to increase the amount of hair sampled when bears rub.

Each of these methods has some degree of bias with regard to size and/or sex of bears sampled (Kendall et al. 2008; Boulanger et al. 2006). Where both methods have been employed, as in Banff National Park, Alberta, Canada, hair traps caught relatively few male grizzly bears, whereas at rub trees, males were more likely to be captured than females (Sawaya et al. 2012).

This method has been adapted for use on black bears in locations around North America, with some modifications. Researchers found that often, bears entered hair traps but did not leave hair on the barbed wire. To increase capture efficiency, Tredick et al. (2007) added a lower wire to their hair traps (20 cm above ground) to force animals to squeeze through more restricted spaces and improve the detection of small bears that could otherwise crawl under wires without leaving hair. However, they concluded that samples from the lower wire were of poor quality and thus did not increase efficiency enough to offset the extra expense and time of construction. Lowe (2011), however, working in south central Louisiana, suspected that large males might be systematically under-represented using 1-strand hair traps; photos from remote cameras showed large males entering enclosures by stepping on or over wires and leaving no hair (Hooker 2010). Estimated capture rate for females was twice as high as for males. Subsequent work in the same study area, using hair traps with an added upper wire at 70 cm resulted in more equal estimates of capture probability for males and females (O’Connell 2013).

Other potential problems stem from the timing of genetic CMR studies, which often run through much of the spring and summer. In temperate regions, this coincides with a bear’s annual molt. In May and June, when most underfur and guard hair are lost, shedding hair is easy to snag, whereas the hairs of new pelage are more tightly bound, so significant changes in capture probability may occur among sampling sessions. If the molt proceeds differently for different sex-age groups, this further complicates the task of categorizing capture heterogeneity and accounting for it in analysis of data.

In our CMR study, we wished to sample all age groups except cubs of the year, because our previous estimates (physical capture and camera capture) excluded cubs. A pilot project suggested that a 2-strand hair trap with wires placed at approximately 45 and 75 cm above the ground would be the most likely to catch bears of all sizes except cubs. Here we examine patterns in the hair we captured so as to discern types and magnitude of sampling bias that barbed wire hair traps introduced into our population estimation procedure.

## **OBJECTIVES**

1. Determine if hair snaring introduces significant sampling bias and/or heterogeneity into CMR sampling, and if so, what type(s).
2. Determine if cubs of the year were sampled by hair snares.

## METHODS

The study area was same CNF study site where previous CMR estimates were obtained (Figure 1). It contains good access via 2 main paved roads, smaller unimproved roads, and forest trails. Ownership is mainly national and state forest, with additional county and private lands.

We erected hair-snare traps using 2 strands of 4-pronged barbed wire wrapped around 3-5 trees, forming an enclosure. Barbed wire was placed at 45 cm and 75 cm off the ground (Figure 2). We erected 1 trap in each of 121 square-mile sections (121 mi<sup>2</sup>). Within each of these grid cells, we set a trap in what we perceived as good bear habitat to maximize visitation. We set traps at least 100m from main roads, but often along trails that bears might use.

We suspended a bag of bacon and a scent lure from a string (above the reach of a bear) across the middle of each trap, and put bait and scent lure on a pile of brush in the middle of the enclosure (Figure 2). Baits and lures were refreshed at each trap visit. We added different types of lures at each trapping session to maintain novelty for the bears. We checked all traps 6x at intervals of 10 days. We did not move traps between sessions. At each trap check, all bear hair was removed from the wire. Each clump of hairs on a barb was collected in a separate envelope, and labeled as to proximity to other barbs with hair, trap number, and date. We coded barbs of hair that were adjacent (next to, on either the same wire or the one above/below) as being from the same cluster. A cluster could include only a single barb.

We set camera traps at some of the hair traps that were visited by bears to gauge whether cubs of the year left hair on wires, and to assess the responses of different bears to the wires and the baits. Hair traps were erected the third week of May, 2012. We checked all 121 hair traps 5 times (605 site-sessions), then dismantled 36 traps that were never visited by a bear, leaving 85 to be checked in session 6 and removed the third week of July.

Hair samples were submitted to Wildlife Genetics International (Nelson, British Columbia, Canada) for genotyping. As our budget was not sufficient to analyze all collected hair samples, we subsampled the collection. In subsampling we made an attempt to maximize the number of different bears that visited the sites, so (1) we included at least 1 sample from each site-session with hair, and (2) we did not submit hairs from multiple barbs within the same cluster. We also submitted hair samples from 4 radiocollared bears and their current offspring living on the study area (collected during den visits) to determine whether they visited the hair traps. The lab also identified likely family groups.

## RESULTS

Bears visited 101 of the 121 hair trap sites, resulting in 377 of 690 (55%) total site-sessions yielding hair (Table 1). We collected hair from 2784 barbs that occurred in 1642 separate clusters of 1–11 adjacent barbs. For genotyping, we initially chose (randomly) 1 barb from each of the 377 site-sessions with hair. We then chose additional random samples from among the remaining 1265 barb clusters.

Of 1113 samples that were analyzed, 14 appeared to be mixtures of >1 bear and 80 failed to amplify. Thus 1019 samples (92%) were successfully genotyped; these were from 96 different sites and 333 site-sessions. Genotyping identified 43 different individuals: 26 males and 17 females. Individual bears were detected up to 132 times each and up to 32 times in a single sampling session. Sex ratio of individuals visiting hair traps was heavy to males in all sampling sessions and did not vary through time ( $\chi^2=0.96$ ,  $df=5$ ,  $P=0.97$ ).

Females that visited hair traps did not differ from males in either the number of sessions in which they were detected (Figure 3;  $\chi^2=2.52$ ,  $df=5$ ,  $P=0.77$ ) or the number of sites they visited (Figure 4;  $\chi^2=0.83$ ,  $df=3$ ,  $P=0.84$ , for M vs.F visiting 1, 2-3, 4-7, or 8+ sites). About a third of both males (31%) and females (29%) were detected in only 1 sampling session. A similar percent of males (31%) and females (24%) were detected at only 1 site during the study. The number of sites and the number of sessions a bear was detected were positively related (Figure 5).

Camera trap photos revealed that many individuals visited the same hair trap multiple

times during a session. The same bear often entered and left a trap at different locations along its perimeter, variously crawling under, between, or over the wire (Figure 6). Multiple individuals sometimes used the same location for entering or exiting traps, conceivably producing mixed hair samples. Only 1% of analyzed samples were unusable for this reason, however, so this did not constitute a significant inefficiency in sampling.

The number of hair traps that yielded bear hair ranged from 30–79 per session (Table 1). The number of different individuals detected in a session ranged from 14–28 (Table 2). Visitation varied through time ( $\chi^2=50.3$ ,  $df=5$ ,  $P<0.0001$ ), with fewer sites visited during the first session (late May) than in subsequent sessions (25% vs. 52 – 75% of hair traps visited;  $\chi^2$  multiple comparisons for proportions;  $\alpha=0.05$ ). Visitation by bears was also higher during the last session than in sessions 2 or 3, at least in part because sites that had not been previously visited were removed for this session (Table 1).

We found that over the course of 6 sampling sessions, at sites visited by bears, the mean number of barbs/site that snagged hair, the number of clusters of barbs/site, and the number of adjacent barbs that comprised each cluster all showed significant declines (Figure 7; one-way AOV:  $F=8.69$ , 4.41, and 12.93, respectively;  $df=5.0$ ,  $P<0.001$ ). These changes, most noticeable in late June and July (sessions 4 – 6) coincided with changes observed in coat condition as bears molted (Figure 8), suggesting that hair became increasingly less likely to snag on barbs when bears entered or exited hair traps as the season progressed.

Bears left nearly twice as many hair samples on lower strands of barbed wire than on upper strands (Table 1). This was true for both males and females, although males were more likely to leave hair on upper wires only ( $\chi^2=3.9$ ,  $df=1$ ,  $P=0.047$ ) or in clusters on both wires ( $\chi^2=11.6$ ,  $df=1$ ,  $P<0.0001$ ) than were females (Figure 9). (Conversely, females were more likely to leave hair only on lower wire;  $\chi^2=19.9$ ,  $df=1$ ,  $P<0.0001$ ). Clusters of barbs included only barbs on lower wires 62% of the time, only barbs on upper wires 16% of the time, and barbs on both wires 22% of the time. Clusters that included both wires decreased from May to July (Figure 10;  $\chi^2=44.1$ ,  $df=10$ ,  $P<0.0001$ ), concurrent with an increase in the proportion left only on lower wires.

Genotyping identified several family groups that were sampled together at the same site and during the same session. Only 1 genotyped family group, a radio-collared mother and her yearlings, occurred where there was a camera. Two males also photographed at these sites were detected by hair. One camera site detected a mother with at least 1 cub present; this cub was not identified among the hair samples analyzed from the site. We had no means of ascertaining whether other detected family groups included cubs of the year.

## DISCUSSION

Capture heterogeneity is a vexing problem in capture–recapture studies of most mammals because a fundamental requirement for deriving unbiased estimates of animal numbers and demographic parameters is that individuals are equally vulnerable to capture. This is rarely the case. Recent advances in analytic methods accommodate some degree of capture heterogeneity among groups and through time, however, these methods still require classifying data such that within identifiable groups likelihood of capture is rather uniform.

Most studies involving physical capture of bears in traps exhibit a trapping bias toward males, despite a living population with more females (at least in hunted populations). Previous trapping in the Chippewa National Forest produced a male-biased capture. Trap vulnerability was also influenced by age and, for females, reproductive status (with or without cubs of the year): subadult males (3–5 years old) and females without cubs were more vulnerable to capture than other bears and juvenile females (1–2 years) and adult males (>5 years old) were less vulnerable (Noyce et al. 2001). Some of these biases shifted through time (e.g. adult males and adult females with cubs were more likely to be caught toward the end of the breeding season in early July than during peak breeding in late May – mid-June). Further compromising any assumptions of equal catchability were differences among individuals that were not sex- or

age-based, likely attributable to individual differences in behavior and/or proximity to traps,

In designing this study we attempted to lessen some of the biases apparent in our earlier capture data by doubling trap density relative to that used in the 1980s and 1990s, making it more likely that small yearling home ranges would include at least 1 hair-trap, and by running the sampling season through mid-July, to sample adult females with cubs and adult males with about the same likelihood as other demographic groups (Noyce et al. 2001). We also specifically ended sampling by mid-July to minimize violation of the assumption of geographic closure, as 40% of bears in the study area, on average (and double that some years), left their summer home range after mid-July to migrate, mainly southward, in search of concentrated food where they could fatten for winter (Noyce and Garshelis 2011). Presumably other bears from elsewhere likewise moved into the study area

Despite these efforts, hair trapping in this study still produced a male-biased capture. We could not discern age-related effects because genetic samples do not provide information about age. The fact that we did not capture a rash of new bears in session 6, yet we caught more new bears than in sessions 4 and 5 (Table 2), suggests that we ended sampling just as seasonal movements began.

Nevertheless, our analysis also suggested that hair trapping may have introduced at least 2 more sources of capture heterogeneity that are unique to this method of capture, adding further complexity to the heterogeneity already present in bait- and trap-based capture projects. As in other studies (Lowe 2011, Sawaya 2012, O'Connell 2013), evidence suggested that the number and height of barbed wire strands potentially introduces size-based differences in capture probability (thus also tied to sex and age). Large bears (most likely males) can be under-represented in studies using a single-wire placed at ~50 cm because they can step over the wire without leaving a hair sample, particularly late in the molt when belly hair is sparse (Hooker 2010). We obtained a photograph of one large bear stepping over even a 75-cm-high wire (Figure 11), though clearly at this height contact with the bear's underside was likely.

The seasonal molt causes declines in capture vulnerability, likely in all individuals, though to varying degrees. Molt varies among individuals in timing (Figure 12), and it is unclear if the timing varies among identifiable demographic groups. We suspect that some bears that visited sites late in the trapping season did not leave hair, or left insufficient hair for genotyping. We did not collect hair from barbs containing only 1 or 2 hairs, and these became increasingly common in July. Nevertheless, the genetics lab used an average of 7.2 hairs per sample, and reported to us that this accounted for the genotyping success exceeding 90% (compared to ~70% in many similar studies).

One aspect of hair trapping in black bear studies that begs more documentation is how to optimize trap design to either include or exclude cubs of the year. Depending on rates of cub mortality in a population, this age class can be up to twice the size of the yearling class. In populations such as Minnesota, where juveniles experience heavy loss each year to hunters, cubs comprise 15–20% of the population. Clearly it is important to know whether or not this age class is included in hair sampling, particularly where an objective is to enumerate the hunted population for management purposes, which, in most places (including Minnesota), does not include cubs of the year. Specific to our case here, a population decline could be obfuscated by exclusion of cubs in the 1980–1990s trap and camera-based population estimates, but inclusion of this large cohort in the recent hair-snaring estimate. Although we attempted to exclude cubs, it is possible that as cubs grew, they became more susceptible to contacting the lower wire while visiting a site with their mother. This possibility requires further investigation with camera traps.

Whereas estimators have been developed that can handle a degree of capture heterogeneity, study design should still include measures to identify likely sources of sampling bias, minimize them by adjusting study design, and, where that is difficult, assess their potential effect on resulting estimates. We plan to further explore some of these questions with better camera documentation of bear behavior at hair traps.

## ACKNOWLEDGMENTS

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Table 1. Bear hair collected at 121 barbed wire hair traps in the Chippewa National Forest during 6 sampling sessions in summer 2012.

Session	Trap sites with hair <sup>a</sup>	Total clusters <sup>b</sup> with hair	Total barbs with hair		
			Lower	Upper	Both
1	30 (25%)	149	206	92	298
2	63 (52%)	308	389	237	626
3	65 (54%)	279	318	152	470
4	79 (65%)	392	446	204	650
5	76 (63%)	303	321	127	448
6	64 (75%)	211	221	71	292
Total	377 (55%)	1642	1901	883	2784

<sup>a</sup> Each hair-snare was checked in each of sessions 1 – 5. Snares that were never visited by bears during that period ( $n = 36$ ) were dismantled prior to session 6.

<sup>b</sup> Barbs with bear hair that were adjacent to each other, either on the same or different wires, were considered the same cluster, possibly representing a single bear entering or leaving a hair snare.

Table 2. Bears detected at hair traps in the Chippewa National Forest during 6 sampling sessions, summer 2012.

Session	Dates of hair collection	Different bears detected			New bears detected
		M	F	Total	
1	25 – 31 May	7	7	14	14
2	5 – 10 June	14	9	23	13
3	15 – 21 June	15	9	24	8
4	25 – 30 June	16	12	28	3
5	5 – 10 July	15	10	25	1
6	13 – 19 July	11	10	21	4

Figure 1. Location of hair-sampling study site in Chippewa National Forest, central bear range.



Figure 2. Set-up of barbed wire hair snare, showing 2 strands of barbed wire, central pile of bait and scent, and suspended bait and scent cup.

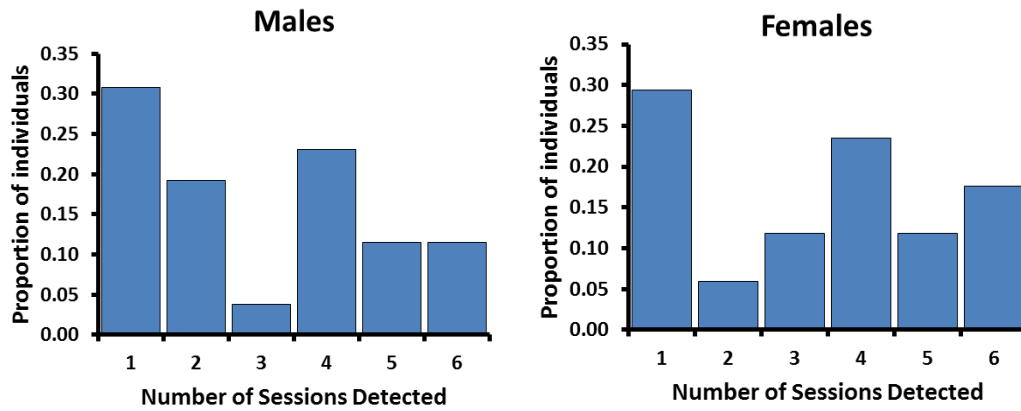


Figure 3. Number of sampling sessions in which black bears (26 M, 17 F) were detected at  $\geq 1$  hair trap during a DNA-based capture-mark-recapture study on the Chippewa National Forest, north-central Minnesota, 2012. Six 10-day sampling sessions ran from 25 May to 19 July.

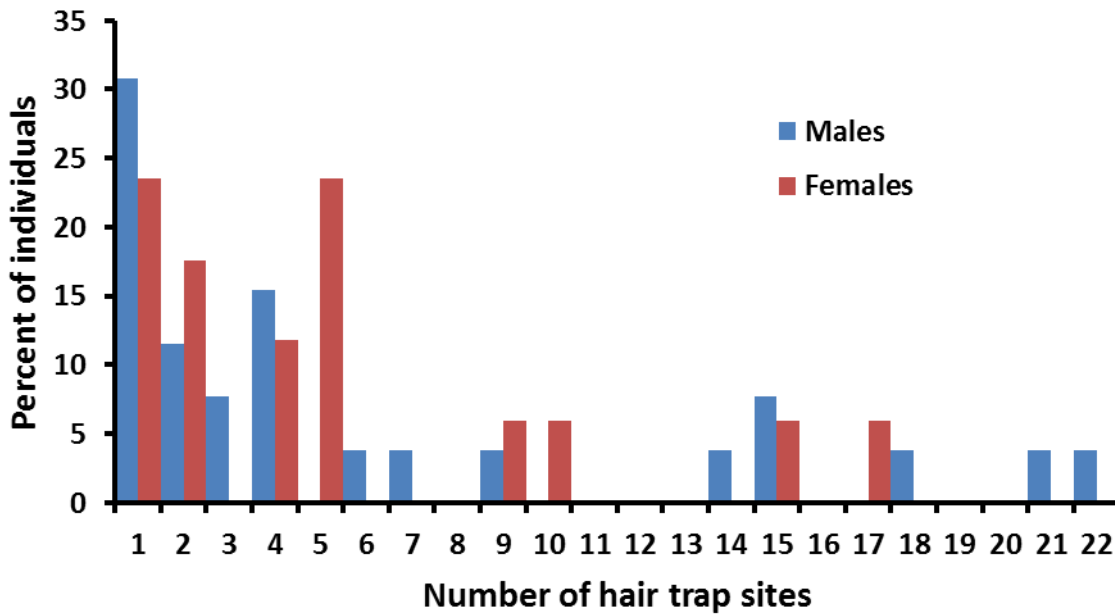


Figure 4. Number of hair trap sites at which individual male and female black bears (26 M, 17 F) were detected during 25 May – 19 July in the Chippewa National Forest, summer 2012. Hair traps were placed in a systematic grid at a density of  $1/\text{mi}^2$ .

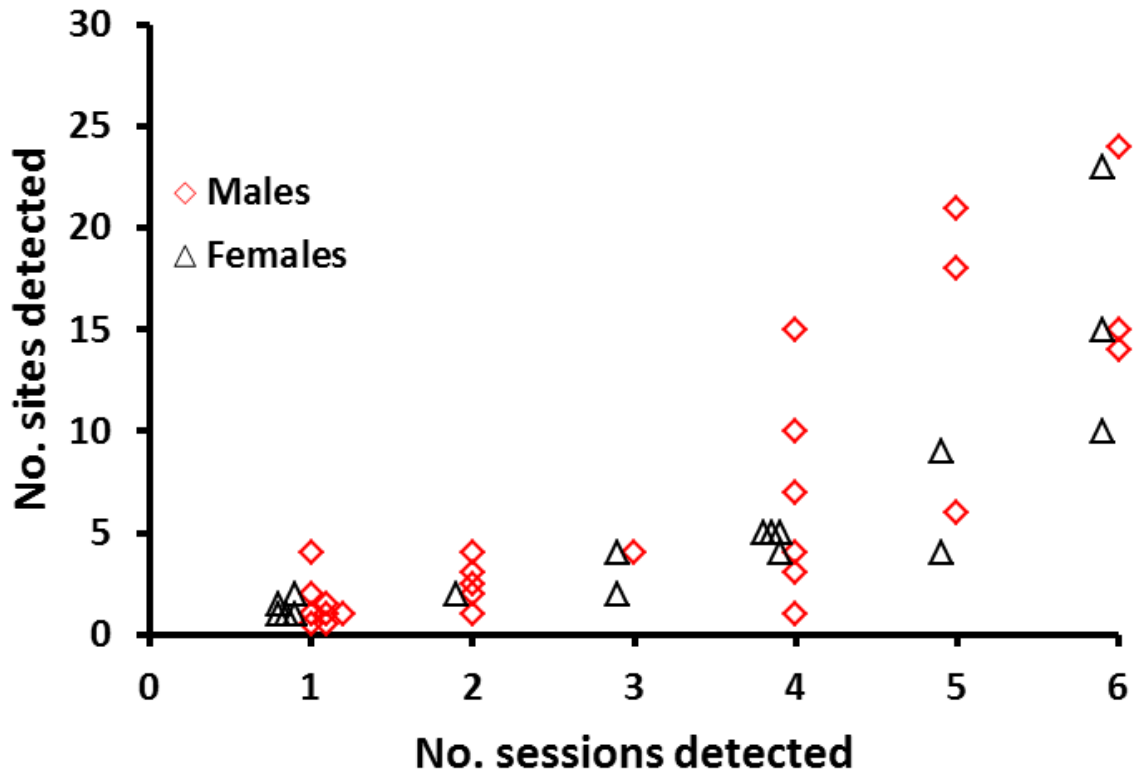


Figure 5. Relationship between the number of hair trap sites and the number of sessions in which individual male and female black bears (26 M, 17 F) were detected during 6 10-day sampling sessions in the Chippewa National Forest, north-central Minnesota, 2012.





Figure 6. Radiocollared and eartagged adult female bear entering a hair trap by going between wires (upper photo) and the same bear entering a hair trap by going below the lower wire (lower photo).

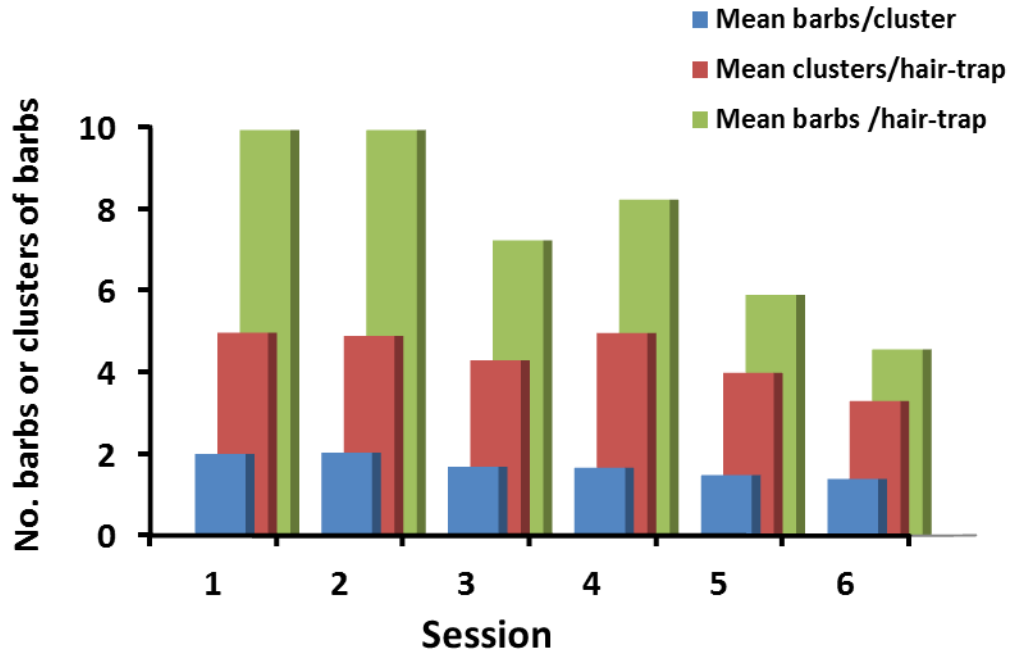


Figure 7. Changes through time in the mean number of barbs that snagged bear hair at barbed-wire hair traps (excluding sites with no hair), the number of clusters of adjacent barbs containing hair, and the number of barbs in each cluster during a DNA-based capture-mark-recapture study in the Chippewa National Forest, north-central Minnesota. Hair traps were checked 6 times, once every 10 days, from 25 May – 19 July, 2012.

Figure 8. (Left) Bear in June still wearing its long winter coat: note the clumps of matted shed hair near the rump, ready to be snagged on the barbed wire. (Right) Bear in mid-July that has nearly finished molting its winter fur: notice areas where a thin layer of longer brown hair is still present over the new black shiny summer coat (which is harder to pluck).

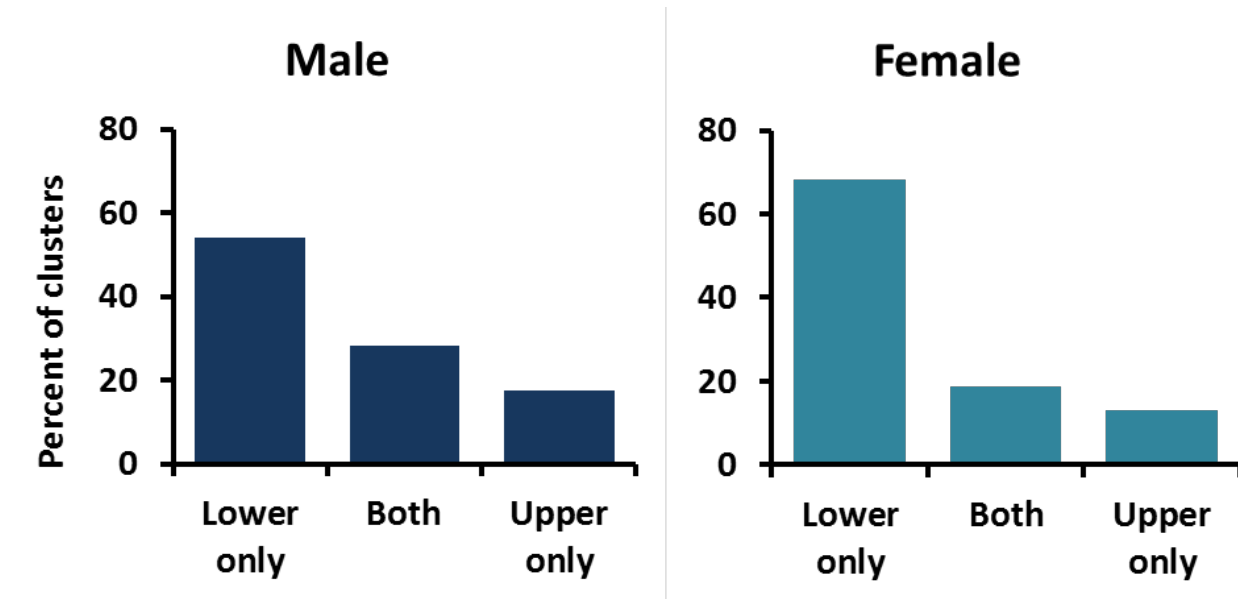


Figure 9. Percent of hair samples from barb clusters (adjacent barbs with hair) comprised of lower wires only, upper wires only, or both upper and lower wires. Only 1 barb from a cluster was genotyped, so clusters were categorized as male or female based on the 1 sampled barb (recognizing that some clusters could have included multiple bears). Males appeared more likely to leave hair on upper barbs only or both upper and lower wires than were females.

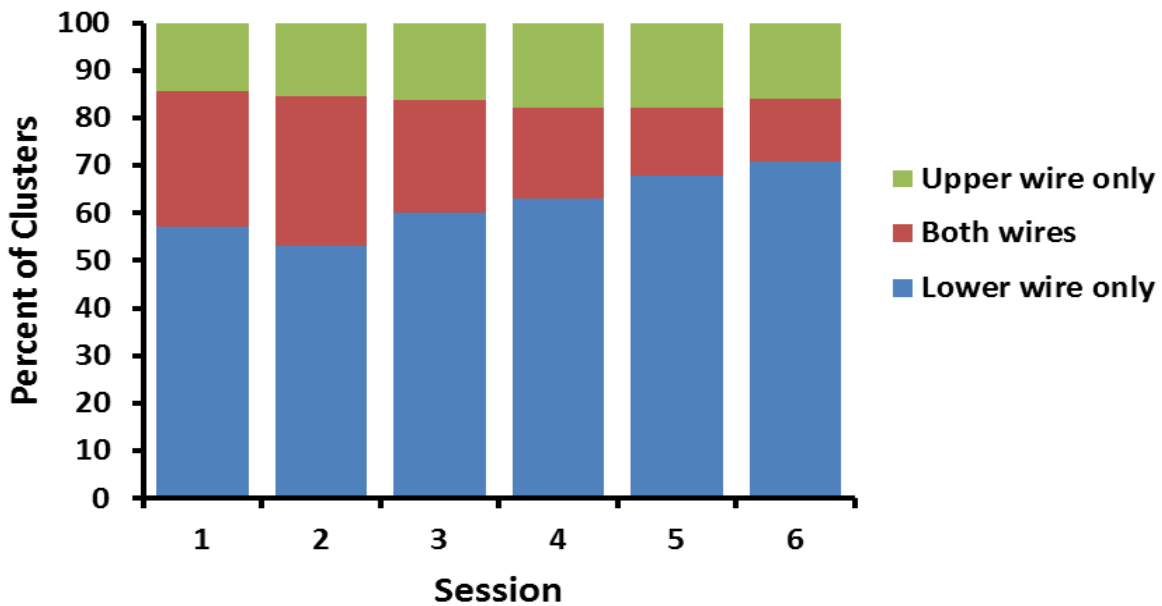


Figure 10. Changes through time in the relative proportion of bear hair samples that occurred in clusters comprised of adjacent barbs on only the lower wire, only the upper wire, or both wires of hair traps in the Chippewa National Forest, north-central Minnesota, 2012. Sampling sessions were 10 days long, spanning 25 May – 19 July.





Figure 11. Large bear stepping over the 75-cm high top wire to enter a hair trap at night. It is likely that its belly hair was caught on the wire.

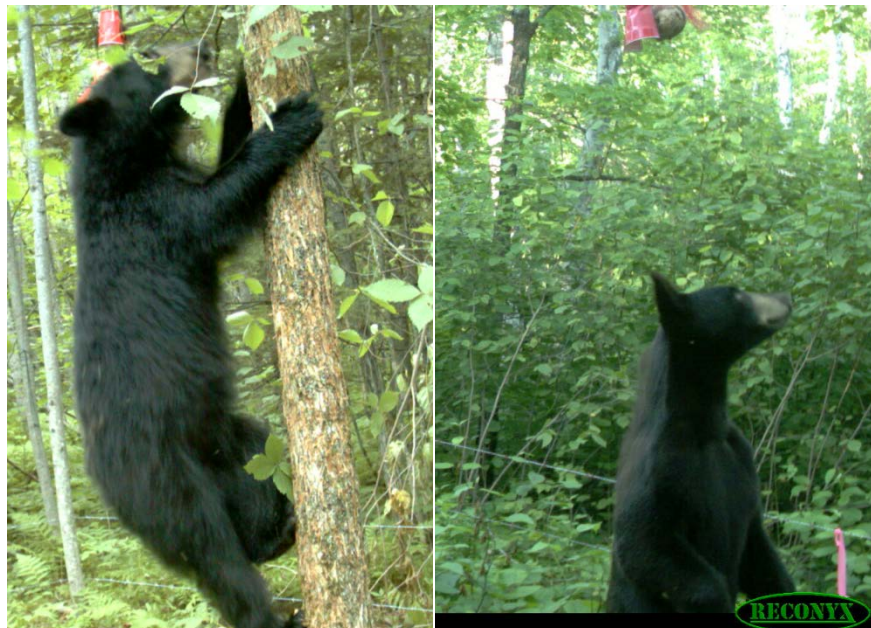


Figure 12. Two bears photographed only 1 week apart — 2 July (left) and 9 July (right) — exhibiting very different degrees of shedding.



## **DELINEATING THE ECOLOGICAL AND GEOGRAPHIC EDGE OF AN OPPORTUNIST: STRATEGIES OF THE AMERICAN BLACK BEAR (*URSUS AMERICANUS*) FOR THRIVING IN A FRAGMENTED, AGRICULTURAL LANDSCAPE**

Mark A. Ditmer<sup>1,2</sup>, David L. Garshelis, and Karen V. Noyce

### **FORAGING PATTERNS BY A FOREST-DWELLING SPECIES IN AN AGRICULTURAL LANDSCAPE: AMERICAN BLACK BEARS AT THE EDGE OF THEIR RANGE**

Human-altered landscapes, including calorie-rich agricultural crops, may promote range expansion for omnivorous, opportunist species, such as the American black bear. Black bears live in the forest, but they are known to take advantage of human-related food sources. To better understand the causes and consequences of anthropogenic foraging, we studied the diet of an expanding population of black bears at the periphery of their range in an agriculturally-dominated region of northwestern Minnesota. We estimated the dietary contributions of various types of seasonally available foods; identified associations between consumption of agricultural foods and sex, age, body size, condition and reproductive status; and assessed relationships between use of agricultural foods and natural food abundance, and the juxtaposition of these food sources on the landscape. We used stable isotope analysis ( $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$ ) of 110 hair samples from 51 bears to estimate the relative dietary contributions of early season vegetation, ants, white-tailed deer, fruit and nuts, corn, and sunflowers (Figure 1). Males and females without cubs consumed appreciable fresh corn and sunflowers in fall (95% credible intervals: males = 19–46% of diet, females = 10–40%), and remnant corn in spring/summer (males = 9–23%, females = 3–16%); females with cubs and juvenile bears relied on crops much less (Table 1). Bear consumption of these crops as judged from isotopic analysis was positively correlated with the proportion of their GPS-collar locations in corn and sunflower fields. Weight and fat thickness were also positively correlated with use of crops for both males and females (Figure 2). Females, but not males, decreased crop consumption in years with high availability of natural fall foods and during years when they were rearing cubs (Figure 2). Male bears were not strongly influenced by landscape composition or proximity of cropfields, unlike females whose agricultural consumption was more closely tied to home range size, local crop availability and landscape composition (Figure 3). Male use of crops increased with increasing physical size, suggesting that a degree of dominance and boldness was associated with the use of this high-calorie resource (Figure 2). Overall, males and females exhibited very different levels of risk-aversion, with male bears more likely to seek out anthropogenic food sources associated with a high risk of human-caused mortality. We found that two crops, corn and sunflowers, had a profound effect on the behavior of bears, despite comprising only ~2% of the land area.

### **BEHAVIORAL AND PHYSIOLOGICAL RESPONSES OF BLACK BEARS TO NATURAL AND ANTHROPOGENIC FEATURES IN AN AGRICULTURALLY-DOMINATED LANDSCAPE**

Human activities and variation in habitat quality and configuration have been shown to influence space use patterns in many species, but few studies have documented the physiological responses of free-roaming animals to these factors. We combined remote bilogger technology, capturing continuous heart rate values, with locational data from GPS collars to investigate the behavioral and physiological reactions of American black bears to a landscape dominated by agriculture (52.5% areal cover) in northwestern Minnesota. Our study occurred at the edge of the range of this species, with small, scattered patches of forest within a mosaic of crop fields and an extensive road network. However, only ~2–4% of the area

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<sup>2</sup> This report represents abstracts from three chapters of Ph.D. thesis

contained crops that bears consumed (corn, sunflowers, oats). We used GPS points to locate the habitat that bears occupied, and to estimate their rates of travel. Heart rates increased with movement rates, rising by over 30% from resting to the quickest travel speeds (Figure 4). We used a modeling approach to distinguish factors that influenced heart rates independent of movement rates. Bears commonly crossed agricultural areas that provided no food or cover and their heart rates, elevated beyond what was expected from their movement rate, were indicative of a stress response. Conversely, edible crops provided multiple metabolic benefits in the form of dense calories, reduced movement, and also reduced heart rates, suggesting that bears foraging there felt at ease. Unexpectedly, female bears had low heart rates and low levels of activity in the most fragmented patches of natural habitat, possibly a sign of human-avoidance behavior or a reaction related to crossing roads. Bears also had relatively low heart rates, given their high rate of travel, at the periphery of their home range, suggesting they were traveling but not foraging. During fall, as bears prepared for hibernation, their heart rates declined 2 weeks before their movements slackened (Figure 5, 6), evidence that their metabolism slowed to enhance fat accumulation; they also shifted to a nocturnal activity pattern (Figure 7), likely to reduce exertion during the heat of the day. The use of a physiological monitor provided new insights on bear biology and ecology that would not have otherwise been apparent.

## **DELINEATING THE ECOLOGICAL AND GEOGRAPHIC EDGE OF AN OPPORTUNIST: HOME RANGE AND CALORIC REQUIREMENTS OF AMERICAN BLACK BEARS IN A FRAGMENTED, AGRICULTURAL LANDSCAPE**

Animals living at the edge of their geographic range often exhibit exceptional physical and behavioral attributes. American black bears living in northwestern Minnesota, the western edge of the eastern black bear's geographic distribution, are the largest and most fecund bears in Minnesota despite, or potentially because of, the agricultural-dominated and fragmented area they inhabit. We estimated annual home ranges; determined how age and gender, roads, habitat, weather, crop types and caloric availability affect weekly home range size. We then used these results to delineate the limits of bear range. Annual home ranges from 60 bear-years (33 female, 27 male) using 29 individual GPS-collared bears (14 female, 15 male) showed that male ranges were 5.7–9.7x larger than females' — the home ranges of these males were the largest ever recorded for the species (Table 2). We calculated weekly home ranges, utilizing 80,441 locations from 38 individual animals (17 female, 21 male) over 75 bear-years (37 female, 38 male, Figure 8). Sizes of weekly ranges were positively associated with high road density and traffic volume, and high-levels of agriculture; they were negatively related to caloric density on the landscape. Oak habitat and crop fields of corn, sunflowers, and oats were associated with smaller home ranges during summer and fall (Figure 9). This negative correlation between amount of crops and home range size was stronger for males. Future range expansion will be dictated by food and cover. Bears required less natural cover in summer and fall months than during spring (Figure 10). Males required less natural cover, so they could potentially range farther west than females (Figure 10). Nevertheless, significant expansion appears unlikely due to limited natural cover and the distance between the natural forage available in the forests and shrublands to the east and the western periphery. Suitable female habitat contracted with consistently low annual availability of food resources, but the presence of crops does not appear to be required for their persistence in the region (Figure 11). Males and females exhibited different strategies for living in this region: males high-graded the landscape by constantly moving among patches of natural habitat during spring through mid-summer but then slowed as they sought out and consumed calorically dense crops. In contrast, females were less risk tolerant and instead sought out home ranges that provided a minimal threshold of calories from natural foods. Overall, bears' annual home range size was not a good metric of habitat quality. By investigating drivers of short-term home range size and the minimal requirements in a landscape we were able to project range limits and understand how bears thrive in a region that was once considered marginal bear habitat.

Table 1. Population estimates of diet for spring/summer and fall food sources broken down by demographic groups found to have a strong influence on individual dietary estimates of crop consumption. Annual estimates of diet from crops were calculated by multiplying lower and upper values of corn and sunflowers by 2/3rds for the spring/summer estimates and 1/3<sup>rd</sup> for the fall.

Food Source	Adult Males	Juvenile males and females	Adult females without cubs	Adult females with cubs
<b>Spring/Summer mixing model (2/3rds distal end)</b>				
Ants	0-11%	2-12%	0-13%	0-7%
Corn	9-23%	0-7%	3-16%	0-11%
Deer	0-3%	0-2%	0-3%	0-2%
Spring vegetation and summer fruit	70-85%	84-94%	75-90%	84-95%
<b>Fall mixing model (1/3<sup>rd</sup> root end)</b>				
Sunflowers	0-7%	0-5%	0-10%	0-2%
Corn	19-39%	1-11%	10-30%	0-11%
Fall fruit and nuts	59-75%	87-96%	67-83%	88-99%
<b>Annual Diet from Crops</b>	<b>12-31%</b>	<b>0-10%</b>	<b>5-24%</b>	<b>0-12%</b>

Table 2. Average annual home range size (km<sup>2</sup>) (95% CI) of GPS-collared bears in northwestern Minnesota based on 33 female and 27 male bear-years, 2007–2013. Only fixes separated by 4 hours or more were included.

Sex	Method <sup>a</sup>	50% isopleth	95% isopleth	100% isopleth
Female	MCP	21.6 (8.3 - 34.9)	72.5 (45.5 - 99.5)	115.9 (80.5 - 151.2)
	Rule Based	18.1 (10.4 - 25.8)	77.8 (51.7 - 103.8)	---
	H-Ref	19.1 (11.5 - 26.7)	91.4 (54.5 - 128.3)	---
	k LoCoH	2.6 (1.6 - 3.6)	30.3 (18.5 - 42.1)	---
Male	MCP	172.8 (107.3 - 238.3)	616.9 (466.1 - 767.7)	861.4 (639.0 - 1083.8)
	Rule Based	168.6 (114.4 - 222.7)	754.0 (550.7 - 957.3)	---
	H-Ref	172.6 (118.7 - 226.6)	834.1 (596.3 - 1071.9)	---
	k LoCoH	7.6 (4.8 - 10.4)	172.9 (129.8 - 215.9)	---

<sup>a</sup> MCP = minimum convex polygon, Ad-hoc = rule-based approach using an ad-hoc method for selecting a bandwidth, H-ref = kernel density estimator with reference bandwidth for H, k LoCoH = fixed k nearest neighbor convex hull.

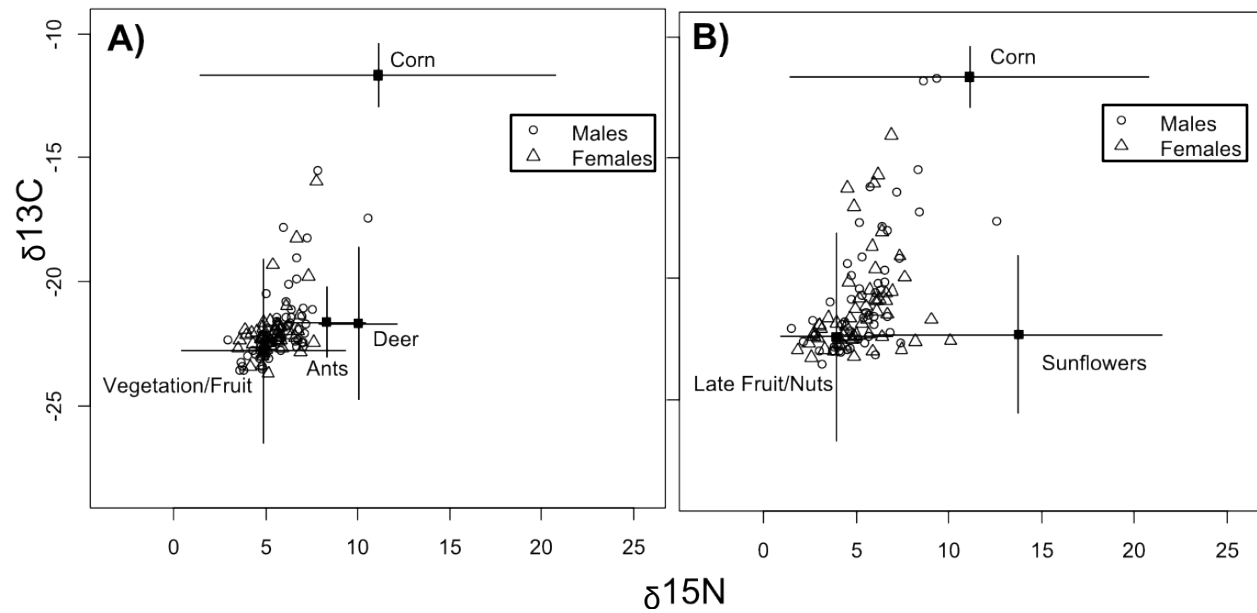


Figure 1. Isotopic signatures of bear-hair compared to bear foods. Bear hairs represent 56 female and 54 male bear-years. Food sources available for consumption are from (A) spring until ~mid-August and (B) ~mid-August until winter in northwestern Minnesota. Isotopic values of hair samples correspond to the distal 2/3rds end of the hair (A) and the 1/3rd root end of the hair (B).

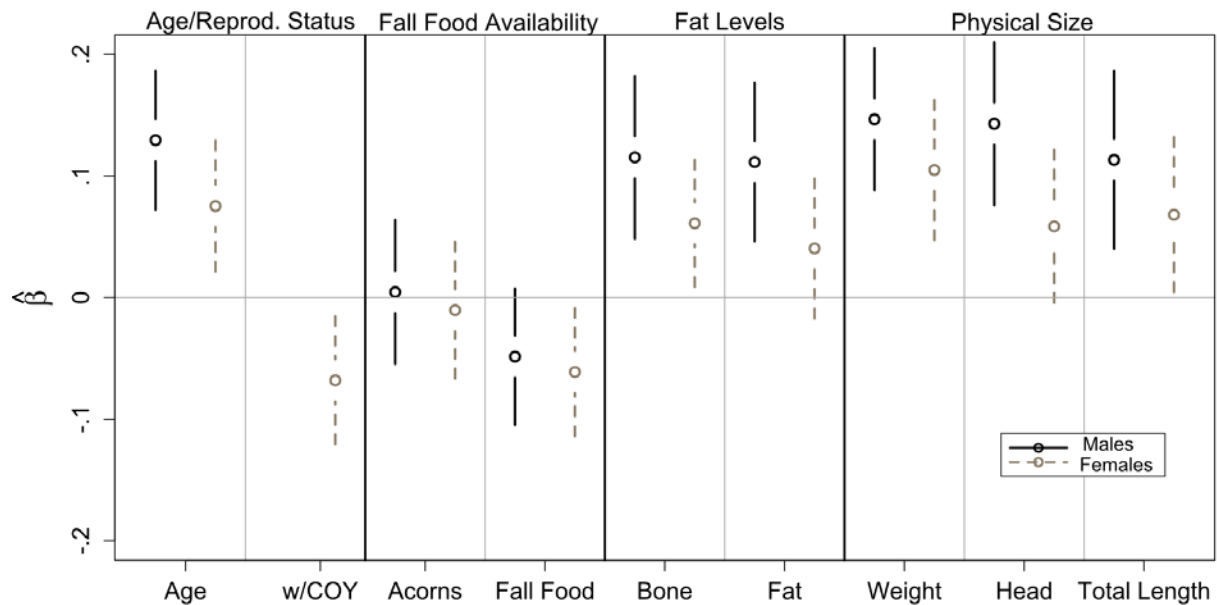


Figure 2. Parameter estimates ( $\hat{\beta}$ ), with 95% confidence intervals, from linear mixed model regressions between estimates of individual bears' consumption of crops and covariates relating to food availability and physical status of animals. Covariates were scaled and centered for comparison purposes.

Covariates:

Age: Age of bear

w/COY: With cubs of the year

Acorns: Estimate of annual acorn mast (acorns/m<sup>2</sup>)

Fall Food: Indices of primary fall foods (hazelnut, dogwood, oak)

Bone: Bone prominence of bear (measure of amount of body fat)

Fat: Skin-fold thickness (measure of amount of body fat)

Weight: Weight of the bear at time of handling

Head: Head girth

Total Length: Total length (from nose-tip to tip of tail)

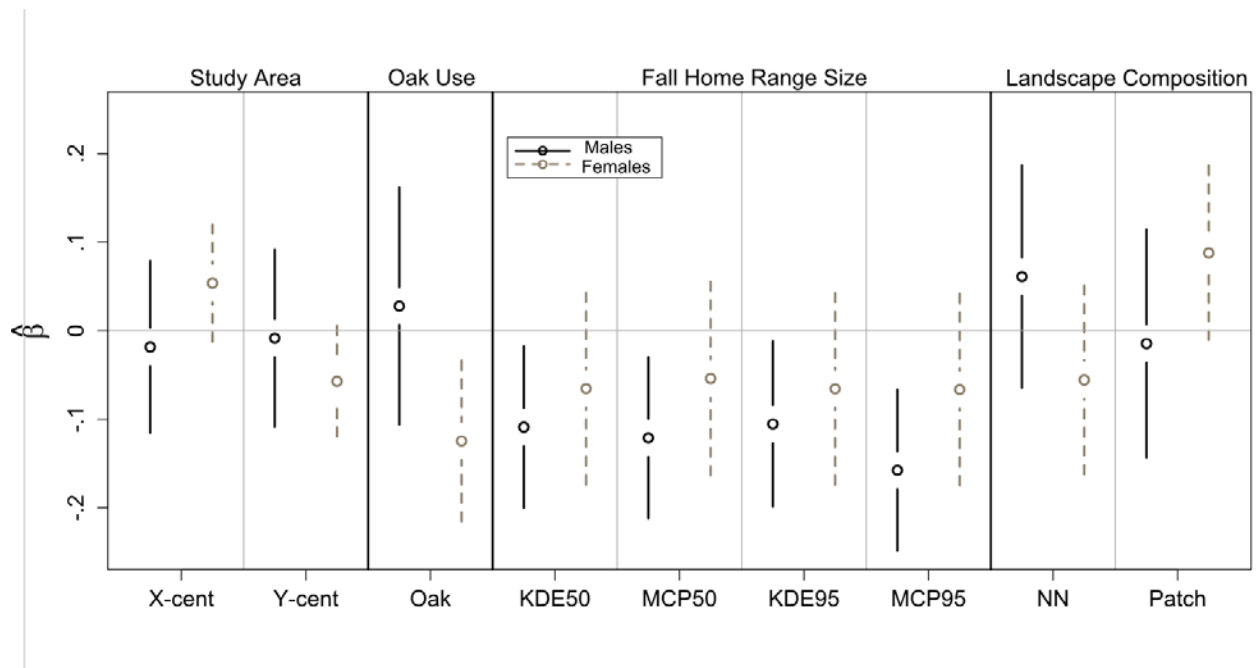


Figure 3. Parameter estimates ( $\hat{\beta}$ ) with 95% confidence intervals, derived from linear mixed model regressions relating estimate of individual bears' consumption of crops with spatial covariates based on GPS data from radio-collars. Covariates were scaled and centered for comparison purposes.

Covariates:

X-cent: Center of activity (east - west) based on trap location, kill site, den or home range centroid

Y-cent: Center of activity (north-south) based on trap location, kill site, den or home range centroid

Oak: % of GPS locations in oak and upland shrub habitats

KDE50: 50% fixed kernel density estimate of fall home range area

MCP50: 50% minimum convex polygon estimate of fall home range area

KDE95: 95% fixed kernel density estimate of fall home range area

MCP95: 95% minimum convex polygon estimate of fall home range area

NN: Average shortest distance between each natural habitat patch with a home range (habitat connectivity estimate)

Patch: Mean size of natural habitat patches within a fall home range (habitat connectivity estimate)



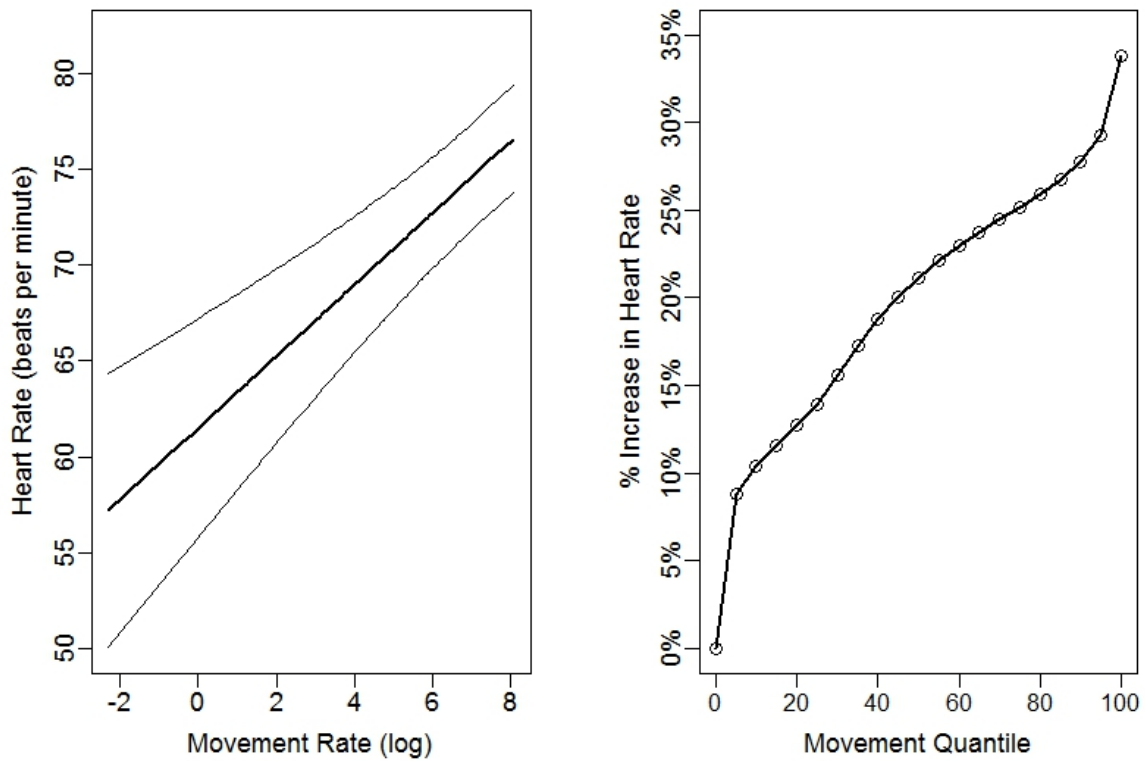


Figure 4. (A) Influence of movement rate ( $\log(m/\text{hour})$ ) on heart rate (bpm,  $\pm 95\%$  CI) of free-roaming black bears in northwestern Minnesota, based on the best-approximating population model with all other covariates held constant at their mean values. (B) Percent increase in mean estimated heart rate with quantile change in log movement rate.



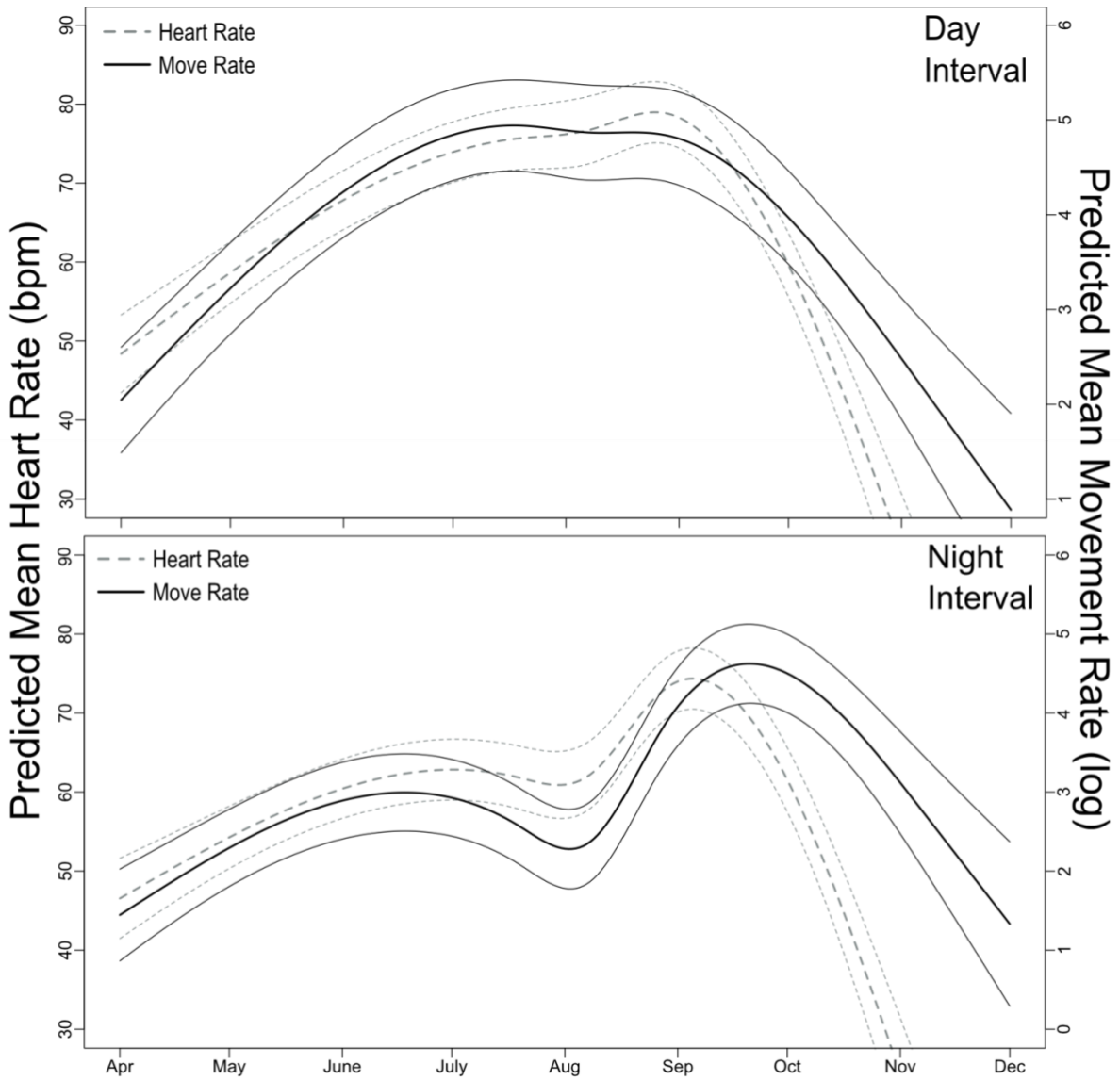


Figure 5. Estimates of mean (A) daytime and (B) nighttime heart rate (bpm,  $\pm$  95% CI) versus mean log movement rate (m/hour;  $\pm$  95% CI) for northwestern Minnesota black bears over the course of a year, 2009–2012.

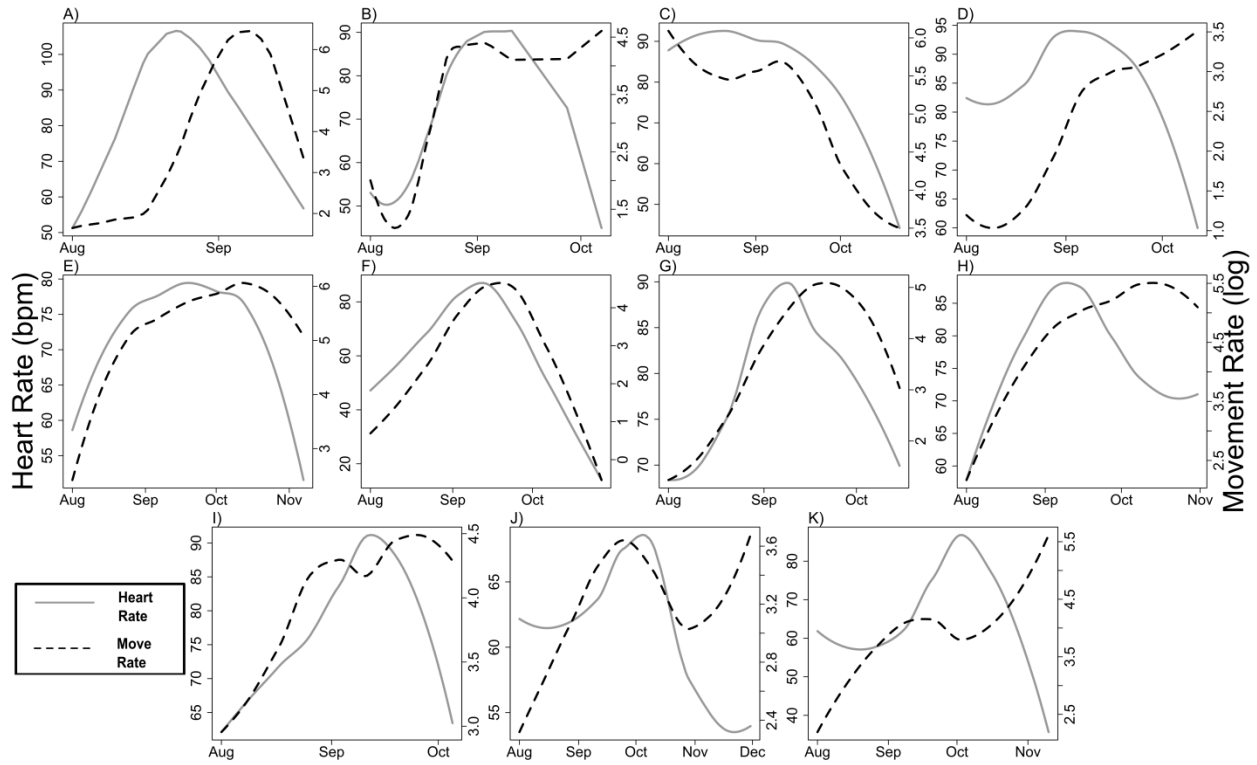


Figure 6. Loess smoothed data of nighttime heart rate (bpm; primary y-axis) and movement rate (log; secondary y-axis) for individual northwestern Minnesota black bears during autumn (August – den entry). All bear-years exhibited the pattern of a reduced average daily nighttime heart rate prior to reduced daily nighttime movement rates except for the largest bear in the study (C).

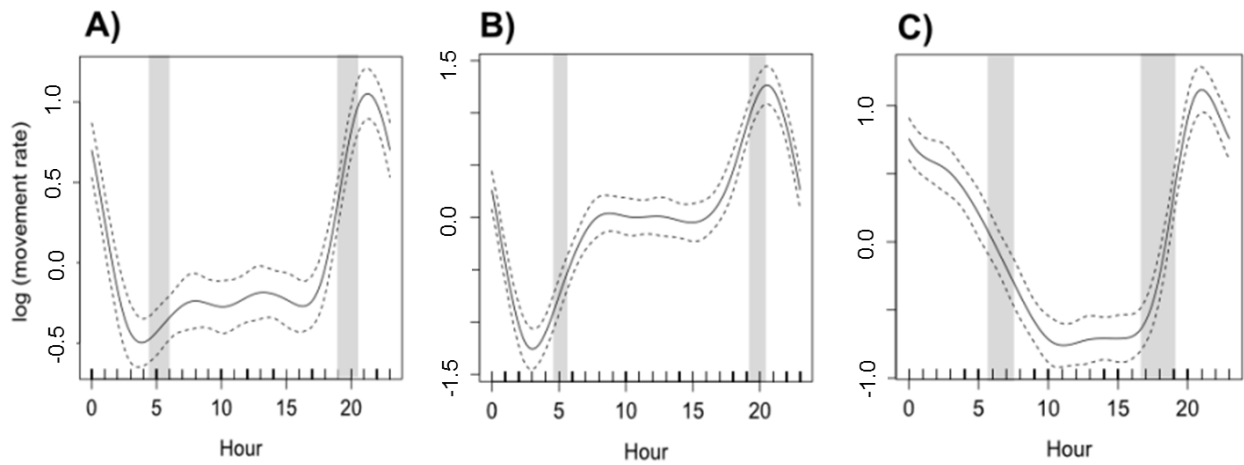


Figure 7. Influence of hour of day on log movement rate ( $\pm$  95% CI) of northwestern Minnesota black bears during (A) spring, (B), summer, and (C) fall, 2009–2012. Gray shaded areas represent the approximate range of sunrise and sunset.

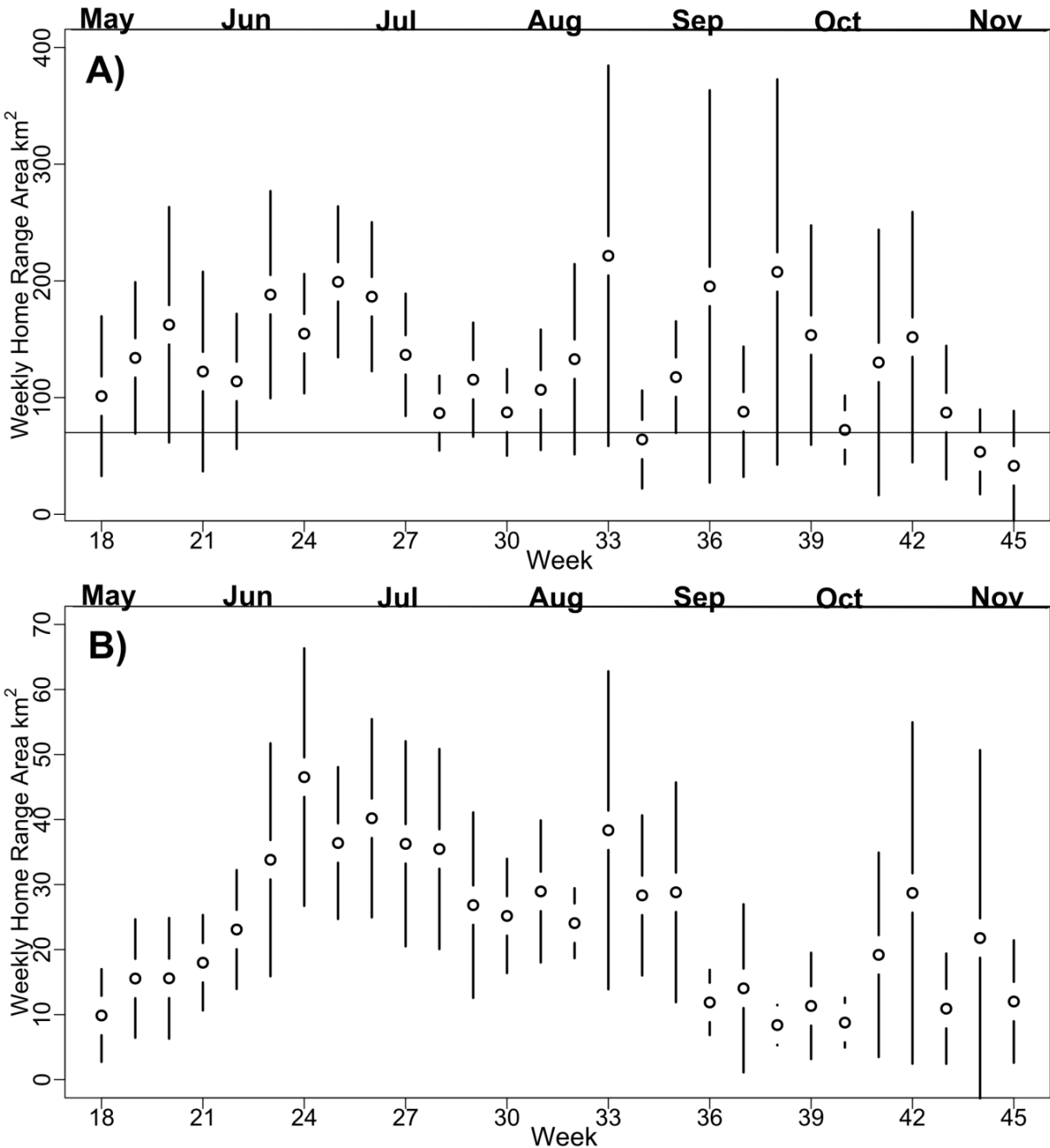


Figure 8. Average weekly home range (WHR) estimates and 95% confidence intervals ( $\text{km}^2$ ) of GPS-collared (A) male and (B) female black bears in northwestern Minnesota, 2007-2013. Seventy-five bear-years (37 female, 38 male) were used to construct 1,453 weekly home range estimates using the 95% isopleth with the rule based method of kernel estimation. Averages and the corresponding confidence intervals were calculated using the point estimates of WHRs for each week. The gray horizontal line in the upper panel represents the maximum value of the females' y-axis ( $70 \text{ km}^2$ ) superimposed on the male data.

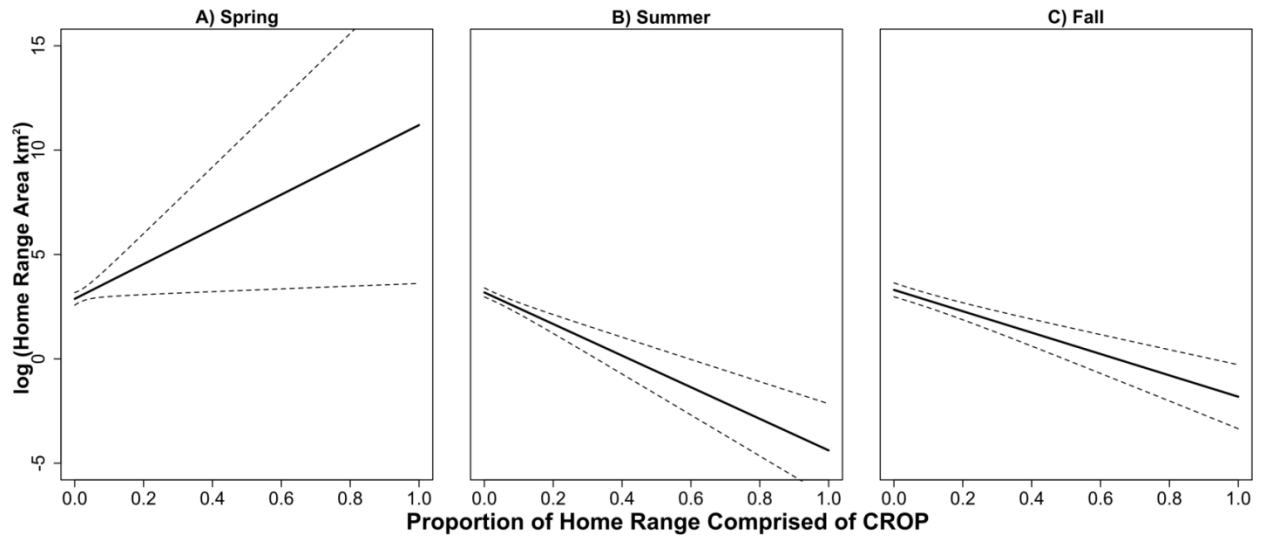


Figure 9. Predicted changes in weekly home range size ( $\log \text{ km}^2$ ) for northwestern Minnesota black bears in response to changes in the proportion of edible crops within their home range during spring (left: ~May 1 – July 1), summer (middle: ~July 1 – Sept. 1) and fall (right: ~Sep 1 – Dec 1).

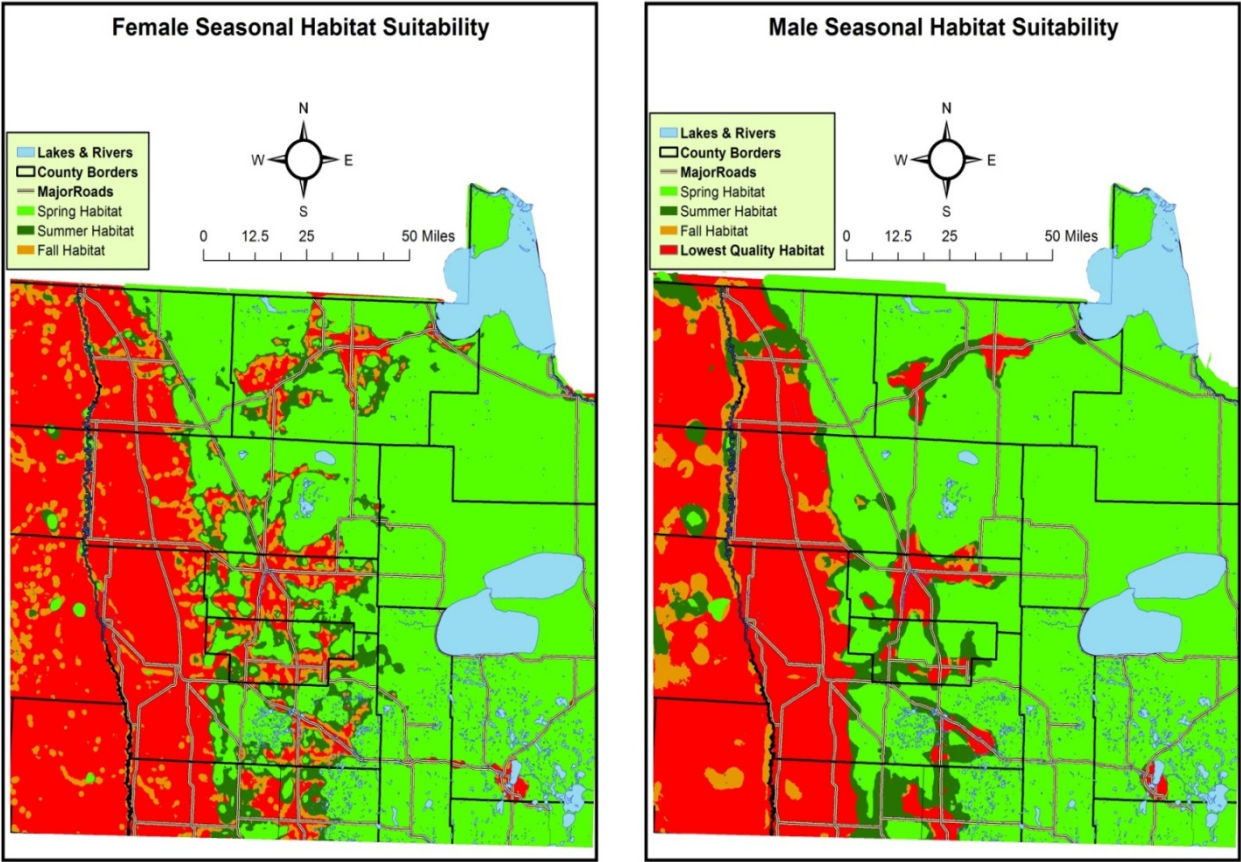


Figure 10. Projected maximum habitat suitability extent during spring (May 1 – July. 1), summer (July 1 – Sept. 1), and fall (Sep 1 – Dec 1) for female (left) and male (right) bears living in northwestern Minnesota and (potentially) eastern counties of North Dakota. The map illustrates the expansion of usable habitat throughout the year. Spring had the least overall coverage so is shown as the top layer with summer and fall maps below. Any areas showing fall habitat are only available during fall whereas all spring area is available during summer and fall.

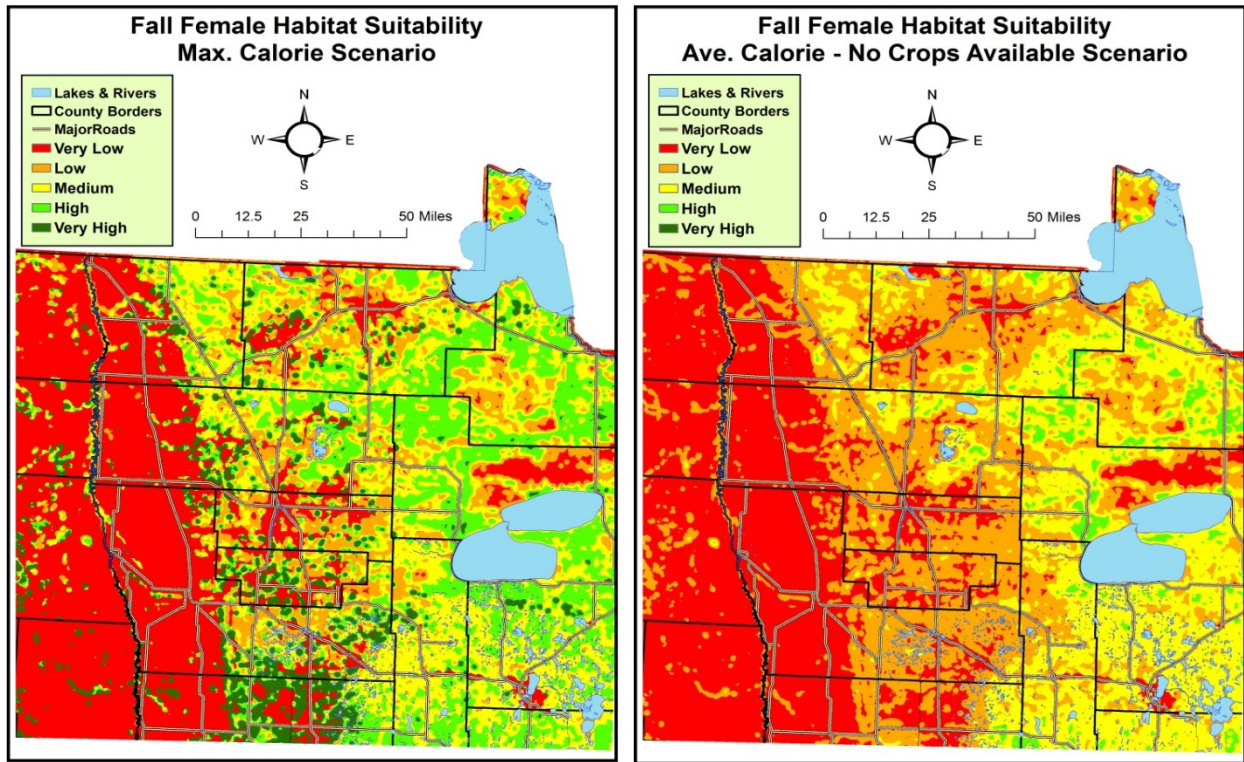


Figure 11. Projected habitat suitability during fall (Sep 1 – Dec 1) for female bears living in northwestern Minnesota and (potentially) eastern counties of North Dakota for years with high natural food conditions and crop availability(left panel) versus average natural food conditions with no edible crops available (right panel). Maps incorporate natural cover requirements, the influence of roads, and caloric values based on weekly home ranges of GPS-collared bears.





## REPRODUCTIVE ECOLOGY OF FISHERS AND AMERICAN MARTENS IN MINNESOTA

John Erb, Pam Coy, and Barry Sampson

### SUMMARY OF FINDINGS

As part of a larger project on *Martes* ecology in Minnesota, we began monitoring reproductive success of radio-collared fishers (*Martes pennanti*) and martens (*Martes americana*) during spring 2008. Including the pilot year of the study, a total of 232 martens (111F, 121M) and 101 fishers (58F, 43M) have been radio-collared. To date, age and reproductive status have been confirmed on 40 adult ( $\geq 2$  years old) female martens. Pooling years, pregnancy rate has been ~57% for 2-year-old martens, and ~85% for martens 3 years or older. Average size of 24 marten litters is 2.92 (range = 1-4), with minimal difference between litter size of 2- versus 3+ year-old females. Based on initial data, it appears marten kits are typically born in late-April through mid-May. A total of 45 marten natal or maternal dens have been located, of which 64% have been in tree (primarily aspen (*Populus tremuloides*) and cedar (*Thuja occidentalis*)) cavities and 36% in underground burrows. We have also confirmed both age and reproductive status for 51 female fishers. Pooling years, pregnancy rate has been ~57% for 2-year-old fishers, and ~93% for fishers 3 years or older. Average size of 37 fisher litters is 2.5 (range = 1-4). Data suggests that litter size for 2-year-old fishers is lower than for older females (2.2 versus 2.7). Based on data collected to date, it appears fisher kits are typically born in early to mid-March, or ~1.5 months earlier than marten kits. A total of 56 fisher natal or maternal dens have been confirmed, all but 2 being in elevated tree cavities. Cavities have been located in both live trees (74%) and snags (26%) with an overall average DBH of 20.5 in. Tree cavities used by female fishers have been located primarily in aspen (68.5%) (*Populus tremuloides*, *Populus grandidentata*) and oak (15%) (*Quercus* spp.) trees. Most female fishers appear to move kits from their natal den to at least 2 different maternal dens prior to June 1.

### INTRODUCTION

American marten and fisher are native to Minnesota, but reliable documentation of their historic distribution is limited. Undoubtedly, northeastern Minnesota was a stronghold for the marten population, though notable numbers likely occurred in the northern border areas as far west as Roseau County. Limited information suggests they occurred as far south as Crow Wing County and as far southwest as Polk County. As a result of unregulated harvest, marten were considered rare in Minnesota by 1900, and extensive logging and burning around the turn of the century further contributed to the near extirpation of marten from Minnesota by the 1930s (Swanson et al. 1945). Fishers in Minnesota appear to have historically occupied a larger geographic area than martens, extending further south and west into the hardwood dominated transition zone, including southeast Minnesota (Swanson et al. 1945, Balsler and Longley 1966). The impacts of unregulated harvest and habitat alteration were equally as detrimental to fisher, with populations substantially reduced by the 1930s.

Legally, fisher and marten were unprotected in Minnesota prior to 1917, after which harvest season length restrictions were implemented. These protections were removed in the mid-1920s, and remained so until all harvest was prohibited in 1929. Seasons remained closed until 1977 for fisher and 1985 for marten, when limited harvests were reinstated. While marten harvest is now legal in approximately the northern 50% of the state, most harvest occurs in counties bordering Canada, particularly in northeast and north-central Minnesota. Fisher harvest occurs in most of the northern 50% of the state, though harvest is comparatively low in extreme northeast Minnesota (Lake and Cook counties). Over the past 10 years, fisher



abundance and harvest have been increasing along the southern and western edge of the 'forest zone' where forest historically transitioned to savanna and prairie and is now characterized by linear forest corridors (streams/rivers) or smaller forest patches interspersed with agriculture. Conversely, fisher abundance appears to have declined significantly over the same period in the core forested areas of north-central and northeast Minnesota. Peak statewide harvest levels have been near 4,000 and 3,500 for marten and fisher, respectively. However, due to apparent multi-year population declines for both species, harvest seasons from 2007 to the present have become progressively more conservative, with the 2013 harvest season lasting only 6 days with a combined fisher/marten limit of 2 per trapper.

While both species appear to have naturally re-colonized a significant portion of their historic range, Minnesota-specific information on reproductive ecology is limited to carcass (corpora lutea, placental scar) data collected from harvested animals primarily from 1985-90 (Kuehn 1989, Minnesota DNR unpublished data). Reproductive data is also available from other geographic areas, but questions remain on the accuracy of various methods to assess reproduction, and the amount of spatial and temporal variation in reproductive parameters. Minnesota-specific data on structures and sites used by fisher for natal and maternal dens is also lacking.

*Martes* pregnancy rate and litter size data are generally quantified from 1 of 4 methods: counts of corpora lutea (CL) in ovaries; counts of blastocysts (BC) in uteri; placental scar (PS) counts; or direct observation of litter size (Gilbert 1987, Mead 1994). Assuming both species are induced ovulators (but see Cherepak and Connor 1992, Frost et al. 1997), CL counts should accurately reflect copulation and ovulation rates, but all CL persist even if only 1 ovum is fertilized. Blastocyst counts reflect the number of fertilized ova, but not all BC may implant in the uterus and develop, and BC are often destroyed in poorly preserved carcasses. Hence, these 2 measures may not only overestimate litter size for parous females, but may also overestimate parturition rate (i.e., females may ovulate, 1 or more ova become fertilized, yet they fail to ultimately den and give birth). Placental scars, formed last in the reproductive process, would seem the most reliable carcass-based estimate of parturition rate and litter size. However, several authors (Gilbert 1987, Payne 1982, Strickland and Douglas 1987) have suggested that PS may not always persist long enough in mustelids to be detected during the harvest season when carcasses are easily collected. Furthermore, PS can persist in some species even if fetuses are resorbed (Conaway 1955) and detection and counts of PS may be affected by observer variability (Johnson et al. 1995). Nevertheless, PS have been reliably used in the past (e.g., Coulter 1966, Crowley et al. 1990), though others have noted that reliable results may only be obtainable when doing microscopic analysis of fresh and properly preserved/prepared uteri (Mead 1994, Frost et al. 1999).

In spite of these concerns, average litter size estimates from reproductive organs do not appear to be substantially biased. Strickland and Douglas (1987), summarizing data from 136 captive marten litters, computed average litter size of 2.9 for martens. This is within the range of average litter sizes reported from ovary or uterine analysis (~ 2.5 – 3.5; Strickland et al. 1982, Strickland and Douglas 1987, Flynn and Schumacher 1995, 2009, Aune and Schladweiler 1997, MN DNR unpublished data). For fishers, the same appears to be true, with an average litter size of 2.8 from 60 captive fisher litters (reviewed in Strickland and Douglas 1987) and 19 wild litters (York 1996), which compares favorably to estimates based on reproductive organs (2.7 – 3.9 (CL), 2.7 – 3.2 (BC), and 2.5 – 2.9 (PC); review in Powell 1993).

Of greater concern is the possibility that ovary, and to lesser degree uterine, analyses might consistently overestimate parturition rate, thereby also underestimating annual variability in parturition rates. Various indications of pregnancy may be detected, though not all of those females may den and produce kits in spring. For example, this might occur if BC fail to implant or fetuses are resorbed as a result of nutritional stress during the period of embryonic diapause (Arthur and Krohn 1991). Overall, CL counts have generally yielded ovulation rates for fisher of  $\geq 95\%$  (Shea et al. 1985, Douglas and Strickland 1987, Paragi 1990, Crowley et al. 1990, MN

DNR unpublished data), while more 'direct' estimates of average parturition rate from radio-marked animals have been lower (46-75%; Crowley et al. 1990; Arthur and Krohn 1991; Paragi 1990; Paragi et al. 1994, York 1996, Truex et al. 1998, Higley and Mathews 2009), and are often highly variable. Conversely, in Minnesota, Kuehn (1989) did not detect changes in fisher pregnancy rate (from CL analysis) in spite of a 64% decline in a presumably important prey species (snowshoe hare; *Lepus americanus*).

For martens, several largely ovarian-based estimates of annual pregnancy rate have often been in the range of 80-90% (Archibald and Jessup 1984, Strickland and Douglas 1987, Aune and Schladweiler 1997, Flynn and Schumacher 1994, Fortin and Cantin 2004, MN DNR unpublished data). However, like for fishers, several marten studies have documented (also based largely on CL counts) lower or more variable pregnancy rates (Thompson and Colgan 1987, Aune and Schladweiler 1997, Strickland and Douglas 1987, Flynn and Schumacher 2009), perhaps a result of fluctuations in prey abundance (Hawley and Newby 1957, Weckwerth and Hawley 1962, Strickland 1981, Strickland and Douglas 1987, Thompson and Colgan 1987, Fryxell et al. 1999, Flynn and Schumacher 2009). We are aware of direct field-based estimates of parturition rate from radio-marked marten in only one state (Maine). Pooling samples across 4 years, the proportion of lactating adult females was 75, 81, and 92% for their 3 different study areas (Phillips 1994, Payer 1999), similar to many of the CL-based pregnancy studies.

Understanding reproductive ecology of these species also necessitates gathering information on natal and maternal den structures and selection of den sites. Natal dens are the structures where kits are born, whereas maternal dens are sites used subsequently by the female with her dependent young. Although data is absent for Minnesota, nearly all reported fisher natal dens have been in cavities of large-diameter trees or snags (Leonard 1986, Paragi et al. 1996, Powell et al. 1997, Truex et al. 1998). In northern studies, the majority of fisher natal dens have been in large diameter aspens (*Populus* spp), and females may use up to 3 or more different maternal dens (Powell et al. 2003, Higley and Mathews 2009). Marten natal and maternal dens are also frequently in tree cavities (Gilbert et al. 1997), but may occur in more varied features (e.g., under-ground burrows, exposed root masses of trees, rock piles, large downed logs; Ruggiero et al. 1998).

Though not further discussed here, the literature is also voluminous with documentation of the importance of tree cavities, large downed logs, and other forest 'structure' for fisher and marten resting sites (see Powell et al. 2003 for a review). Initial results from this study (Joyce 2013) appear consistent with other published findings on the importance of forest structure for marten den and rest sites. Given the continuing pressure to maximize fiber production from forests (i.e., short forest rotation, biomass harvesting, etc), the forest structural attributes critical to fishers and martens could become limiting in the future, if not already. Hence, acquiring Minnesota-specific information is critical to better inform forest management activities.

## **OBJECTIVES**

As part of a larger project on *Martes* ecology (Erb et al. 2009), we began efforts to better describe the reproductive ecology of fisher and marten in Minnesota. Specific objectives are to:

- Document denning chronology;
- Determine structures used for natal and maternal dens;
- Quantify vegetative characteristics in the area surrounding natal and maternal dens;
- Develop a resource selection model specific to the denning season;
- Derive field-based estimates of pregnancy rate and litter size;
- Evaluate kit survival; and
- Assess the potential influence of age, diet, prey fluctuations, forest attributes, and winter severity on reproductive success.

After initial evaluation of field methods during the pilot year of the study, spring 2009 marked the beginning of full-scale research activities. We defer a more complete evaluation of results until additional data are collected or additional analysis is completed. Herein we present basic information on field methods and only report preliminary findings related to denning chronology, dens structures, and pregnancy rates and litter sizes. For initial analysis related to den and rest site selection for martens, we refer the reader to Joyce (2013).

## STUDY AREA

Marten research is focused on 1 study area located in northeastern Minnesota (Figure 1; Area 1), though an occasional marten has been captured and radio-collared in Area 2 (Figure 1). Area 1 (~700 km<sup>2</sup>) is ~ 90% public ownership, including portions of the Superior National Forest and state and county lands. Fishers are also present in this area at low to moderate density.

Fisher research has taken place in 3 areas (Figure 1; Areas 1, 2, and 3). The work in Area 3 is a collaborative effort between Camp Ripley Military Reservation, Central Lakes Community College, and the Minnesota Department of Natural Resources. While we do include animals captured in that area in our basic summaries, we do not discuss other aspects of that project in this report. Area 2 (1075 km<sup>2</sup>), our primary fisher study area, is ~ 67% public ownership, including portions of the Chippewa National Forest and state and county lands. Extremely few martens occupy Area 2.

## METHODS

We used Tomahawk (Tomahawk Live Trap, Hazelhurst, WI) cage traps to capture both fishers (Model 108) and martens (Models 106 and 108) during winter. Traps were typically baited with deer (*Odocoileus virginianus*) or beaver (*Castor canadensis*) meat, and we placed commercial lure in or above the traps. We enclosed traps inside white plastic 'feed sacks' or burlap bags and further covered traps with natural vegetation. All traps were checked daily.

To immobilize animals, we used metal 'combs' to restrict the animal to a small portion of the trap, or restrained the animal against the side of the trap by pulling its tail through the cage mesh. Animals were injected with a hand-syringe using a 10:1 mixture of ketamine and xylazine (fisher: 30 mg/kg ketamine and 3 mg/kg xylazine; marten: 20 mg/kg ketamine, 2 mg/kg xylazine) (Kreeger et al. 2002). After processing, the xylazine was reversed with yohimbine at a dosage of 0.1 mg/kg (marten) or 0.15 mg/kg (fisher). Fisher were either ear-tagged with a monel # 3 tag in one ear (National Band and Tag Co., Newport, KY) and a 2-piece plastic mini-tag (Dalton I.D. Systems, UK) in the other ear, or with a monel # 3 tag in both ears. Marten were ear-tagged with a monel #1 tag (National Band and Tag Co., Newport, KY) in each ear.

During processing, we placed animals on heating pads connected to a power inverter and 12 volt battery. Portable shelters and propane heaters were also used to keep animals warm during processing. We monitored respiration, pulse, and rectal temperature during anesthesia. We weighed and sexed animals and typically removed a first pre-molar for aging. Morphological measurements taken included body length, tail length, hind foot length, and chest, neck, and head circumference. We removed guard hair samples for possible genotyping, and for evaluating the use of stable isotope analysis for deciphering food habits (Ben-David et al. 1997). To assist with determining which female fishers would likely produce kits, blood samples were drawn when possible to measure serum progesterone levels (Frost et al. 1997). Antibiotics were administered subcutaneously to all animals prior to release as a precaution against infection (Kreeger et al. 2002) from minor wounds that may have occurred while in the trap, and because of certain invasive procedures utilized during handling (ear-tagging, removal of tooth).

During the pilot year, we deployed several radio-collar designs on fishers, including an Advanced Telemetry Systems (ATS; Isanti, MN) M1585 zip-tie collar (~43 g), an ATS M1930 collar (~38 g), and a Lotek Wireless Inc. (Newmarket, ON, CA) SMRC-3 collar (~ 61 g; deployed on adult males only). Since the pilot year, we have primarily deployed ATS M1940 (~43 g) or Sirtrack (Havelock North, New Zealand) TVC-162 collars (~45 g) on fishers. The majority of martens have been fitted with Holohil Systems Ltd. (Carp, ON, CA) MI-2 collars (~31 g). We retrofitted each collar with a temperature data logger (I-button model DS1922L; Maxim Integrated, San Jose, CA) to provide ancillary information on winter activity and spring den attendance patterns, as well as to provide information on time of death for other study objectives.

We ground-tracked collared females to locate possible den structures. When a suspected den structure was located, we then deployed remotely-activated cameras (Reconyx PC-85, RC-55, or HC600, Reconyx, Inc, Holmen, WI) to monitor female activity. We considered a female to have given birth only if kits were confirmed via sound or video/camera, or if other reliable evidence (e.g., obvious lactation, placental scars, or kit bite marks on collar) was obtained when an animal was subsequently handled as a mortality or recapture. Litter size was ascertained via visual confirmation in most cases, though we also utilized placental scar counts on any females that died during summer or fall, and for which other methods failed to produce a count. To confirm or count kits at dens located in tree cavities, we used an MVC2120-WP color video camera (Micro Video Products, Bobcaygeon, Ontario), attached to a telescoping pole if necessary, and connected to a laptop computer. Dens were only examined when the radio-marked female was not present. If video inspection equipment did not work at a particular den structure, we deployed remote cameras in an effort to obtain pictures of kits when they emerged or were moved by the female (Jones et al. 1997).

When a natal or maternal den was confirmed, we recorded den location (above/on/below-ground) as well as various location-specific details (e.g., tree species, log/tree diameter, burrow entrance attributes, etc.). We note that since birth is never observed, and kits may be moved to new dens within days following birth, distinguishing natal dens from maternal dens can rarely be done with certainty. Hence, we pool natal and maternal dens for purposes of general summaries herein.

We are also collecting more detailed information on vegetative characteristics of the site surrounding each den structure, with a goal of developing a biologically meaningful den site selection model using methods and metrics that should be available from existing and periodically collected forest sampling data (e.g., see Zielinski et al. 2006). Following the United States Forest Service's Forest Inventory and Analysis (FIA) protocol, we quantify vegetative characteristics in a 1-acre (120 ft. radius) area surrounding the den structure by sampling in 4 circular subplots, each being 0.04-acre (24-ft radius) in size. One subplot is centered on the den structure, with the other 3 subplots centered 120 feet from the den at 360°, 120°, and 240°. Within each subplot, 3 24 ft. coarse woody debris sampling transects are established, originating from the subplot center, and oriented at 30°, 150°, and 270°. Deviating from FIA protocol, we also establish 3 (not 1, as with FIA) 0.003-acre (6.8 ft radius) circular micro-plots for estimating sapling density, each micro-plot situated at the end of the 3 coarse woody debris sampling transects. Details of vegetation sampling methods within each subplot will be outlined in subsequent years as results become available. Herein, we simply note that we are collecting quantitative data on 1) mean DBH and basal area of live trees, overall and by species; 2) % overhead (angular) canopy; 3) sapling density; 4) understory cover density; 5) density and volume of snags and stumps; 6) volume of coarse woody debris; 7) distance to improved road; and 8) distance to water. Canopy structure will also be categorized based on number and distribution of canopy layers. Lower-resolution (1 ppm) LIDAR data will also be analyzed in all study areas, along with higher-resolution (8 ppm) data for a portion of the marten study area.

To better understand any observed fluctuations in reproductive parameters, we are also collecting data on factors that may influence reproductive success, including winter severity and

prey fluctuations. In each study area, a temperature monitor was placed on the north-facing side of a tree in each of 6 cover types. Each sensor records temperature every 30 minutes from December 1 – June 1. At approximately 10-day intervals from 1 December – 1 April, we also recorded snow depth and 2 measures of snow compaction at 3 locations along transects situated in each of 6 cover types. Two snow compaction tools were constructed using PVC pipe, one each with an end-cap similar in diameter to a typical marten and fisher track in the snow. Each pipe length was then adjusted to ensure the pipe-specific load ( $\text{g}/\text{cm}^2$ ) was similar to marten and fisher foot-loading measures (females) reported by Krohn et al. (2004). Depth of snow compaction was recorded by dropping each load tool from 1 in. above snow level and measuring compaction depth.

Prey sampling transects have also been established in both study areas. Prey sampling is being conducted primarily to document between-area differences in prey abundance, annual within-area fluctuations in prey, and ultimately to assess whether fisher or marten habitat use, diet, survival, or reproductive success is correlated with prey dynamics. Prey-sampling transects ( $n \approx 125$  in each study area) consist of 10 sampling locations (2 parallel lines of 5 stations) spaced 20m apart, with transects distributed in 6 cover types throughout each study area. Transects are generally oriented perpendicular to roads or trails, with the first plot 30m off the trail. In spring, we count snowshoe hare (*Lepus americanus*) pellets in a 1- $\text{m}^2$  plot at each sampling station (McCann et al. 2008). During fall, small mammal snap-trapping occurs for 2 consecutive days at the same sampling stations, similar to protocols used on an existing small mammal survey in Minnesota (Aarhus-Ward 2009). During both spring (hare pellet sampling) and fall (small mammal trapping), we also count the number of red squirrels (*Tamiasciurus hudsonicus*) observed or heard along each transect. Rather than using 10-min point counts (e.g., Mattson and Reinhart 1996, Bayne and Hobson 2000) with our small mammal/hare pellet stations as the sampling points, we record the number of unique squirrels detected per transect (summarized per unit time) while checking pellet plots and small mammal traps. Information on white-tailed deer and ruffed grouse (*Bonasa umbellus*) populations may be available from existing surveys or population models.

## RESULTS AND DISCUSSION

Including the pilot year of the study, a total of 232 martens (111F, 121M) and 101 fishers (58F, 43M) have been radio-collared. Because tooth aging has not yet been completed for all animals, some of which may be only 1 year of age (i.e., not capable of producing kits), we present results only for animals known to be  $\geq 2$  years of age during spring den visits, or those of unknown age but for which we have confirmed parturition at the time of this writing (i.e., until age is known, we do not include animals that we have confirmed to be nulliparous). As of this writing, spring 2014 reproductive status assessment is largely complete for radio-collared fishers, but still ongoing for a few martens.

Treating females that were alive during multiple parturition periods (years) as independent units, and excluding females known to be  $< 2$  years of age, we have confirmed age and reproductive status for 40 female martens (Table 1). Pooling years, pregnancy rate has been 57% for 2-year-old martens ( $n=14$ ), and 85% for martens 3 years or older ( $n=26$ ) (Table 1). We have been able to confidently assess litter size for 24 marten litters, for which average litter size is 2.92 (range = 1-4); litter size averaged 2.83 for 2-year-olds ( $n=6$ ) and 3.0 for 3+ year-old ( $n=16$ ) martens (Table 1). Data suggests marten kits are typically born from late-April through mid-May.

A total of 45 natal or maternal dens have been confirmed for martens (Table 2). For temporal reference, 37 (82%) of the marten dens were documented to be used from mid-April through June 1<sup>st</sup>, with the low number ( $n=8$ ) of maternal dens located after this a result of time constraints and increasing difficulty in finding dens in summer. Of the 45 dens, 64% were in tree cavities whereas 36% were in underground burrows (Table 2). Of the 29 dens in tree

cavities, 69% have been in live trees whereas 31% have been in snags. Pooling live trees and snags, most tree cavity dens used by martens have been in aspen (n=11) and cedar (n=10), with from 1 – 3 dens located each in tamarack (*Larix laricina*), red maple (*Acer rubrum*), black ash (*Fraxinus nigra*), and white pine (*Pinus strobus*) (Table 2). We have completed DBH measurements on 16 of the 29 den trees, yielding an average DBH of 18.6" (range = 13.3 – 30.0") (Table 2). Of the 16 underground dens, 8 were characterized as being in soils with an abundance of medium to large rocks, 5 were under the base of larger trees or associated with shallow roots or sphagnum 'soils' adjacent to the base of the tree, while 3 were under 'tip-ups' or stumps (Table 2). Most female martens appear to move their kits from their natal den to 1 or more maternal dens in the first 8 weeks following birth.

Similar to martens, we treat female fishers that were alive during multiple parturition periods (years) as independent units. Excluding individuals known to be 1 year of age during the parturition period, we have confirmed both age and reproductive status for 51 female fishers (Table 1). Pooling years, pregnancy rate for female fishers has been 57% for 2-year-olds (n=21), and 93% for fishers 3 years or older (n=30) (Table 1). We have been able to confidently assess litter size for 37 fisher litters from which average litter size is 2.5 (range = 1–4); litter size averaged 2.25 for 2-year olds (n=12) and 2.7 for 3+ year-old (n=23) fishers (Table 1). Based on data collected to date, it appears fisher kits are typically born in early to mid-March, or ~1.5 months earlier than marten kits.

A total of 56 fisher natal or maternal dens have been located to date (Table 3). For temporal reference, 52 (93%) of the fisher dens confirmed were documented to be used in March and April, with the low number (n=4) of maternal dens located after June 1 resulting from time constraints and increasing difficulty in finding dens in summer. Of the 56 dens confirmed, all but 2 were in elevated tree cavities; the remaining 2 maternal dens were in large hollow logs either on or suspended above the ground (Table 3). Of the dens in tree cavities, 74% have been in live trees whereas 26% have been in snags. Pooling live trees and snags, most tree cavity dens used by fishers have been in aspen (n=37) and oak (n=8), with from 1 – 3 dens located each in sugar maple (*Acer saccharum*), red maple, white cedar, and white pine (Table 3). We have completed DBH measurements on 53 of the 55 fisher den trees, yielding an average DBH of 20.5" (range = 13.9 – 29.1") (Table 3). Similar to martens, most female fishers appear to move their kits from their natal den to 1 or more maternal dens in the first 8 weeks following birth.

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Table 1. Parturition rate and litter size for radio-collared<sup>1</sup> female fishers and martens in Minnesota from 2008-2014.

Species*Age	Parturition Rate		Litter Size		
	# females	% with litters	# litters	Average	Range
<b>Martens</b>					
2-year-olds	14	57	6	2.83	1 - 4
3+ year-olds	26	85	16	3.0	1 - 4
All	42 <sup>2</sup>	74	24 <sup>3</sup>	2.92	1 - 4
<b>Fishers</b>					
2-year-olds	21	57	12	2.25	1 - 4
3+ year-olds	30	93	23	2.7	1 - 4
All	51	78	37 <sup>3</sup>	2.5	1 - 4

<sup>1</sup> Excludes unknown-aged nulliparous females and all 1-year-olds. Multiple years for same female treated as independent.

<sup>2</sup> Includes 2 females with age ≥ 2, but otherwise unknown age

<sup>3</sup> Includes known litters from unknown-aged females

Table 2. Natal and maternal den structures (n=45) used by radio-collared female martens in Minnesota from 2008 - 2014.

Den Structure	# dens	% of total	Average DBH (in.)	DBH Range (in.)
Above-Ground, All Tree Cavities	29	64.4	18.6	13.3 – 30.0
Cavity, live tree	20	42.2	19.6	15.9 – 30.0
Cavity, snag	9	20.0	15.3	13.3 – 20.4
All Aspen cavities	11	24.4	18.0	13.3 – 23.8
All Cedar cavities	10	22.2	19.2	16.1 – 21.5
All Tamarack cavities	3	6.7	16.8	Only 1 measured to date
All Red Maple cavities	2	4.4	16.3	15.9 – 16.7
All Black Ash cavities	2	4.4	16.1	14.3 – 17.8
All White Pine cavities	1	2.2	30.0	
Below-Ground Dens	16	35.6		
Burrow, under base of tree	5	11.1	14.5	9.0 – 18.6
Burrow, rocky soils/outcrop	8	17.8		
Burrow, under stump/tip-up	3	6.7		

Table 3. Natal and maternal den structures (n=56) used by radio-collared female fishers in Minnesota from 2008 - 2014.

Den Structure	# dens	% of total	Average DBH (in.)	DBH Range (in.)
Above-Ground, Tree Cavities	54	96.4	20.5	13.6 – 29.1
Cavity, live tree	40	71.4	20.9	13.9 – 29.1
Cavity, snag	14	25.0	19.6	13.6 – 26.1
Aspen cavities	37	66.1	20.4	15.1 – 29.1
Oak cavities	8	14.3	20.5	15.1 – 28.0
White pine cavities	3	5.3	23.7	21.9 – 25.6
Sugar Maple cavities	2	3.6	20.6	19.1 – 22.1
Red Maple cavities	2	3.6	20.8	18.0 – 23.6
Cedar cavities	2	3.6	17.1	13.9 – 20.3
Hollow Log	2	3.6	16.9	15.4 – 18.3

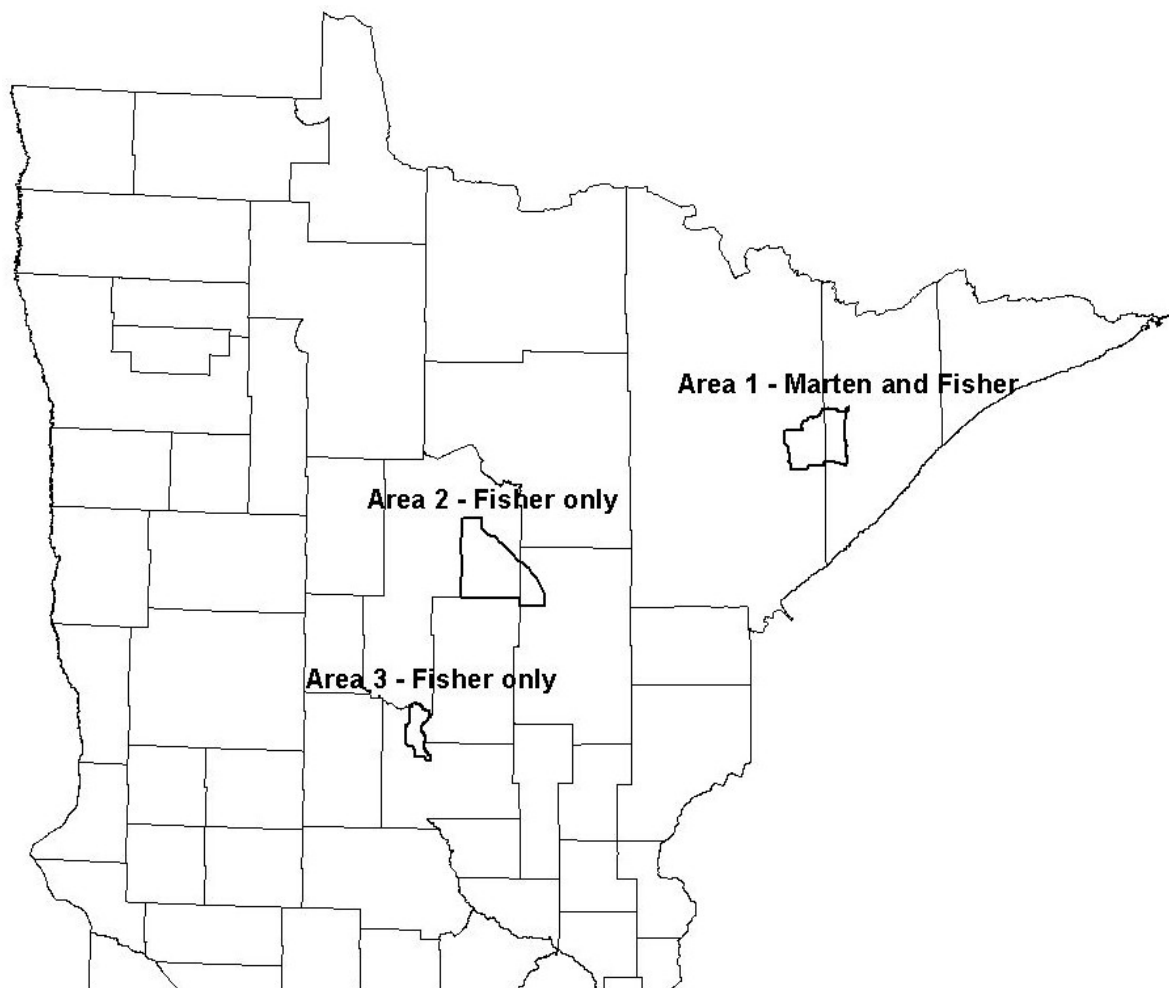


Figure 1. Fisher and American marten study areas in Minnesota, 2008-2014.



## **SURVIVAL AND CAUSES OF MORTALITY FOR FISHERS AND MARTENS IN MINNESOTA**

John Erb, Pam Coy, and Barry Sampson

### **SUMMARY OF FINDINGS**

As part of a larger project on *Martes* ecology in Minnesota, we began monitoring survival of radio-collared fishers (*Martes pennanti*) and martens (*Martes americana*) during winter 2007-08. Including the pilot year of the study, a total of 232 martens (111F, 121M) and 101 fishers (58F, 43M) have been radio-collared. An additional 6 animals (3 martens, 3 fishers) were ear-tagged only. Of the 232 martens radio-collared, 33 are still actively monitored (15F, 18M), radio-contact has been lost on 61, 7 whose collars are inaccessible have either slipped their collars or died, and 131 deaths have been confirmed (of which 12 were censored due to death within 2 weeks of capture). Of the 119 non-censored marten deaths (53F, 66M), most have been from legal fur trapping (n=47; 34M, 13F) and predation (n=55; 33F, 22M). Approximately 91% of the marten predation deaths have been attributed to mammalian carnivores and 9% to raptors. Although natural mortality of martens has occurred in most seasons, it is highest in spring and lowest in fall. No significant sex bias has been observed in overall mortality - female martens make up 46% of our sample and 44% of the known deaths. However, marten harvest mortality has been male-biased (70% male) while natural mortality has been female-biased (60% female).

Of the 101 fishers radio-collared, 10 are still being monitored (6F, 4M), radio contact was lost on 33, 7 whose collars are inaccessible have either slipped their collars or died, and 51 deaths (32F, 19M) have occurred. Of the 51 fisher deaths, most have been from predation (n=25; 20F, 5M) and fur trapping (8 in-season, 8 accidentally out-of-season). Two fishers have been car-killed and 5 others died from unknown but apparent natural mortality. Similar to martens, natural mortality for fishers is highest in spring and lowest in fall. Of 20 female fishers predated, 19 were killed by other mammalian carnivores. Conversely, 4 of the 5 male fisher predation mortalities were attributed to raptors (all bald eagles). Of particular note, 18 of the 20 female fishers killed by predators were adults, and 17 of them were killed while they still had dependent young in natal dens, indirectly resulting in the death of all their offspring. The deaths of these 18 nursing females and their litters represent ~33% of the reproductive 'opportunities' for adult female fishers monitored during the kit-rearing season since the study began. We hypothesize that the observed timing and magnitude of female mortality is a result of increased activity and increased vulnerability during the kit-rearing season. Because the magnitude of the mortality would not likely have been sustainable for an extended period, we suggest that survival patterns have probably changed in the last 10 years. We continue to explore several hypotheses, but suspect a partial explanation to be that cumulative changes in the environment may have had both direct (e.g., reduction in denning habitat quality) and indirect (e.g., weather and habitat more favorable to competing bobcats) effects on survival of female fishers in the core of Minnesota's fisher range.

### **INTRODUCTION**

American marten and fisher are native to Minnesota, but reliable documentation of their historic distribution is limited. Undoubtedly, northeastern Minnesota was a stronghold for the marten population, though notable numbers likely occurred in the northern border areas as far west as Roseau County. Limited information suggests they occurred as far south as Crow Wing County and as far southwest as Polk County. As a result of unregulated harvest, marten were considered rare in Minnesota by 1900, and extensive logging and burning around the turn of the century further contributed to the near extirpation of marten from Minnesota by the 1930s

(Swanson et al. 1945). Fishers in Minnesota appear to have historically occupied a larger geographic area than martens, extending further south and west into the hardwood dominated transition zone, including southeast Minnesota (Swanson et al. 1945, Balsler and Longley 1966). The impacts of unregulated harvest and habitat alteration were equally as detrimental to fisher, with populations substantially reduced by the 1930s.

Legally, fisher and marten were unprotected in Minnesota prior to 1917, after which harvest season length restrictions were implemented. These protections were removed in the mid-1920s, and remained so until all harvest was prohibited in 1929. Seasons remained closed until 1977 for fisher and 1985 for marten, when limited harvests were reinstated. While marten harvest is now legal in approximately the northern 50% of the state, most harvest occurs in counties bordering Canada, particularly in northeast and north-central Minnesota. Fisher harvest occurs in most of the northern 50% of the state, though harvest is comparatively low in extreme northeast Minnesota (Lake and Cook counties). Over the past 10 years, fisher abundance and harvest have been increasing along the southern and western edge of the 'forest zone' where forest historically transitioned to savanna and prairie and is now characterized by linear forest corridors (streams/rivers) or smaller forest patches interspersed with agriculture. Conversely, fisher abundance appears to have declined significantly over the same period in the core forested areas of north-central and northeast Minnesota. Peak statewide harvest levels have been near 4,000 and 3,500 for marten and fisher, respectively. However, due to apparent multi-year population declines for both species, harvest seasons from 2007 to the present have become progressively more conservative, with the 2013 harvest season lasting only 6 days with a combined fisher/marten limit of 2 per trapper.

While both species appear to have naturally re-colonized a significant portion of their historic range, Minnesota-specific information on survival and causes of mortality is limited. Except for harvest data, we are aware of only 1 published field study in Minnesota. Specifically, Mech and Rogers (1977) opportunistically radio-collared 4 marten and reported survival and home range information for those animals. This information is specific to marten, now nearly 30 years old, and based on a very limited sample size. Gathering cause-specific mortality information can be useful for informing population models, detecting unknown mortality agents, and focusing management activities on issues of concern.

Krohn et al. (1994) estimated 11% annual non-harvest mortality for adult fisher in Maine, while York (1996) estimated 19% and 7% annual non-harvest mortality (incl. 4% poaching mortality on males) for adult male and female fisher, respectively, in Massachusetts. Excluding the first 4-5 months of life, juvenile fisher non-harvest mortality rates have been estimated to be 28% in Maine (Krohn et al. 1994), and 0% (females) and 23% (males) in Massachusetts (York 1996). While mortality may be higher in the first months of life than the rest of the year, if we assume a similar non-harvest mortality rate during the first 4-5 months of life, we calculate that annual non-harvest mortality for juvenile fishers would be ~ 56% in Maine. Combining minimum summer survival estimates for kits with telemetry estimates of survival the rest of the year, York (1996) estimated ~ 67% (males) and 22% (females) annual non-harvest mortality for juvenile fishers in Massachusetts. Kelly (1977, in Paragi et al. 1994) reportedly estimated 18% annual mortality of juveniles and 44% annual mortality for adult fisher in New Hampshire. More recently, Koen et al. (2007) estimated annual mortality rate (including harvest mortality) of fishers in Ontario to be 55-67% for males, and 29-37% for females. While non-harvest mortality of adult fishers is often presumed to be 'low', it has not always proven to be the case. Furthermore, there is limited data on which to assess the amount of geographic or temporal variation in non-harvest mortality of fisher.

Natural mortality, particularly via predation, appears more common with martens. Marten survival data is available from Wisconsin (McCann et al. 2010), Maine (Hodgman et al. 1994, 1997), Ontario (Thompson 1994), Oregon (Bull and Heater 2001), British Columbia (Poole et al. 2004), Alaska (Flynn and Schumacher 1995, 2009), Quebec (Potvin and Breton

1997), and Newfoundland (Fredrickson 1990). Although we do not summarize details of these studies here, a couple conclusions are worthwhile. First, when comparing across studies, annual adult non-harvest mortality rates varied from ~ 0.07 – 0.48. Juvenile data was rarely separated, but a few studies pooled ages, and mortality rates also were within the above interval. While this variability may be attributable to both sampling and biological variability, the wide range suggests that it is risky to assume results from any area are applicable elsewhere. Secondly, at least 1 study (Maine; Hodgman et al. 1997) has documented significantly higher natural mortality for females compared to males, and other researchers have postulated this to be common given the typical male-biased harvest, 50:50 sex ratio at birth, and often balanced adult sex ratio (Strickland et al. 1982, Strickland and Douglas 1987). Due to male-biased harvest and our *assumed* sex-related equality in non-harvest mortality, our marten population model previously projected a very female-biased population, contradicting our preliminary capture results and suggesting that our model inputs were overestimating female survival, underestimating male survival, or incorrectly assuming a 50:50 birth sex ratio.

## OBJECTIVES

As part of a larger project on *Martes* ecology in Minnesota (Erb et al. 2009), we began monitoring survival and causes of mortality for fishers and martens. After initial evaluation of field methods during the pilot year of the study, winter 2008-09 marked the beginning of full-scale research activities. While details are not discussed here, we are also collecting data on various potential correlates to survival (e.g., prey dynamics, winter severity, diet, habitat use, activity patterns, and body condition). Our primary objectives are to:

- Determine causes of mortality;
- Estimate cause- and sex-specific mortality rates;
- Document seasonal patterns of mortality; and
- Examine potential effects of winter weather, prey fluctuations, competitor density, activity patterns, and habitat on survival probability.

Herein we present basic descriptive information regarding number of captures and number and causes of deaths. We defer a more comprehensive statistical analysis until a later time.

## STUDY AREA

Marten research is focused on 1 study area located in northeastern Minnesota (Figure 1; Area 1), though an occasional marten has been captured and radio-collared in Area 2 (Figure 1). Area 1 (~700 km<sup>2</sup>) is ~ 90% public ownership, including portions of the Superior National Forest and state and county lands. Fishers are also present in this area at low to moderate density.

Fisher research has taken place in 3 areas (Figure 1; Areas 1, 2, and 3). The work in Area 3 is a collaborative effort between Camp Ripley Military Reservation, Central Lakes Community College, and the Minnesota Department of Natural Resources. While we do include animals captured in that area in our basic summaries, we do not discuss other aspects of that project in this report. Area 2 (1075 km<sup>2</sup>), our primary fisher study area, is ~ 67% public ownership, including portions of the Chippewa National Forest and state and county lands. Extremely few martens occupy Area 2.

## METHODS

We used Tomahawk (Tomahawk Live Trap, Hazelhurst, WI) cage traps to capture both fishers (Model 108) and martens (Models 106 and 108) during winter. Traps were typically baited with either deer (*Odocoileus virginianus*) or beaver (*Castor canadensis*) meat, and commercial lure was placed in or above the traps. We enclosed traps inside white plastic 'feed sacks' or burlap bags and further covered traps with natural vegetation. All traps were checked daily.

To immobilize animals, we used metal 'combs' to restrict the animal to a small portion of the trap, or restrained the animal against the side of the trap by pulling its tail through the cage mesh. Animals were injected with a hand-syringe using a 10:1 mixture of ketamine and xylazine (fisher: 30 mg/kg ketamine and 3 mg/kg xylazine; marten: 20 mg/kg ketamine, 2 mg/kg xylazine) (Kreeger et al. 2002). After processing, the xylazine was reversed with yohimbine at a dosage of 0.1 mg/kg (martens) or 0.15 mg/kg (fishers). Fishers were either ear-tagged with a monel # 3 tag in one ear (National Band and Tag Co., Newport, KY) and a 2-piece plastic mini-tag (Dalton I.D. Systems, UK) in the other ear, or with a monel # 3 tag in both ears. Martens were ear-tagged with a monel #1 tag (National Band and Tag Co., Newport, KY) in each ear.

During processing, we placed animals on heating pads connected to a power inverter and 12 volt battery. Portable shelters and propane heaters were also used to keep animals warm during processing. We monitored respiration, pulse, and rectal temperature during anesthesia. We weighed and sexed animals and typically removed a first pre-molar for aging. Morphological measurements taken included body length, tail length, hind foot length, and chest, neck, and head circumference. We removed guard hair samples for possible genotyping, and for evaluating the use of stable isotope analysis for deciphering food habits (Ben-David et al. 1997). To assist with determining which female fishers would likely produce kits, blood samples were drawn when possible to measure serum progesterone levels (Frost et al. 1997). Antibiotics were administered subcutaneously to all animals prior to release as a precaution against infection (Kreeger et al. 2002) from minor wounds that may have occurred while in the trap, and because of certain invasive procedures utilized during handling (ear-tagging, removal of tooth).

During the pilot year, we deployed several radio-collar designs on fishers, including an Advanced Telemetry Systems (ATS; Isanti, MN) M1585 zip-tie collar (~43 g), an ATS M1930 collar (~38 g), and a Lotek Wireless Inc. (Newmarket, ON, CA) SMRC-3 collar (~ 61 g; deployed on adult males only). Since the pilot year, we have primarily deployed ATS M1940 (~43 g) or Sirtrack (Havelock North, New Zealand) TVC-162 collars (~45 g) on fishers. The majority of martens have been fitted with Holohil Systems Ltd. (Carp, ON, CA) MI-2 collars (~31 g). We retrofitted each collar with a temperature data logger (I-button model DS1922L; Maxim Integrated, San Jose, CA) to provide ancillary information on winter activity and spring den attendance patterns, as well as to provide information on time of death for other study objectives.

All radio-locations, except for some taken during the den-monitoring period, were obtained from fixed-wing aircraft at approximately weekly intervals. When a radiocollar emits a mortality signal, we usually investigate and recover the animal or collar within 1-2 days. To determine cause of mortality, we use a combination of field investigation and animal necropsy. Starting in the second year of the project, we also began collecting forensic samples (hair by wound, wound swabs) from all animals exhibiting signs of being predated, particularly if a mammalian predator is suspected. Forensic samples are submitted to the University of California-Davis Veterinary Genetics Laboratory. If non-predation natural causes are suspected after initial analysis (i.e., no visible trauma), the animal is submitted to the University of Minnesota's Veterinary Pathology Lab for a full pathological exam.

## RESULTS AND DISCUSSION

Including the pilot year of the study, a total of 232 martens (111F, 121M) and 101 fishers (58F, 43M) have been radio-collared. An additional 6 animals (3 martens, 3 fishers) were ear-tagged only. Tooth aging has not yet been completed for all animals; however we note that because capture operations took place during winter, all animals were a minimum of ~ 8 months of age at initial capture. We have yet to derive formal estimates of survival rate. Instead, we provide a simple overview of the fate of collared animals in this summary.

Excluding 12 martens that were censored due to death within 14 days post-release, 33 (15%) of the 220 collared martens are actively being monitored, radio contact has now been lost on 53 (24%), 8 (4%) slipped their collars, the status of 7 (3%) is uncertain due to unrecoverable collars, and 119 (54%) have died (Table 1). Of the 119 non-censored deaths, most have been from legal fur trapping ( $n=47$ ) and predation ( $n=55$ ) (Table 2). Nine animals died of other natural causes, including being crushed by a tree, perforation and blockage of the intestine from a piece of bone, starvation related to an intestinal polyp, and 6 from unknown but assumed natural causes (Table 2).

Of the 53 non-censored marten deaths that we believe could be attributed to either avian or mammalian predation, evidence suggests 48 (91%) were killed by mammalian predators and 5 (9%) by raptors. Although predation deaths have occurred in nearly all months, predation and overall natural mortality is highest in the spring and lowest in the fall (Figure 2). Forensic (DNA) analysis of samples collected from predated marten (mammalian predation only) is still incomplete. To date, field evidence and DNA analysis suggests bobcats (*Lynx rufus*) as a common mammalian predator, with red fox (*Vulpes vulpes*) and fisher also confirmed in several cases.

Excluding martens censored within 14 days of capture, our sample of radio-collared marten has been comprised of 46% females. In comparison, female martens have accounted for 44% of the total marten deaths, 30% of the total deaths due to harvest, and 60% of the predation deaths. Although there is no apparent sex-bias to overall mortality, marten harvest has been notably male-biased while natural mortality has been female-biased.

Of the 101 fishers radio-collared, 10 (10%) are still being monitored, radio contact has been lost with 18 (18%), 15 (15%) collars have come unattached, the fate of 7 (7%) (unrecovered collars) is uncertain, and 51 (50%) have died (Table 1). General cause of death (human versus natural) could be assigned to 48 of the 51 fisher deaths. Of these, 18 (37.5%) were attributable to humans (8 trapped during legal season, 8 accidental trapping, and 2 car-killed) whereas 30 (62.5%) were attributable to natural causes (25 predated, 5 unknown natural cause) (Table 2). The seasonal pattern of natural mortality for fishers has been similar to that of martens, being greatest in spring and lowest in fall (Figure 3).

Of the 25 predated fishers, 20 were females (Table 2). Of the 20 females predated, only 1 was attributed to an avian predator (Great-horned owl (*Bubo virginianus*) suspected). Conversely, 4 of the 5 male fisher predation deaths were attributed to bald eagles (*Haliaeetus leucocephalus*), though we can't rule out scavenging in 1 case where only the radio-collar was retrieved directly underneath an active eagle nest. The fifth male fisher predated appears to have been killed by a wolf. We are awaiting forensic DNA analysis on many fishers killed by mammalian predators. However, similar to martens, field evidence and forensic DNA analysis suggests bobcats as the most common predator, with canids (wolf or coyote) suspected in 2 fisher deaths.

Of particular note, 18 of the 20 female fishers killed by other predators were adults ( $\geq 2$  years old), and 17 of those 18 were killed while they still had dependent kits in natal or maternal dens, indirectly resulting in the death of all their dependent offspring. The deaths of these kit-rearing females represent a complete loss of ~33% of the fisher litters monitored during spring and early summer since the study began.



We continue to suspect that 2 broad factors may explain the high mortality of kit-rearing female fishers during late-winter and spring: increased activity, and increased vulnerability (independent of activity level). Given the potential for negative energy balance during parts of winter, compounded in early spring by the added energy demands of gestation and lactation, female fishers may need to increase activity in spring to meet energy demands. Combined with the need to locate suitable (and multiple) natal or maternal dens, this activity, much of which may be in localized areas near den trees and hence more predictable/detectable to other carnivores, may increase predation risk. Preliminary data from temperature data loggers attached to radio-collars suggests that fishers do spend increasing amounts of time (compared to winter) outside of den and rest sites during late-winter and spring. Secondly, independent of their activity level, fishers may be more vulnerable in spring because concealment cover is diminished (i.e., before 'green-up'), other predators may also increase activity in spring, and potential prey for carnivores is at the low point in the annual cycle. Collectively, this may yield a period of high energetic need that overlaps a high risk competitive environment for female fishers. Progressing into summer and fall, concealment cover is maximal, prey abundance (for all carnivores) is maximal, and energetic demands (and hence movements) of female fishers decrease as kits are weaned.

Regardless of the explanation, it seems unlikely that the level of predation we have observed on nursing female fishers during the study would be sustainable for long periods, which may partially explain the decline in fisher abundance in core areas over the previous decade. However, many of the correlates to the timing of predation mortality that we have mentioned are not new challenges for adult female fisher, and since 1977 the core fisher population appears to have only been in decline over the last ~ 9 years suggesting that other more recent changes may be altering dynamics. Likely explanations for the observed and presumably new mortality pattern for female fishers continue to be assessed, including potential declines in fisher habitat quality in core fisher range and changes in habitat and weather that may have contributed to an increase in competing bobcats. Overharvest of fisher, particularly in the 4 years preceding the start of this study, may also have contributed to the decline in fisher abundance, but would not explain the high natural mortality of female fishers, and to some extent female martens, that we have observed during the study.

## **ACKNOWLEDGEMENTS**

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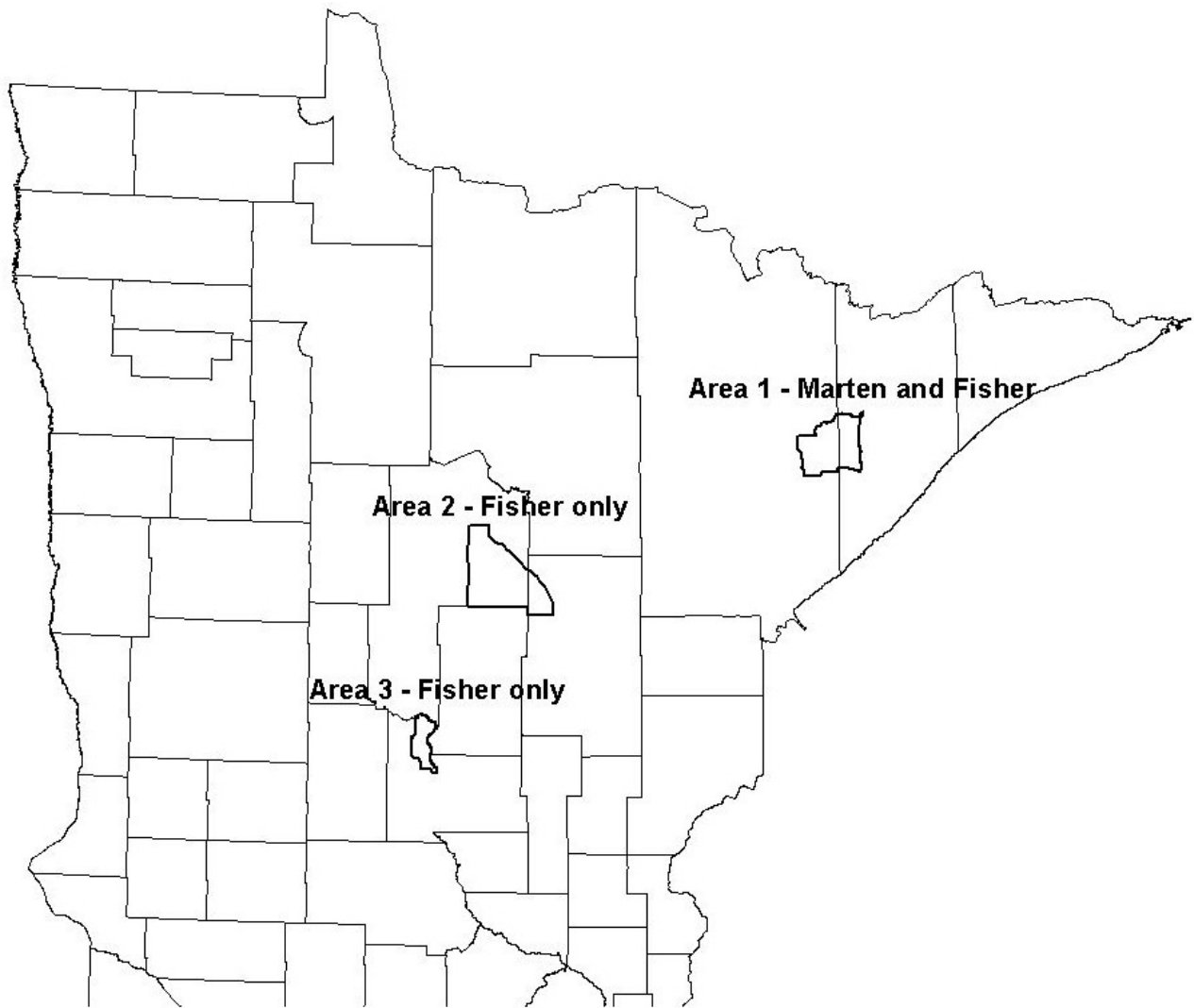


Figure 1. Fisher and marten study areas in Minnesota 2008-2014.

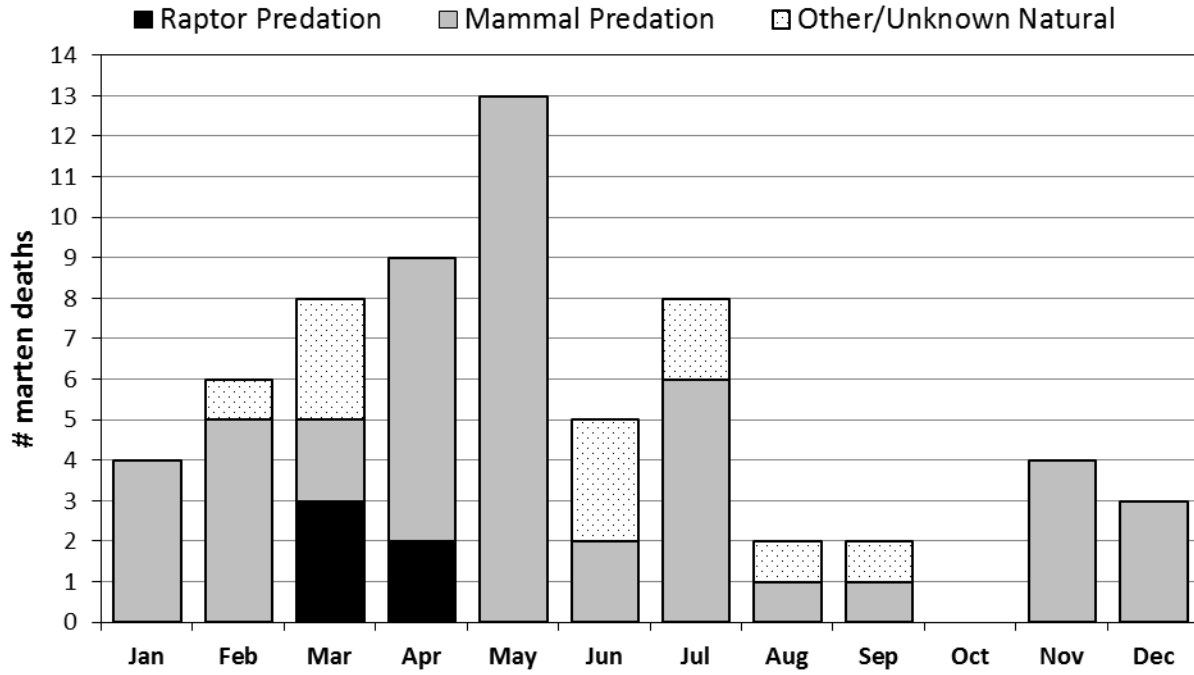


Figure 1. Seasonal timing of natural mortality for martens in northeast Minnesota, 2007 - 2014.

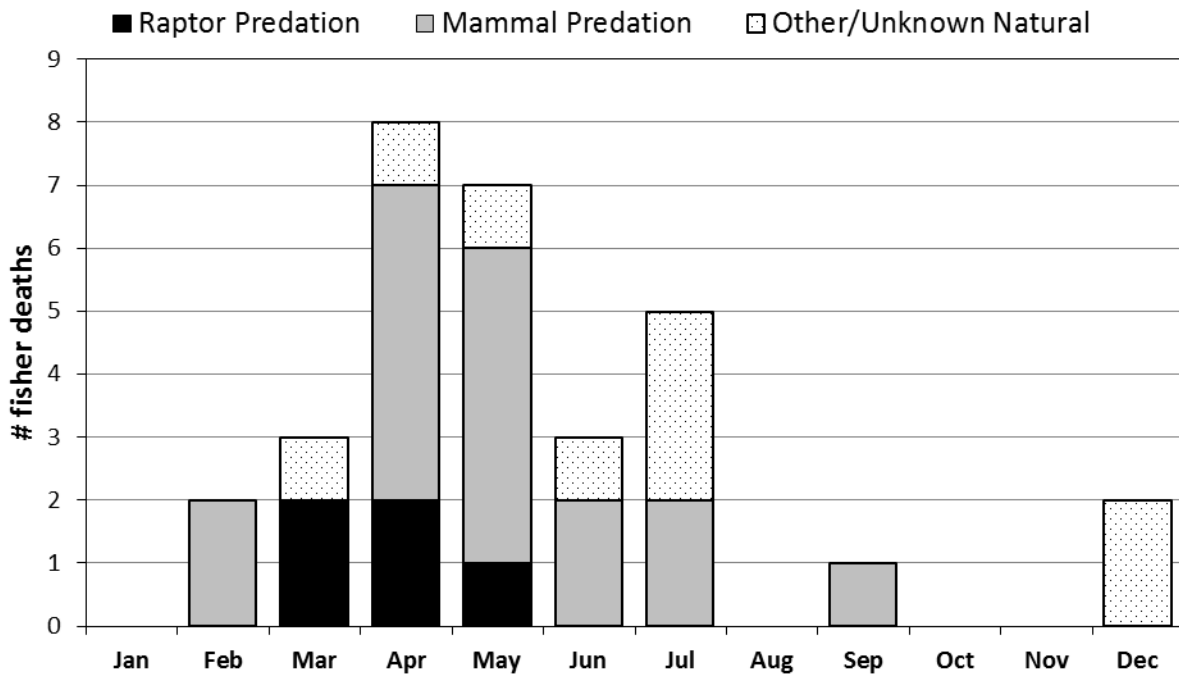


Figure 2. Seasonal timing of natural mortality for fishers in northeast Minnesota, 2007 - 2014.

Table 1. Number and status of fishers and martens radio-collared<sup>1</sup> in Minnesota from 2007 - 2014.

Sex*Species	# Active	# Deaths	# Missing	# Slipped Collars	Unknown <sup>2</sup>	Total
Male Martens	18	66	23	4	1	112
Female Martens	15	53	30	4	6	108
Male Fishers	4	19	10	6	4	43
Female Fishers	6	32	8	9	3	58

<sup>1</sup> Excludes radio-collared animals that died within 2 weeks of capture and release

<sup>2</sup> Unknown represents collars not yet retrieved from tree cavities or underground locations (presumed dead or slipped collars), or retrieved but with uncertainty whether the animal slipped the collar or had died

Table 2. Cause of death for fishers and martens radio-collared<sup>1</sup> in Minnesota from 2007 - 2014.

Sex*Species	Predation	Natural Accident	Disease/ Illness	Unknown Natural	Car-Killed	Trapped In Season	Trapped Out of Season	Collar Complication	Unknown <sup>2</sup>	Total
Male Martens	22	2	1	4	0	34	2	1	0	66
Fem. Martens	33	0	0	2	0	13	2	2	1	53
Male Fishers	5	0	0	3	1	4	5	0	1	19
Fem. Fishers	20	0	0	2	1	4	3	0	2	32

<sup>1</sup> Excludes radio-collared animals that died within 2 weeks of capture and release

<sup>2</sup> Unknown represents animals where evidence was insufficient to assign to natural versus human-related cause.

# IDENTIFYING BARRIERS TO MOVEMENT AND THE EFFECTIVENESS OF CORRIDORS FOR CONNECTING CORE AREAS: LANDSCAPE GENETICS OF PRAIRIE GROUSE IN FRAGMENTED LANDSCAPES

Charlotte Roy, Eric Nelson<sup>1</sup>, and Andrew Gregory<sup>2</sup>

## SUMMARY OF FINDINGS

Cooperators and staff began collecting feathers at sharp-tailed grouse (*Tympanuchus phasianellus*) and greater prairie-chicken (*Tympanuchus cupido*) leks during the spring of 2014. Samples will be sent for genetic analysis at the close of the season. Feather collection will continue in 2015.

## INTRODUCTION

The grassland habitats that prairie grouse require have become increasingly fragmented as a result of competing pressures on the land (Berg 1997). Core habitat areas are isolated from each other by unsuitable areas that may prevent successful movement and the colonization of newly created habitat. The Minnesota Prairie Conservation Plan recognizes the importance of providing dispersal corridors to connect isolated core areas and identifies the greater prairie-chicken as an indicator species for upland prairie and grassland habitat (Minnesota Prairie Plan Working Group 2011). Similarly, sharp-tailed grouse must be able to move among isolated grassland, brushland, savanna, and peatland habitat patches (Berg 1997), through areas that may pose difficulty for successful movement. If the resistances of various landscapes to movement are understood, then more effective corridors can be identified, and management efforts can be prioritized using this information (Epps et al. 2007, Braunisch et al. 2010, Spear et al. 2010).

Landscape genetics is an emerging field that provides methods to examine connectivity on the landscape by combining a GIS with information about genetic variation in a population (Braunisch et al. 2010, Lowe and Allendorf 2010, Sork and Waits 2010, Haig et al. 2011). This tool can be used to examine effective dispersal (gene flow) on the landscape, without having to rely on telemetry techniques, which can be expensive and may require large numbers of marked animals if successful dispersal events are infrequent (Coulon et al. 2004, Spear et al. 2010). Landscape genetic methods have been used in recent years to identify barriers to dispersal, including human development, non-habitat land cover types, and distance in species like capercaillie (*Tetrao urogallus*, Braunisch et al. 2010), northern bobwhite (*Colinus virginianus*, Berkman et al. 2013a,b), and prairie-chickens (Gregory 2011). Thus, landscape genetics can be used to examine the movements of birds in a spatially explicit manner.

## OBJECTIVES

- 1- To identify barriers to movement for sharp-tailed grouse and greater prairie-chickens in Minnesota (e.g., distance, urban development, treed areas) as measured by genetic connectivity
- 2- To identify landscape features and types that enable movements of prairie grouse among areas of suitable habitat in Minnesota (e.g., agriculture) as measured by genetic connectivity

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- 3- To improve corridor planning and provide guidance to keep connected populations connected

## **METHODS**

Wildlife managers and cooperators surveyed prairie-chickens and sharp-tailed grouse at leks throughout Minnesota in the spring of 2014. Feathers were lost at leks during male contests, copulations, and as a result of other activities. Following completion of 1 survey at each lek (Figs.1 and 2), managers and cooperators collected feathers from dispersed locations on the lek. This maximized the probability of sampling different individuals. Each cluster of feathers, or single feather if necessary, was placed in an envelope and labeled with the lek location, date, and a unique number for each individual (e.g., UTM coordinates, date, species). Information from each envelope was recorded in a database, and the samples will be shipped to a commercial lab (Wildlife Genetics International, British Columbia) for extraction of genetic material when all samples have been received for the season.

If necessary, feather samples can be supplemented with hunter-harvested birds. Harvested samples can be restricted to adult birds which will be identified based on plumage characteristics (Bihrlé 1993), so that analyses are not confounded by the unknown dispersal status of juveniles. The sex of birds can be determined molecularly (Fridolfsson and Ellegren 1999), if not possible from available plumage (Bihrlé 1993).

At the commercial genetic lab, genetic material will be extracted and amplified at numerous microsatellite loci. Microsatellites are highly variable, neutral (non-coding) genetic loci. Recent studies of prairie-chickens and sharp-tailed grouse have identified microsatellite loci that are polymorphic in these species (and populations, see citations in Gregory 2011 and Malone 2012). This genetic information can be linked to spatial information to examine the connectivity of the landscape. Areas that share greater connectivity will be similar genetically, whereas areas with restricted connectivity will be more dissimilar genetically. Analytical methods will be revisited for the most recent advances prior to initiating data analysis.

## **DISCUSSION**

Results of this study will provide information about landscape features that isolate habitat fragments and those that serve to promote connectivity. Following the methods of Gregory (2011) or Barton et al. (2010) we can also use the described landscape genetic analyses to understand the relative influence of different landscape elements to promote or inhibit dispersal. This information will be useful to target management efforts in ways that can more effectively accomplish the goal of connecting core areas, enhancing local habitat conditions, and providing new habitat sufficiently close to existing leks to promote colonization.

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Figure 1. Prairie-chicken leks ( $n = 188$ ) surveyed in northwestern Minnesota during 2013. County boundaries and Minnesota GAP cover classes are provided for context. These leks will be the focus of feather collection efforts in 2014 and 2015.

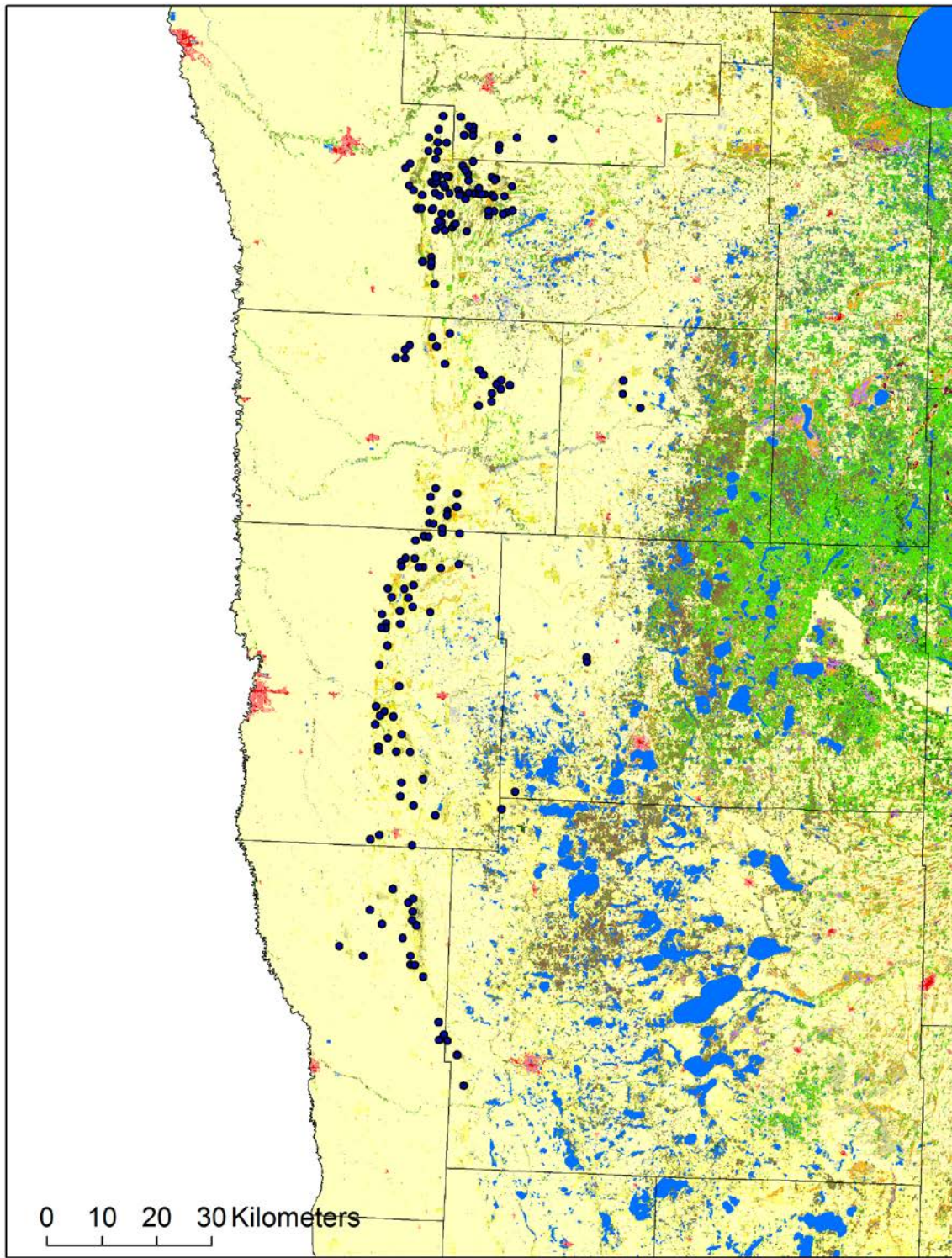
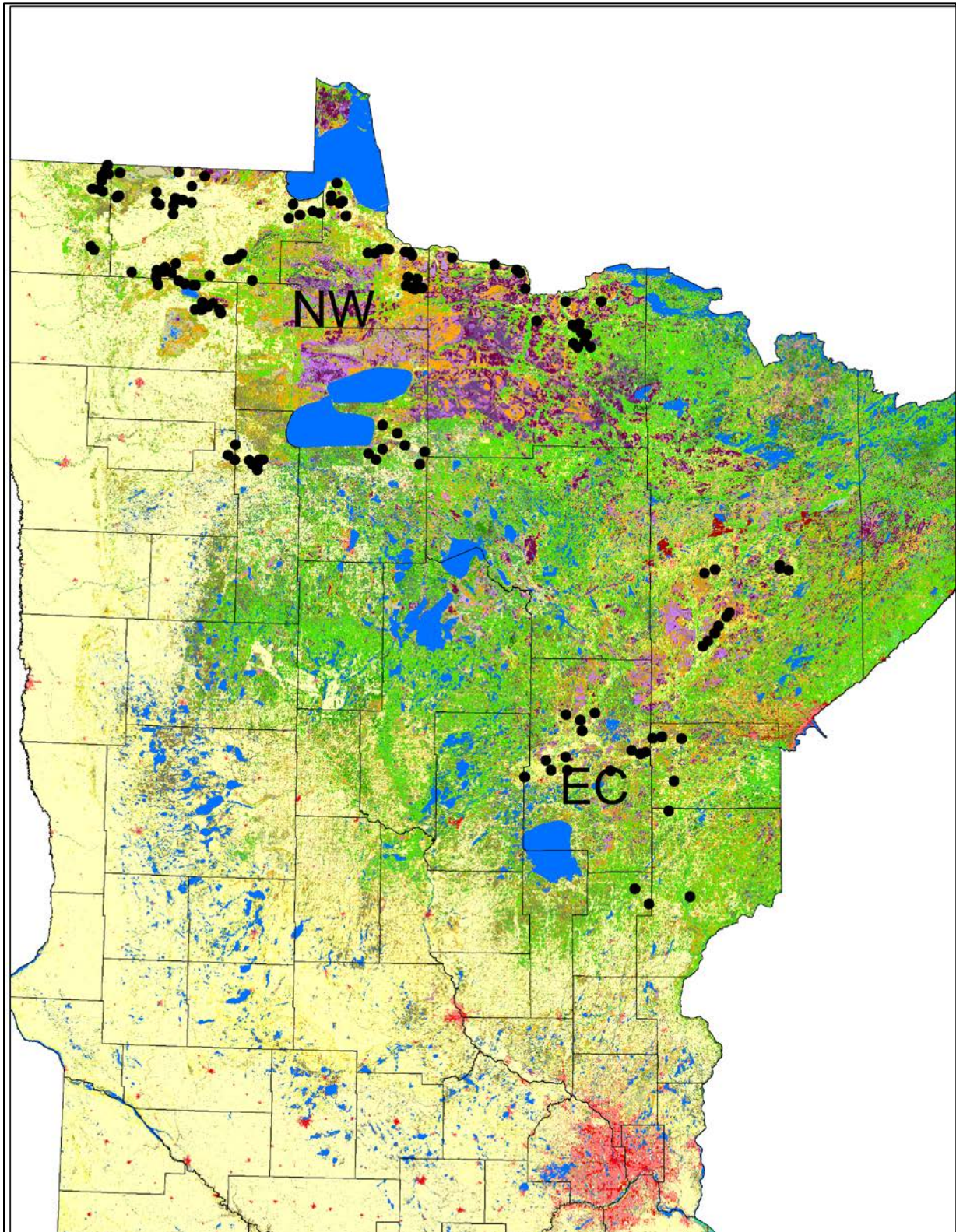




Figure 2. Sharp-tailed grouse leks ( $n = 139$ ) surveyed in northwestern (NW) and east-central (EC) Minnesota during 2013. County boundaries and Minnesota GAP cover classes are provided for context. These leks will be the focus of feather collection efforts in 2014 and 2015.



# DEVELOPING SURVEY AND STUDY METHODOLOGY FOR SPRUCE GROUSE: A PILOT STUDY

Charlotte Roy, Mike Larson, and John Giudice

## SUMMARY OF FINDINGS

Data collection began in April 2014 for a 2-year pilot study. Results will be forthcoming in future reports.

## INTRODUCTION

The spruce grouse, *Falci pennis canadensis*, is a boreal forest inhabitant existing at the southern edge of its range in the Lake States of northern Minnesota, Wisconsin, and Michigan. Spruce grouse occupy forest stands with a variety of coniferous tree species including spruce, fir, pine, hemlock, and tamarack, and habitat selection varies with the forest community in various portions of spruce grouse range (Boag and Schroeder 1992). In the Lake States, spruce grouse prefer areas of black spruce (*Picea mariana*) with some jack-pine (*Pinus banksiana*) interspersed (Robinson 1969, Pietz and Tester 1982, Anich et al 2013), but have also been reported using balsam fir (*Abies balsamea*), tamarack (*Larix laricina*), eastern hemlock (*Tsuga canadensis*) and northern white cedar (*Thuja occidentalis*). In nearby Wisconsin, closed canopy coniferous uplands near lowland conifer swamps were important, especially mature black spruce-tamarack swamps and jack pine 16-29 years old (Anich et al. 2013). Similarly in Minnesota, but with a more modest sample, spruce grouse used lowland conifers with black spruce and mixed bogs during the breeding season, and used jack pine during winter (Pietz and Tester 1982). Boag and Schroeder (1992) indicated that early successional stands were preferred but Anich et al. (2013) found mature forest to be important.

Spruce-fir forests are predicted to be vulnerable to climate change at the southern edge of their range (Scheller and Mladenoff 2005, Prasad et al. 2007, Iverson et al. 2008). Thus, spruce grouse are expected to experience a range contraction due to climate change-induced habitat loss, and will likely have a more limited distribution in Minnesota in the future. A northward shift in spruce grouse distribution is already underway in the state, as they once occurred in Nemadji State Forest (Pine and Carlton Counties), Itasca State Park (Clearwater County), and Hubbard County (W. Berg, pers. comm., Janssen 1987).

Minnesota is unique among the Lake States in having a sizeable spruce grouse population that still permits spruce grouse hunting. The spruce grouse is considered a Species of Special Concern in Michigan (Michigan DNR 2005) and was listed as threatened in Wisconsin in 1997 (Wisconsin DNR 2004). Yet, the only data we collect for spruce grouse is part of the Small Game Harvest Mail Survey (Dexter 2013), which provides an estimate of harvest reported by hunters between 9,000 and 27,000 birds per year over the last 10 years (Dexter 2013). However, spruce grouse harvest is more reflective of ruffed grouse hunter numbers than spruce grouse numbers, and thus these data cannot be used as a population index (Gregg et al. 2004). Thus, we have sparse data on spruce grouse distribution, and no data on population size or trends, despite a responsibility to manage spruce grouse during a period of expected habitat loss.

Minnesota currently has long term survey efforts for all grouse species except spruce grouse, including annual lek surveys of prairie grouse and annual roadside drumming counts of ruffed grouse (*Bonasa umbellus*). Other states like New York (Fritz 1979) and Wisconsin (Worland et al. 2009) have executed surveys of spruce grouse. However, these surveys were conducted during a few years, were labor intensive, and were not designed to be long term monitoring projects. Any long term, large scale monitoring effort of spruce grouse in Minnesota

would need to be easy to execute, repeatable, and representative of spruce grouse populations. Logistical constraints of doing large scale surveys often constrain survey design. In this case, spruce grouse occupy habitats that are very difficult to access away from roads. A roadside survey would possess the logistical ease desirable for a statewide effort, but several potential biases would need to be understood and addressed.

Surveys for spruce grouse are usually conducted using playback of female cantus calls (Fritz 1979, Boag and McKinnon 1982, Whitcomb et al. 1996, Lycke et al. 2011, among others). Schroeder and Boag (1989) compared counts obtained with playback of female calls to counts from a wing-clapping index and concluded that 'the female call may be a useful compromise between the accuracy of a complete count and the time efficiency of the wing-clapping index' and that both were useful for estimating the size of male spruce grouse (Franklin's subspecies) populations in Alberta. Because spruce grouse of the subspecies in Minnesota do not execute the wing-clap display, we will use playbacks of cantus calls to survey spruce grouse.

## **OBJECTIVES**

1. Assess the feasibility of a roadside survey for spruce grouse to determine distribution and population trends of spruce grouse
2. Estimate capture success and identify constraints to radio-tracking for future study

## **STUDY AREA**

The pilot survey will be conducted on Red Lake Wildlife Management Area (WMA) and Beltrami Island State Forest. This study area is on the southwestern edge of the assumed current range of spruce grouse, where changes would be expected to occur before changes more centrally within their Minnesota range. The long term goal is to expand the survey throughout spruce grouse range in northern Minnesota, if the survey is deemed feasible, and for it to be repeated at a regular interval (to be determined).

## **METHODS**

We developed a pilot spruce grouse survey using playback of female cantus calls (Fritz 1979, Boag and McKinnon 1982, Schroeder and Boag 1989, Whitcomb et al. 1996, Lycke et al. 2011). In spring 2014, we attempted to survey as many survey routes/points as possible to inform point survey duration (1-15 min), the duration of time it takes to complete a route, and the duration of responsiveness of spruce grouse to cantus calls (both time of day and duration of season). The duration of responsiveness to playback varies among years depending on spring phenology (i.e., shorter period of responsiveness in early springs, Anich, pers. comm.), which is consistent with findings in our study area by Larson (unpublished data) in 2011 and 2012. The duration of responsiveness may also vary throughout the day, although Lycke et al. (2011) determined that birds responded to cantus calls between sunrise and noon.

Survey routes (10 points per route) were placed along low maintenance road segments that bisected or were adjacent to forest stand types known to be used by spruce grouse in the Lake States in the spring (i.e., mature black spruce and tamarack swamps, and young jack pine; Robinson 1969, Pietz and Tester 1982, Anich et al 2013; Fig. 1). Points were spaced  $\geq 300$  m apart along routes to ensure independence among points, based on estimates that playback can be heard 100-150 m from the speaker (Schroeder and Boag 1989, Lycke et al. 2011, Anich, unpubl. data). Surveys were conducted during April and May beginning at sunrise. We conducted surveys when winds were  $< 10$  mph and when precipitation was absent or light. Each point count lasted 15 min (Lycke et al. 2011, Anich et al. unpubl. data) with broadcast of the

cantus call once each minute. Intervals were subdivided into five 3-min intervals, and we recorded both new detections and redetections of each bird during each interval for time-to-detection analysis (Alldredge et al. 2007). The distance to each bird detected was recorded in <50, 50-100, 101-150, >150 m intervals. Each observer recorded date, arrival time at each stop, approximate wind speed, temperature, type of forest (jack pine, black spruce, tamarack, white cedar, red pine, other), type of detection (flutter flight, approach, etc.) and background noise (e.g., wind) on a relative scale (e.g., none, low, medium, high).

After the completion of playback surveys, we surveyed the same points with trained dogs and their handlers and attempted to locate all birds <150 m from the survey point. Other researchers have had success using dogs to locate spruce grouse (Robinson 1969, Keppie 1987, Ratti et al. 1984). We attempted to quantify the variability in skill among dogs by surveying the same points with multiple dogs. We also incorporated a grouse pellet survey along a circular transect 75 or 100 m from each point, with transect length depending on the distance that dogs roamed from their handlers, so that both surveys could be completed at the same time. We counted all grouse pellet piles within 1 m of the traversed path and calculated the length of the path traversed. We distinguished ruffed grouse pellets from spruce grouse pellets on the basis of size, shape, and color.

## **Data Analysis**

We plan to use time-to-detection methods to estimate available birds (Alldredge et al. 2007, Johnson 2008). Time-of-detection methods treat each time interval as a separate trapping occasion in a mark-recapture framework that can be analyzed in Program MARK (White and Burnham 1999). Auditory surveys will be compared to data from dogs and pellet surveys using double-sampling techniques (Cochran 1977, Thompson 2002). We will use results of this pilot study to determine whether a statewide survey is feasible by examining the number of routes and stop locations necessary to estimate population trends and distribution statewide.

## **DISCUSSION**

The goal is to develop a statewide survey that would provide an index of population size that could be used to estimate the trend over time and monitor changes in distribution. Current predictions of climate change suggest that the impending impact on spruce grouse will be large. Activities related to the second objective will begin the second year of the pilot study.



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## **DETERMINING CAUSE- SPECIFIC MORTALITY IN MINNESOTA'S NORTHEAST MOOSE POPULATION**

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### **SUMMARY OF FINDINGS**

The primary goal of this study is to improve our understanding of non-anthropogenic (i.e., health-related) mortality of the northeastern Minnesota's declining moose population. Our objectives are to determine causes of non-hunting mortality (i.e., identify specific disease or parasite agents, predation), by responding to mortalities within 24 hours of death, prior to decomposition of tissues. In the first 17 months of this multiyear study, we've captured and radio collared 142 adult moose (mean age = 6 years, range 1 to 14) and a total of 35 collared moose have died since this study began, which included 7 capture-related mortalities that will be censored from subsequent survival analyses. Overall causes of death are as follows: 15 predator-related mortalities (54%), which includes 9 wolf kills, 3 likely wolf kills, and 3 wolf-caused injuries that led to secondary lethal bacterial infections; and 13 health-related mortalities (46%), which includes 3 brainworm (*Parelaphostrongylus tenuis*) infection, 3 winter tick (*Dermacentor albipictus*) infestations, 1 liver fluke (*Fasciola hepatica*) infection with secondary bacterial infection, 1 trauma with associated septicemia, 1 accident (drowning), 1 kidney disease with cachexia, and 3 undetermined health issues. Whole carcasses were retrieved for 9 of these moose, with field necropsies performed on remaining 19 animals. Response times from initial mortality notification (e.g. text message or email) to a team in the field at the death site were  $\leq 24$  hours in 16 cases (58%), between 24 and 48 hours in 6 cases (21%), and  $>48$  hours in 6 cases (21%). Delays in mortality responses  $>24$  hours have been due to collar or technology failures and wolves actively feeding on the moose carcass and thus preventing the collar from sending a mortality alert.

### **INTRODUCTION**

Historically, moose were found throughout the forested zone of north central Minnesota. By the 1960's there were two distinct populations, the northwest (NW) population of the aspen parklands and northeast (NE) population of the boreal forest (Fuller 1986). In the mid-1980's the NW population began a precipitous decline, falling from 4,000 to  $<100$  animals by the early 2000's (Murray et al. 2006, Lenarz 2007). Murray et al. (2006) identified pathogens, including liver flukes (*Fascioloides magna*) and brainworm (*Paralaphostrongylus tenuis*), as the principal cause of death for 37-62% of radio-collared animals; 25% of additional mortalities were likely pathogen-induced, but limited necropsy evidence was inconclusive. They also observed that many moose in NW MN dying of natural causes were malnourished, as evidenced by 51% of

carcasses having bone marrow fat (BMF) contents below a critical threshold (< 30%) and trace mineral deficiencies (i.e., copper and selenium).

Subsequently, in NE MN, Lenarz et al. (2009) reported a 21% average non-hunting mortality rate for radiocollared moose, which was much higher than the 8-12% reported for moose elsewhere in North America (Larsen et al. 1989, Ballard 1991, Kufeld and Bowden 1996). Specific causes of most of the non-anthropogenic mortality (89%) could not be determined, as assessing cause-specific mortality was not the primary objective of the study (Lenarz et al. 2009). Many of the deaths appeared health-related, with prime age animals dying during unusual times of the year or carcasses found intact with little evidence of scavenging.

Aerial surveys also indicate the NE population is declining. Since the estimated peak at 8,840 moose in 2006, the 2014 estimated moose population (4,350) is 50% lower and time series analysis of estimates since 2005 indicate a significant downward trend (DelGiudice 2013). Butler et al. (2013a) documented evidence of exposure of NE MN moose to a variety of disease agents (e.g., West Nile Virus, eastern equine encephalitis, malignant catarrhal fever), which could be potential mortality factors. Additionally, a recent study of sick and vehicle-killed moose ( $n=62$ ) from 2003-2013 had found 85% of animals were undernourished and infected with a variety of disease agents, including brainworm (45%), liver flukes (60%), and winter ticks (*Dermacentor albipictus*) (22%) (Wuenschmann et al., 2014). Researchers have hypothesized that brainworm was responsible for historic declines in moose populations (Karns 1967, Prescott 1974, Lankester 1987), but it is questionable whether brainworm currently represents a major threat to the NE MN population; clinical signs consistent with brainworm infection were first reported in MN moose in 1912 (Fenstermacher and Olson 1942). Lenarz et al. (unpublished data) found that brainworm may have caused an average 19% (0-32%) of the population's total annual mortality.

## METHODS

Moose were captured within the study area (Fig. 1) by aerial darting (Quicksilver Air Inc., Alaska) with carfentanil (4.5mg or 6.0mg), thyaferentanil (16mg) and xylazine (150mg or 30mg) from a helicopter; immobilizations were reversed with naltrexone (425-575mg) and tolazoline (400mg). Blood (serum and whole blood) was collected at capture by venipuncture of the jugular vein. Serum was screened for evidence of exposure to 10 disease agents following the same protocol as described by Butler et al. (2012b). Additionally, serum was submitted for a large animal serum chemistry profile and reproductive hormones to assess physiological status, overall health, and pregnancy status (Franzmann and LeResche 1978, Haig et al. 1982, Duncan et al. 1994). Serum progesterone levels were determined by the Smithsonian Institute; levels >2.0 ng/mL were considered pregnant. Whole blood in Ethylenediaminetetraacetic acid (EDTA) was used to make blood smears and complete and differential blood cell counts were performed, which may be indicative of condition and health status (Duncan et al. 1994), presence of tick-borne illnesses, and evaluation for the presence of microfilaria. An incisor (I4) was removed for aging by cementum annuli (Sergeant and Pimlott 1959). A general fecal floatation examination for parasites was performed. A thorough physical examination was performed, including assessment of body condition score (very thin, thin, normal, fat), winter tick load, and hair loss. Total body length and girth (cm) were measured to estimate body weight of

moose (Hundertmark and Schwartz, 1998) and hair samples were collected from the withers. Any mortalities that occurred within two weeks of capture were censored from the study.

Moose were fitted with mortality-sensing collars utilizing Global Positioning System (GPS) and Iridium two-way communication technologies (Vectronic Aerospace GmbH; Berlin, Germany). Collars transmit location and status data to a base station (Forest Lake, MN) at user-defined intervals. The base station also analyzes location data to identify animals that have “localized” (e.g., remained within a 20m radius for >24 hours), to assist with detecting sick animals that are potentially moribund. When a mortality or localization event occurred, the mortality response team was notified via text and email messages. Mortality implant transmitters (MITs; Vectronic Aerospace GmbH) were placed orally into the reticulum of a subset of the captured moose. These devices are similar to a cow magnet in size, log internal temperatures every 15 minutes, and transmit a subset of this data through the collar. Additionally, MITs are meant to provide immediate notification of mortality via detection of minimal internal activity (e.g., lack of a heart beat) and this notification is also sent via text and email message to the moose mortality response team. External temperature loggers (Hobo TibdbitV2; Onset Corporation, Bourne, MA) were affixed to the GPS collar and were programmed to collect ambient temperature every 60 minutes.

Moose mortality response teams have 8 primary team leaders that have undergone extensive necropsy training, and they are supported by about 30 secondary and tertiary team members (including Minnesota Department of Natural Resources [MNDNR], tribal, academic, and other personnel) available upon request. Every effort is made to remove carcasses intact from the field and deliver them to the University of Minnesota Veterinary Diagnostic Laboratory (UMN VDL) for a complete necropsy by a board-certified pathologist. Teams are able to utilize special equipment, including trucks with 2000lb winches, an amphibious ARGO, chainsaw winch (e.g. Lewis winch), heavy duty snow machines with long tracks, all-terrain vehicles, and specialized rubber mats with built-in hitches for dragging carcasses. Primary members have also taken specialized training with DNR forestry and fire units to be able to sling out a moose carcass via helicopter. If a moose was found to be alive, but obviously ill, it was euthanized (via gunshot). If carcass extraction is not possible, a thorough and complete field necropsy was performed, guided by an established protocol, and samples were submitted to the UMN VDL for diagnostic evaluation (Butler et al., 2013b).

## **RESULTS AND DISCUSSION**

### **Overall survival and timing of mortalities**

A total of 28 collared moose have died since this study began; which excludes 7 capture-related mortalities that are censored from subsequent survival analyses. Overall survival rate is 74.6% from February 4, 2013 to June 30, 2014. Causes of death are as follows: 15 predator-related mortalities (54%), which includes 9 wolf kills, 3 likely wolf kills, and 3 wolf-caused injuries that led to secondary lethal bacterial infections; and 13 health-related mortalities (46%), which includes 3 *P. tenuis* infection, 3 winter ticks, 1 liver fluke infection with secondary bacterial infection, 1 trauma with associate septicemia, 1 accident, 1 was cachectic with kidney disease and endometriosis, and 3 undetermined health issues (Fig 2). Timing of these mortalities

suggest that most deaths occur in spring (64%, March–May); however, moose died in all seasons (Winter 18%, Summer 14%, and Fall 4%) (Fig 3).

### Annual variation in survival

The annual survival rate for adult moose ( $n=107$ ) in Year 1 (4 February, 2013–5 February, 2014) of this study was 80% (95%CI = 72.0 to 87.9%; Fig 4). A total of 21 collared moose (17 females, 4 males) died. There were 78 moose remaining with active collars, excluding censored animals ( $n=12$ ) at the end of Year 1. Causes of death are as follows: 11 predator-related mortalities (52%), which includes 6 wolf kills (3 of which had predisposing health conditions that likely contributed to their death), 3 likely wolf kills, and 2 wolf-caused injuries that led to secondary lethal bacterial infections; and 10 health-related mortalities (48%), which includes 2 *P. tenuis* infection, 3 winter ticks, 1 liver fluke infection with secondary bacterial infection, 1 trauma with associate septicemia, and 3 undetermined health issues. Whole carcasses were retrieved for 8 of these moose, with field necropsies performed on the remainder. A total of 12 collared moose were censored in Year 1 due to capture-related mortalities ( $n=4$ ), slipped collar ( $n=1$ ), and collars that stopped transmitting ( $n=7$ ).

From February 6-11, 2014, 31 additional moose (12 males, 19 females) and 5 female moose with failed collars (originally captured in 2013) were captured and recruited into the study. Sample size for the start of Year 2 was 104 collared moose. As of June 30, 2014, survival rate is 93.4% (Fig 4). A total of 7 collared moose (6 females, 1 male) have died. Causes of death are as follows: 4 predator-related mortalities (57%), which includes 3 wolf kills and 1 wolf-caused injury that led to a secondary lethal bacterial infection; and 3 health-related mortalities (43%), which includes 1 *P. tenuis* infection, 1 accident (fell through the ice and drowned), and 1 moose that was cachectic with kidney disease and endometriosis. Nine collared moose from Year 2 are censored thus far, which includes 3 capture-related mortalities, 1 slipped collar, and 5 collars that stopped transmitting. Thus far in Year 2 (from capture to 30 June) moose survival is 2x higher than the same time period in Year 1. It appears that winter survival was enhanced by the prolonged winter 2013, which may have suppressed winter tick numbers, as we have not reported any winter tick mortalities during winter 2014. Also, the historically severe winter of 2014 likely reduced deer numbers in our study area, which would lessen disease exposure risks of moose to *P. tenuis* and liver flukes.

### Serologic and Parasite Screening at Capture

Progesterone levels from blood samples obtained during 2014 captures indicated a 77% pregnancy rate; similar to 75% from last winter (Butler et al., 2013b). Biological samples obtained during 2014 captures were analyzed and serologic evidence of exposure to West Nile Virus (8 of 34, 23.5%), malignant catarrhal fever (1 of 34, 2.9%), various serovars of *Leptospira interrogans* (6 of 34, 17.6%) and *Borrelia* (2 of 34, 5.9%) was documented. Fecal floatation was used to screen moose at capture for parasites, and 3 of 28 (11%) were identified as having infections. These parasites included *Nematodirus* sp. (2 moose) and coinfections with Strongyle-type ova (1 moose). Analyses of the CBC and serum chemistry results are pending. Mean age of captured moose was 5.3 years ( $n=23$ , range 1 to 13). The body condition score of each moose was evaluated and recorded whenever possible; 6 were classified as thin (16.7%), 21 as normal (58%), and 9 as fat (25%). None of the animals were classified as very thin. Comparatively, during 2013 captures, a third of all moose captured were either thin or very thin (Butler et al. 2013b). Estimated mean body mass at capture was 395kg ( $n=29$ , range 336-461) in Year 2, compared to 388kg ( $n=99$ , range 269-522) in Year 1.

## **Mortality Response Times**

Whole carcasses were retrieved for 12 of these moose, with field necropsies performed on the remaining 23 moose. Response times from initial mortality notification (e.g. text message or email) to a team in the field at the death site were  $\leq 24$  hours in 20 cases (57%), between 24 and 48 hours in 9 cases (26%), and  $>48$  hours in 6 cases (17%). Delays in mortality responses  $> 24$  hours have been due to collar failures and wolves actively feeding on the moose carcass and thus preventing the collar from sending mortality alert.

## **Collar and MIT Functioning**

Of the 100 collars originally purchased by the MNDNR, 41 have either had mortality signal malfunctions (e.g., collars being locked in mortality mode) or stopped transmitting entirely, and have been replaced under warranty by the manufacturer. The collars locked in mortality (LIM) mode are transmitting locational data; however, staff have to monitor these animals ( $n=21$ ) daily to ensure movement is occurring. To assist with this more intensive monitoring, LIM collars collect and transmit data more frequently and the localization function evaluates movements more often (12 hour intervals). The localization function has increased our ability to recognize animals that are moribund, but are not actually dead, allowing us to euthanize the animal. This allows never before documented clinical signs to be observed (e.g., teeth grinding in a *P. tenuis* infected moose) and key samples (e.g., fresh blood) to be collected, which is vital when trying to determine cause of death. This function has also helped us identify wolf-kills faster, as wolves will feed under the collar and prevent it from going into mortality mode until they have left the carcass.

Initially, we deployed 28 MITs in Year 1 and their functionality is as follows: 4 MITs were spit out by the moose immediately after deployment, 2 stopped functioning after 14 months (presumably failed batteries), 6 were recovered in dead moose, and 16 are currently actively deployed and operational. In Year 2, we attempted to deploy additional MITs and 12 were spit out by moose immediately after deployment (in some cases, capture team witnessed this and recovered MIT) and 18 were successfully deployed. Currently, there are 32 radio collared moose with functioning MITs alive in the study.

## **Preliminary Animal Migration Behavior Analyses**

Year 1 moose movement data were analyzed to assess migration behavior. Out of 70 active moose that survived through the first year of the study, 34% ( $n=24$ ; 17 females, 7 males) migrated to a distinct spring-summer-fall (SSF) range. Average distance from winter range to SSF range was 24km (12.8km for females, 19.9km for males). Average SSF home range (MCP) was 23.8km<sup>2</sup> for females, and 51.8km<sup>2</sup> for males. Average winter home range (MCP) was 11.2km<sup>2</sup> for females and 47.5km<sup>2</sup> for males. Average home range (MCP) for non-migrating moose ( $n=46$ ) was 74.3km<sup>2</sup>. A preliminary analyses of cover types within these moose home ranges suggest that woody wetlands are used the most by moose at all times of the year, but far

more (21.6% higher) in the summer than in winter; moose are using deciduous and coniferous more in the winter. Further analyses of habitat use and movement data are pending.

## **Funding and Future Project Direction**

This \$1.2 million dollar project initially began through funding from the Legislative Citizen Commission on Minnesota's Resources (LCCMR; \$600,000) and in-kind contributions from the MNDNR, University of Minnesota, tribal partners, and nonprofit organizations (e.g., MDHA, Northstar Museum, and Save Minnesota Moose). Additional funding through LCCMR was recently secured for \$600,000 to continue the study and expand on the use of MITs to gain insight at the potential role ambient temperature may play in moose survival and productivity. Sample size of at least 100 collared adult moose will be maintained through 2015 and 2016 by capturing new individuals as needed over the next 2 winters. Further, 30 additional MITs will be deployed. New methods for successful MIT delivery and a calibration of the internal temperature data will be developed in conjunction with the Moose Research Center in Alaska in 2015.

## **ACKNOWLEDGEMENTS**

This project is very demanding and would not be possible without the assistance of the following groups and individuals: the Environmental and Natural Resources Trust Fund for funding the majority of this project, Dr. Arno Wuenshmann and Dr. Anibal Armien (UM VDL) for their diagnostic investigations of the mortalities, Mike Schrage (Fond du Lac Natural Resources) and Andy Edwards (1854 Treaty Authority) for their assistance in the field and during captures, Richard Gerhold and Caroline Grunenwald (University of Tennessee) for assisting with the identification of microfilaria, Ulrike Munderloh (University of MN, Department of Entomology) for testing samples for tick-borne illness, J. P. Dubey (USDA, ARS) for neospora testing, our team of primary responders (Nancy Hansen, Dave Ingebrigtsen, Dawn Plattner, and John Giudice; MNDNR), our team of secondary responders (Bob Fashingbauer, Bob Kirsch, Bryan Lueth, Carolin Humpal, Jim LaBarre, Leslie McInenly, Lindsay Shartell, Meadow Kouffeld-Hansen, Steve Piepgras, Tim Pharis, Tom Rusch, and Jeff Hines; MNDNR), Dan Ryan and Dave Grosshuesch (US Forest Service), Brandon Seitz (Grand Portage National Monument), EJ Issac and Seth Moore (Grand Portage Band), and Lance Overland (Fond du Lac Resources), Nick Bogyo (1854 Treaty Authority) for their assistance in the field, and the MNDNR enforcement pilots (Jason Jenson, John Heineman, Tom Bucker, and Bob Geving) for their assistance during captures, USDA-Wildlife Services (Paul Wolf) for use of their necropsy trailer, and Tyler Obermoller, Kaytee Firnett, Jeanna Lodel, Beth Martin, Amanda McGraw, and Amy Kingsley for assistance with data management and gearing-up for captures. Rob Fasteland (MNDNR Forestry) and the Lake & Cook County Highway Department staff for snow plowing and maintaining helispots used during capture events. Special thanks to special operations staff for remote hook/sling and radio training, including Bill Schuster, Lee Kessler, Mike McLaughlin, and Pat Coughlin. This project was also funded in part by the Wildlife Restoration Program (Pittman-Robertson).

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Figure 1. Study area in northeast Minnesota where 142 moose have been captured and radio collared (2013–2014) to study cause-specific mortality.

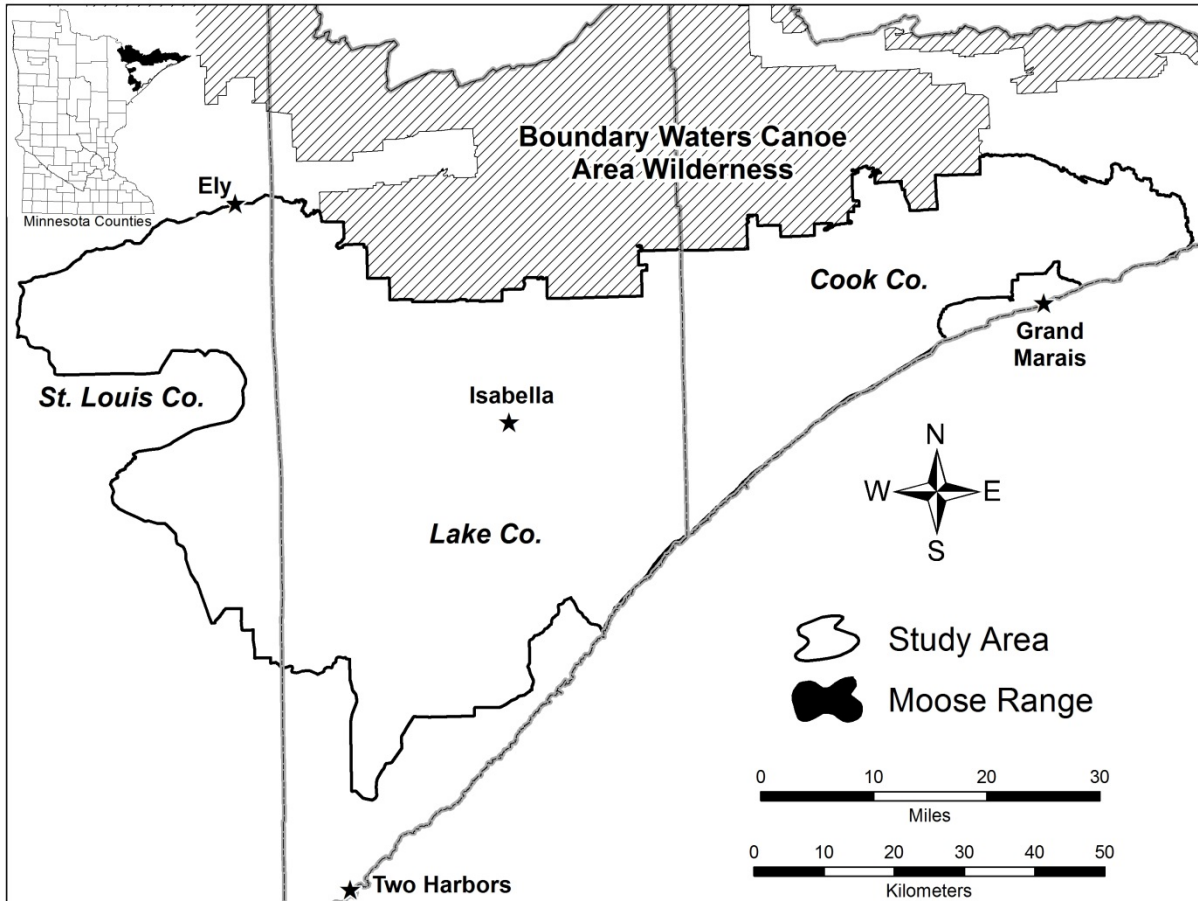


Figure 2. Cause-specific mortality of radiocollared, adult moose ( $n=28$ ) from February 2013 to June 2014, northeast Minnesota.

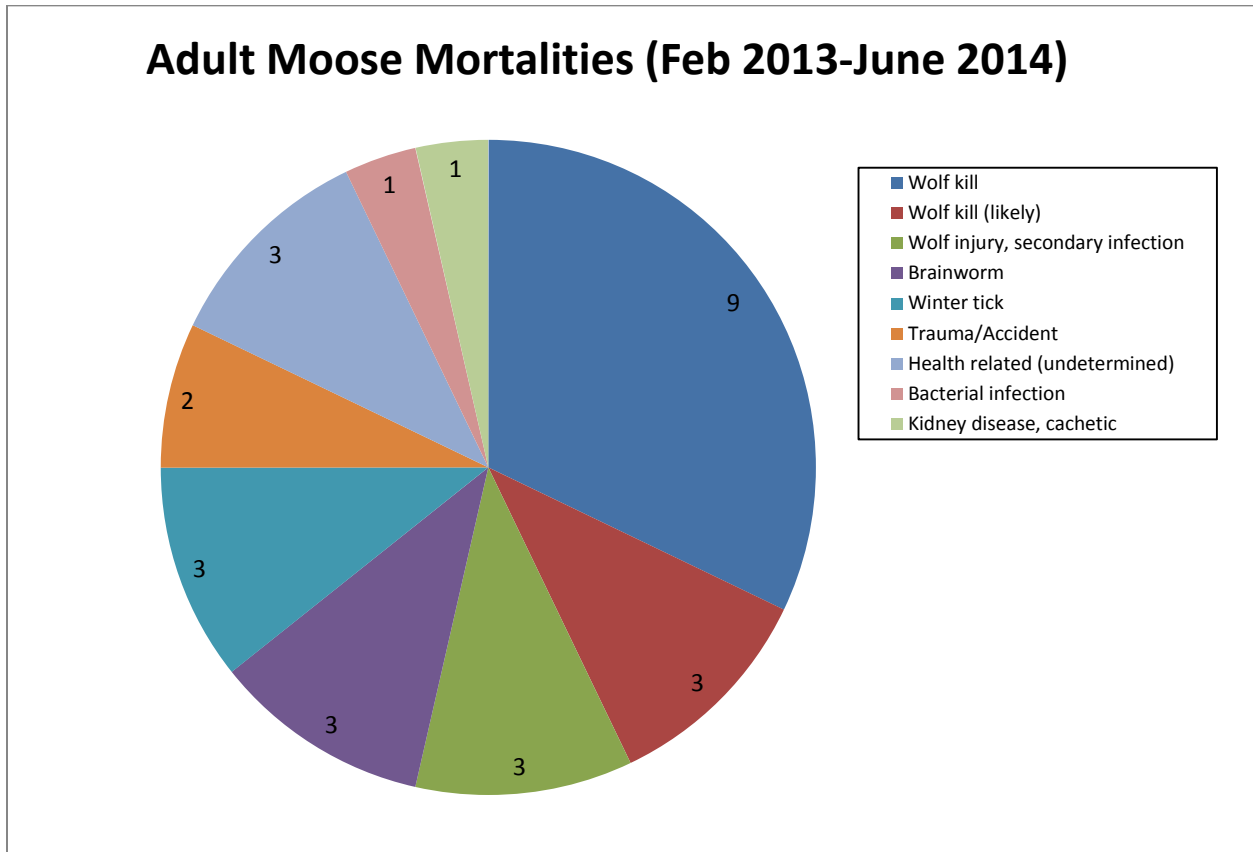


Figure 3. Timing of mortalities for adult moose ( $n=28$ ) from February 2013 through June 2014, northeast Minnesota.

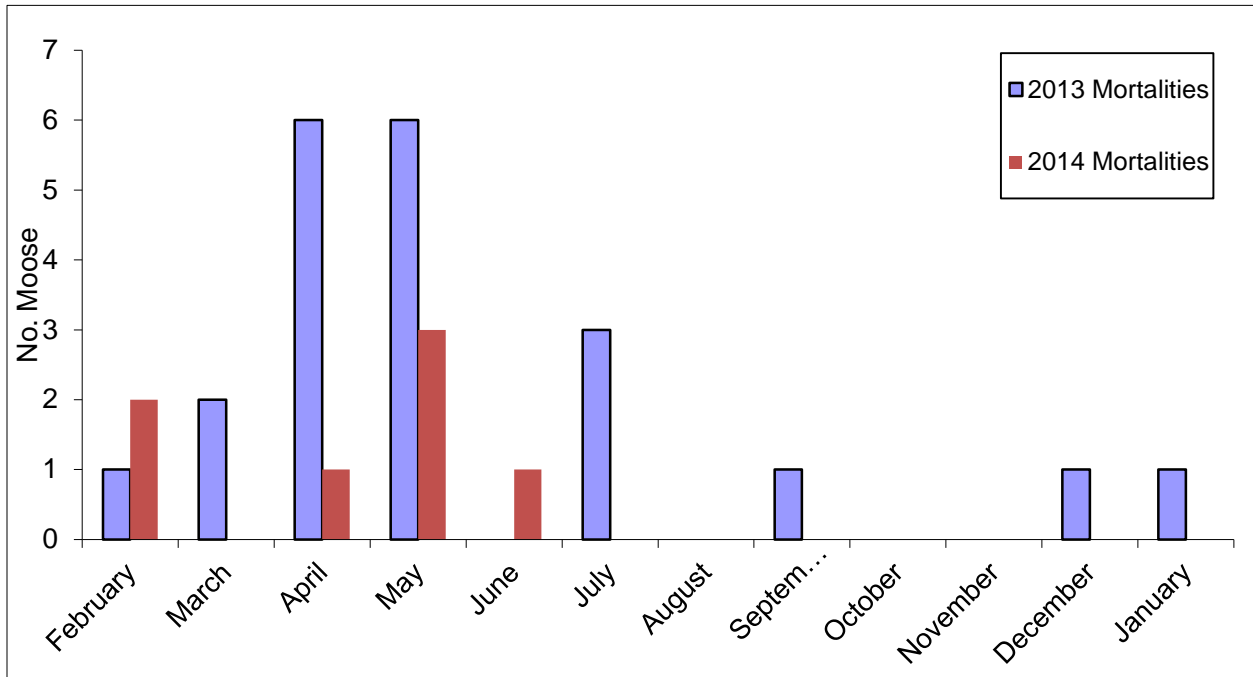
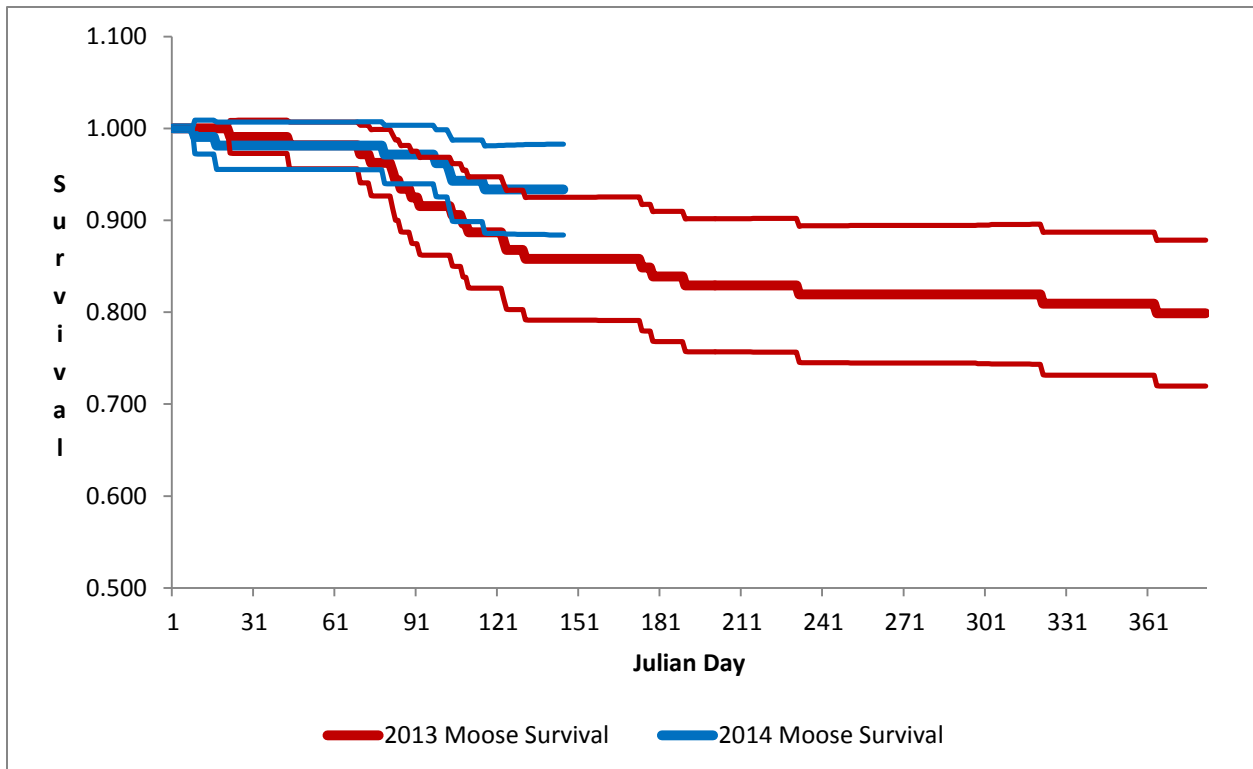


Figure 4. Annual survival of radio-collared moose captured in 2013 and 2014, with 95% confidence intervals included.





## CHRONIC WASTING DISEASE MANAGEMENT IN A MINNESOTA DEER HERD: COORDINATED RESPONSE TO THE SOUTHEAST DETECTION, 2010–2013.

Erik Hildebrand<sup>1</sup>, Michelle Carstensen, Lou Cornicelli, David C. Pauly, and Margaret H. Dexter

### INTRODUCTION

In November 2010, the first chronic wasting disease (CWD) positive wild white-tailed deer (*Odocoileus virginianus*) was detected through hunter-harvested surveillance by the Minnesota Department of Natural Resources (MNDNR) in southeastern Minnesota. Surveillance efforts were focused within an area surrounding a CWD-positive captive elk facility in Pine Island, which was discovered infected with the disease in early 2009. This captive elk herd, comprised of about 600 animals, was subsequently depopulated through federal indemnification, and a total of 4 elk were confirmed with the disease. Epidemiological investigations conducted by the United States Department of Agriculture (USDA) and Minnesota Board of Animal Health (BAH) concluded that there was an apparent longstanding infection within this captive elk facility. This wild deer index case was located within 3 miles of the CWD-positive captive elk facility.

In response to this disease detection, MNDNR enacted its CWD Response Plan (available at: [Minnesota DNR CWD response plan](#)). Initial aerial survey results indicated a high density of deer in the area near Pine Island (31 deer/km<sup>2</sup>) as well as numerous recreational feeding sites. The combination of these two factors heightened concern about disease transmission potential. A ban on recreational deer feeding was immediately enacted in a 4-county area, and a supplemental surveillance effort was conducted in February–April 2011 to improve our understanding of disease prevalence and distribution in the local deer herd. To prevent further disease spread the MNDNR (1) created a CWD Management Zone (Deer Permit Area (DPA) 602), (2) restricted whole-carcass movements outside of the zone, and (3) required mandatory sampling of all adult ( $\geq 1.5$  years of age) deer harvested by hunters (4) continued aggressive disease surveillance of hunter-harvested deer in the CWD Management Zone for 3 consecutive years (2011–2013). No additional cases of CWD were discovered in wild deer. Surveillance efforts are complete and DPA 602 was dissolved for the 2014 deer hunting season. In this paper, we describe how MNDNR responded to this detection of CWD in wild deer, including surveillance methods, management strategies, costs, and lessons learned.

### METHODS

#### 2011 Aerial Deer Survey, Recreational Feeding Activity, and Winter

Prior to beginning an intensive sampling effort, MNDNR used a fixed-wing aircraft to conduct an aerial survey of the CWD surveillance area to assess deer numbers and distribution. This survey was conducted in late January-early February 2011, with estimates of 6,200 deer within the 793-km<sup>2</sup> (306-mi<sup>2</sup>) winter surveillance area, equating to an estimated 7.3 deer/km<sup>2</sup> (19 deer/mi<sup>2</sup>) density (Figure 1). Deer densities were highest within a 23-km<sup>2</sup> (9-mi<sup>2</sup>) area surrounding the wild index case; 600 deer were counted using a helicopter census and estimated  $>31$  deer/km<sup>2</sup> (80 deer/mi<sup>2</sup>) (Figure 2). This information was used to guide sharpshooting activities and estimate the percentage of deer removed from the area through these subsequent targeted surveillance efforts.

Another key step in preventing further spread of CWD was to ban the recreational feeding of deer. On February 14, 2011 MNDNR issued a special rule that made recreational deer feeding illegal in a 4-county area (Dodge, Goodhue, Olmsted and Wabasha), surrounding the location of the CWD-positive deer (Figure 3). During the aerial surveys (fixed-wing and helicopter), there were a total of 35 recreational feeding sites observed. The ban was aimed at reducing the potential for the disease spread by eliminating artificially-induced deer concentration sites. MNDNR Enforcement staff began education and

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enforcement of the new rule immediately and compliance was extremely high.

CWD samples were obtained through landowner shooting permits (LSP) February 1-28, 2011, agency-sponsored culling conducted by USDA-Wildlife Services, (USDA-WS) February 22-early April 2011, and opportunistic sampling (e.g., vehicle-killed, sick or found dead deer) in this area. Car-killed deer disposition permits were not given to public persons who hit deer and wanted the carcass in DPA 602; local, county, and state enforcement personnel implemented these guidelines year round.

### **Hunter-harvested Surveillance**

Chronic Wasting Disease testing of hunter-harvested deer in DPA 602 was mandatory and included Special Youth, Archery, Firearm, and Muzzleloader hunting seasons. For the archery and muzzleloader seasons, MNDNR staff placed deer head collection boxes at high-volume registration. Boxes were checked daily, tissue samples were extracted twice weekly by trained MNDNR staff, and shipped overnight to a certified testing laboratory. During the regular firearm season, MNDNR personnel staffed 5 registration stations daily to collect samples from harvested deer, with assistance from veterinary and natural resources students on weekends. Within DPA 602, each registration station collected:

- Retropharyngeal lymph nodes (RLN) from all deer  $\geq 1.5$  years of age, and a small amount of muscle tissue was collected and placed in 95% ethanol for future genetic work.
- A front incisor was extracted from all deer  $\geq 2.5$  years old for aging by cementum annuli.
- Both fawn and adult deer were issued carcass tags by authorized MNDNR staff.
- MNDNR shipped RLN samples daily to the receiving laboratory in order to achieve a 3-business day turnaround time for test results
- Adult carcasses were prohibited from being taken out of DPA 602 until they were confirmed CWD-negative

All samples were inventoried, entered into a database, and sent to either the University of Minnesota's Veterinary Diagnostic Laboratory (St. Paul, MN) or Colorado State University (Fort Collins, CO) for enzyme-linked immunosorbent assay (ELISA) testing. Any presumptive positive samples from ELISA testing would be confirmed using immunohistochemistry (IHC) testing at the National Veterinary Services Laboratory in Ames, Iowa.

Hunters were able to check their test results on the MNDNR website using either their MNDNR number or the carcass tag number issued. At the time of sample collection, hunter information was recorded, including the hunter's name, a telephone number, MNDNR number, and location of kill. Maps were provided to assist the hunters in identifying the location (Township, Range, and Section) of the kill. Cooperating hunters were given a cooperator's patch and entered into a raffle to win a .50 caliber muzzleloader donated by the Minnesota Deer Hunter's Association (MDHA). During the firearm season, individual test results were available on-line within 3 business days of submission. Archery and muzzleloader results took slightly longer because of the bi-weekly sampling strategy.

Carcass movement restrictions within DPA 602 prevented local venison processors outside the zone from processing deer until negative CWD test results were obtained; this affected taxidermists as well. To accommodate hunters seeking a taxidermy mount from deer harvested within DPA 602, we provided local area taxidermists with necessary training and supplies to collect the retropharyngeal lymph node sample needed to test for CWD. At times, taxidermists came into the CWD management zone to cape the deer head for hunters, then the cape and antlers (with cleaned skull cap) were able to leave the zone immediately.

## RESULTS AND DISCUSSION

### Winter 2011 Sampling

Following a well-attended public meeting of >350 landowners, hunters, and concerned citizens in the Pine Island area, a winter sampling effort was initiated to gain a better understanding of disease prevalence and distribution in the local deer herd. Landowners within the boundaries of the winter surveillance area were able to obtain landowner shooting permit (LSP) for culling deer on their property during February 2011. More than 300 LSP were issued, with each landowner allowed to take an unlimited number of deer, designate up to 15 additional shooters under their LSP, use of high powered center-fire rifles, and the use of artificial lights was permitted. This area is historically regulated as a shotgun-only zone during the firearm season, so the ability to use rifles during this sampling effort was viewed as a unique opportunity by many landowners. Once deer were harvested, the landowner was required to contact MNDNR staff within 24 hours; samples were then collected in the field at private residences, each carcass was given a unique identification tag, and landowners were directed not to transport carcasses outside the winter surveillance area until they were notified of test results. Meat that was boned out or cut and wrapped either commercially or privately, and quarters or other portions of meat with no part of the spinal column or head attached were allowed to leave the zone immediately.

Late February 2011 through early April 2011, MNDNR contracted with USDA-Wildlife Services to use sharpshooting at bait piles to obtain additional deer samples. Deer were targeted during late evening and overnight hours, and intact carcasses were transported to the central processing facility located within the winter CWD surveillance. MNDNR and USDA-WS disease biologists eviscerated carcasses immediately upon delivery, collected medial retropharyngeal lymph nodes, a central incisor tooth for aging, muscle sample, recorded pregnancy rates, and issued a unique carcass tag to each individual animal. Entrails were deposited in a lined dumpster. All carcasses were held in a refrigerated trailer at 33-35°F until test-negative results were reported (typically within 3 business days). Once negative results were received, we distributed carcasses to the public from a venison disposition list which consisted of more than 400 people. Notified recipients arrived at the facility daily to load, transport and process one or two deer for consumption.

Through this combined winter surveillance effort, a total of 1,180 deer (752 adults, 428 fawns) were sampled within 16 km (10 miles) of the index wild deer case; all deer were negative for the disease (Figure 4). Sampling included deer taken by landowner shooting permits ( $n = 491$ ), agency-sponsored sharpshooting ( $n = 603$ ), vehicle-kills ( $n = 59$ ), and opportunistic sick deer ( $n = 27$ ). Shooting permits were issued to 323 landowners. Of those, 47% of permit-holders harvested  $\geq 1$  deer. The majority of permittees (57%) took 1 or 2 deer and approximately 5% took >10 deer from their properties.

The estimated total cost of the winter sampling effort was \$419,000. The majority (\$229,000) resulted from the USDA-WS sharpshooting contract, staff overtime (\$82,000), and diagnostic testing (\$30,000). The remaining expenditures were related to staff travel, building leases, and equipment leases or rentals.

### Hunter-harvested Surveillance

Following the winter 2011 winter sampling effort, hunter-harvested surveillance became the primary method for obtaining adequate samples for continued CWD monitoring. Fortunately, MNDNR had been conducting hunter-harvested CWD surveillance throughout the state since 2002, with increased focus in southeastern MN due to the infection rate in wild deer in adjacent counties of southern Wisconsin. A recent, intensive surveillance effort in 2009-2010 (which included the discovery of the one positive wild deer in 2010), in which over 3,200 deer were tested for CWD (Figure 5), demonstrated no widespread infection in the region. These data, in combination with historical data from 2002-2008 and the winter 2011 sampling, gave us increased confidence that the disease was not widespread or present at prevalence >0.5%.

The creation of CWD Management Zone, DPA 602, was an important step in efforts to manage

the disease, as it provided an enforceable boundary to restrict the potential flow of prions out of the area. Within DPA 602, MNDNR had the authority to change hunting season lengths, bag limits, offer special disease management tags (unlimited amount at a reduced cost of \$2.50/each) to increase harvest of antlerless deer, and make it mandatory to present the animal for CWD testing upon registration.

From 2011–2013, a total of 4,050 deer ( $n = 1,125, 1,195, \text{ and } 978$  for 2011, 2012, and 2013, respectively) were sampled for CWD within DPA 602 (Figure 6). All deer were negative for the disease. Approximately \$703,000 was spent in efforts to collect and test these hunter-harvested samples over the 3-year period. These results provide strong evidence that Minnesota was on the front end of a CWD outbreak in wild deer. Our inability to detect any additional infected deer in the immediate vicinity of the index case or in surrounding DPA's is encouraging. It is plausible that this disease is recent on the landscape in southeast MN, and that few additional wild deer have been exposed.

## Lessons Learned

The MNDNR had recently responded to an outbreak of bovine tuberculosis (bTB) in cattle and wild deer in northwestern Minnesota (2005-2012) and experiences gained from the management of that disease were very relevant to this CWD outbreak. While bTB and CWD are different diseases with unique transmission routes, initial strategies to manage these outbreaks were similar. Most importantly were efforts to reduce potential transmission pathways by restricting recreational feeding (hunting over bait has been illegal in MN since 1991) and reducing local deer numbers. Some of the tools used in the bTB outbreak, such as agency culling, proved to be highly successful at reducing deer numbers in high-risk areas in both disease outbreaks.

Numerous successes were attained throughout this entire CWD monitoring and response effort, which include:

- Gaining public support for all agency efforts was achieved by maintaining an informed public through publishing/reporting updated information as it became available.
- Having the MNDNR CWD Response Team availability 7 days a week to address concerns and interests expressed by the general public, landowners and hunters.
- The 3-day turnaround time for CWD test results eased hunters' concerns regarding spoilage of their harvested animal.
- The rapid test result time also gave credibility to this project and kept MNDNR staff apprised of additional positive results.
- Accommodating taxidermists and meat processors affected by both the test result reporting time and the carcass movement restrictions, allowed these vendors to perform their work and keep their livelihood and profession active.
- A special page was set up on the MNDNR website for CWD results and updates. Hunters were given a business card with the web address and instructions on how to access their results using either the carcass tag number assigned to their deer or their MNDNR number. Results were posted on the website as soon as they were made available.
- Hunters had the ability to access their results via computer at any time or by phone Monday-Friday during office hours.

While public support for MNDNR's strategies to manage the disease in DPA 602 was evident at the beginning of the outbreak, we did detect rising concerns about continued surveillance efforts once the disease was not widely detected in subsequent years. This "CWD Fatigue" syndrome has been described in other states that have long-standing efforts to manage the disease where public tolerance of control efforts fades over time. Moreover, the high cost of responding to a CWD detection event, which totaled \$1.12 million for this 3-year effort, brings added scrutiny to the need for an aggressive response. The MNDNR believed these efforts were necessary to manage and monitor this CWD outbreak and was thankful for the public and agency support to ensure the health of the local deer herd in southeast MN.



## **Future Surveillance Plans**

Disease surveillance in the CWD Management Zone has been discontinued and DPA 602 will be dissolved by fall 2014. Targeted CWD surveillance of deer exhibiting clinical signs of illness will continue statewide. The MNDNR plans to sample 450 hunter-harvested deer for CWD in DPAs 348 and 349 during the regular firearm season in fall 2014. This effort is in response to a recent detection of CWD in a free-ranging white-tailed deer in Alamakee County, Iowa.

## **ACKNOWLEDGEMENTS**

We would like to thank all the MNDNR Wildlife and Enforcement staff, who volunteered to assist with this disease outbreak surveillance project. Specifically, we'd like to recognize the support received from Rochester Wildlife Staff -- Don Nelson, Mike Tenney, Barb Perry, Nancy Reber, Clint Luedtke, Conservation Officers -- Dan McBroom, Kevin Prodzinski, Phil George and Dean Olson. We also wish to thank the students and faculty from the University of Minnesota, Colleges of Veterinary Medicine and Natural Resources, for assisting in our fall sampling efforts. Special thanks to Julie Hines and Bob Wright for fulfilling our GIS mapping needs and Pete Takash for his efforts in communications and internet-related work. We would also like to thank Beth Martin, Tom Enright, Amanda McGraw, Katie Pilarski, Claire Bagniewski, and Jennifer Dippel for their help through this effort. We appreciate the support of the USDA-Wildlife Services disease biologist Paul Wolf and Danny Storlie and USGS Epidemiologist LeAnn White, along with DNR Pilots Brad Maas and Tom Bucker with flying deer survey work.

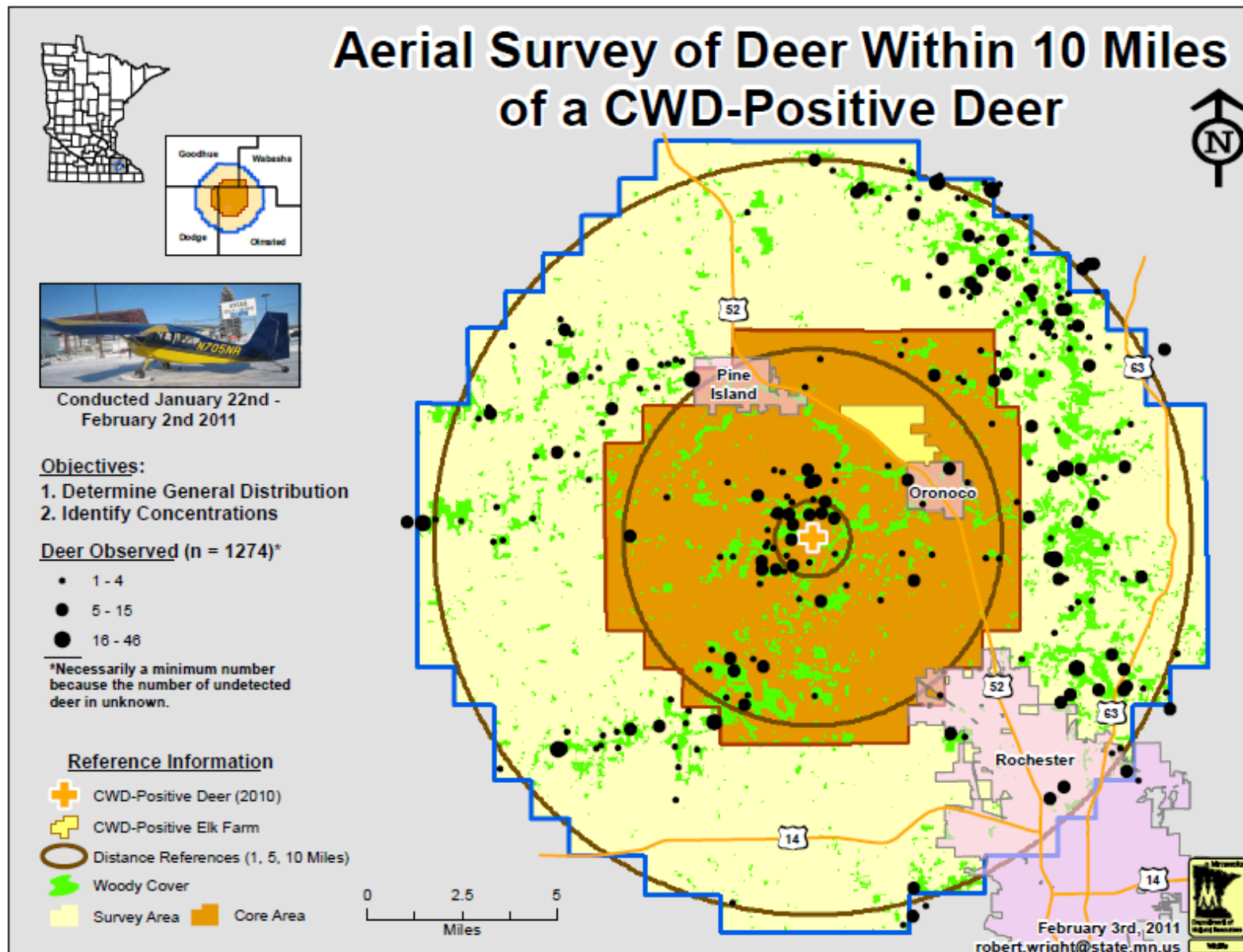


Figure 1. Fixed-wing, aerial survey results for 793-km<sup>2</sup> (306-mi<sup>2</sup>) area surrounding the location of the white-tailed deer that tested positive for chronic wasting disease (CWD), southeastern Minnesota, January–February 2011.

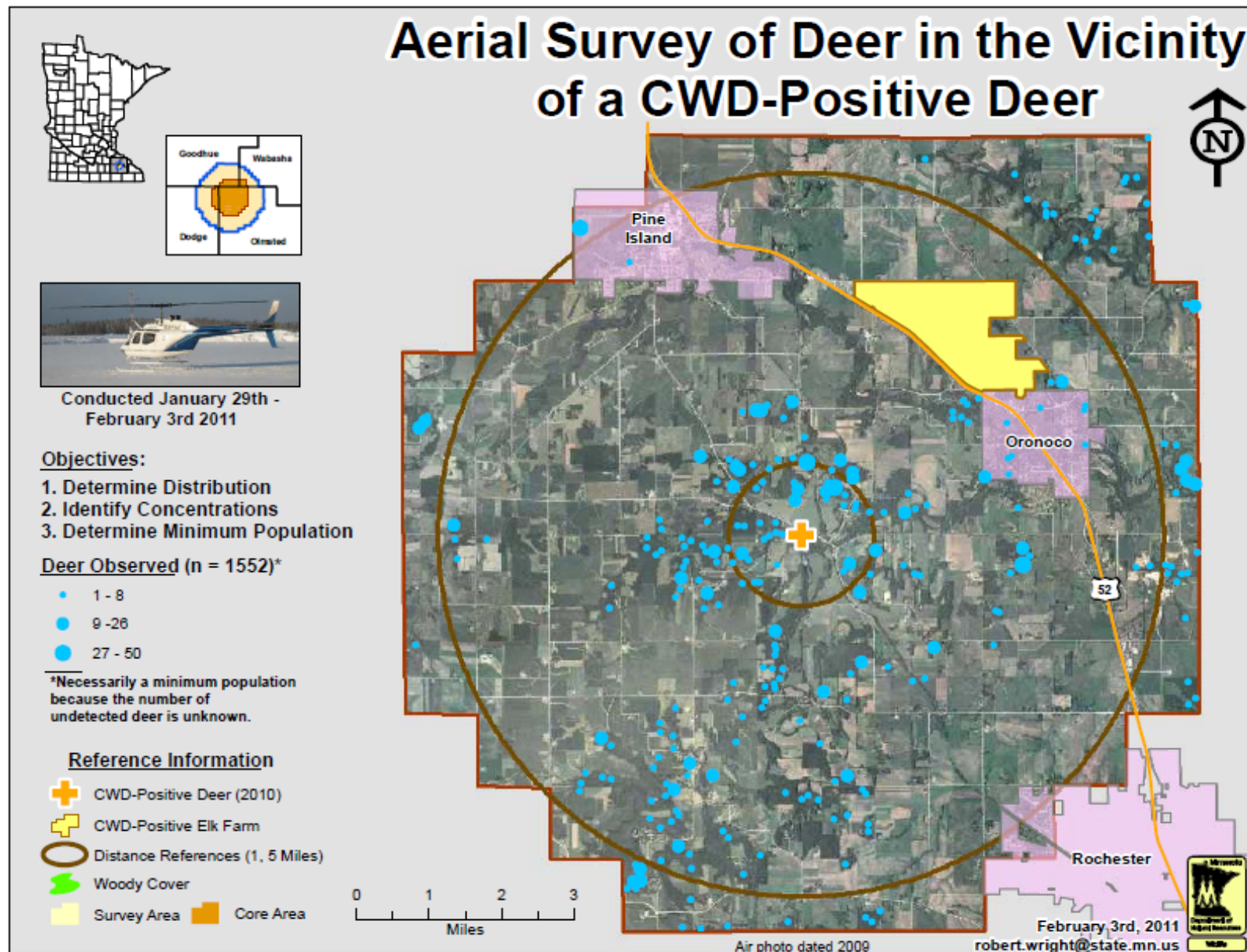


Figure 2. Helicopter white-tailed deer census for the 259-km<sup>2</sup> (100-mi<sup>2</sup>) Core Area within the winter 2011 chronic wasting disease (CWD) surveillance area, southeastern Minnesota, January–February 2011.



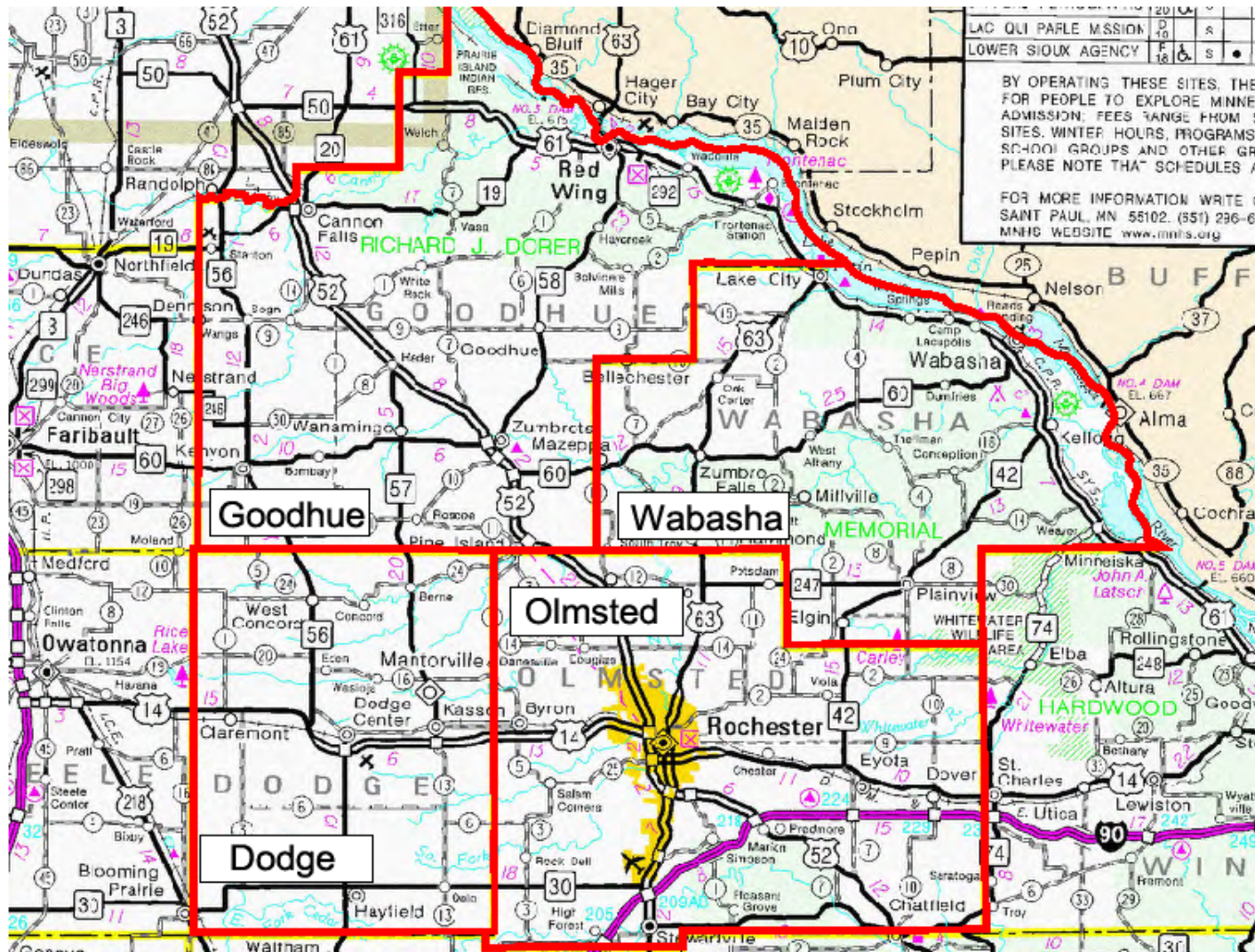


Figure 3. Four-county area in southeastern Minnesota where recreational feeding of wild white-tailed deer was banned in January 2011, following the discovery of chronic wasting disease in Olmsted County.

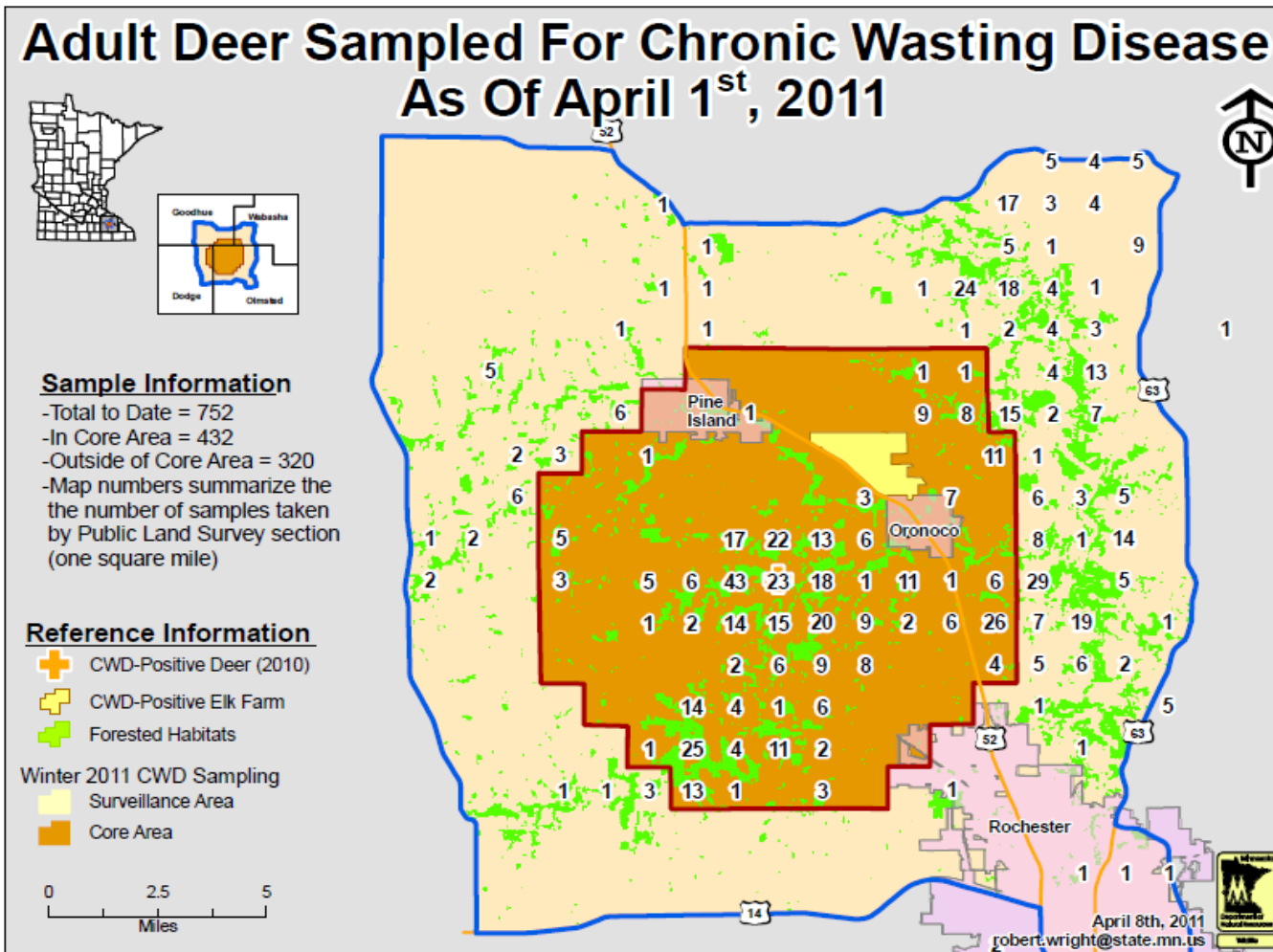


Figure 4. Section totals and distribution of adult (>1 year old) white-tailed deer ( $n = 752$ ) sampled for chronic wasting disease (CWD) during winter 2011, southeastern Minnesota.



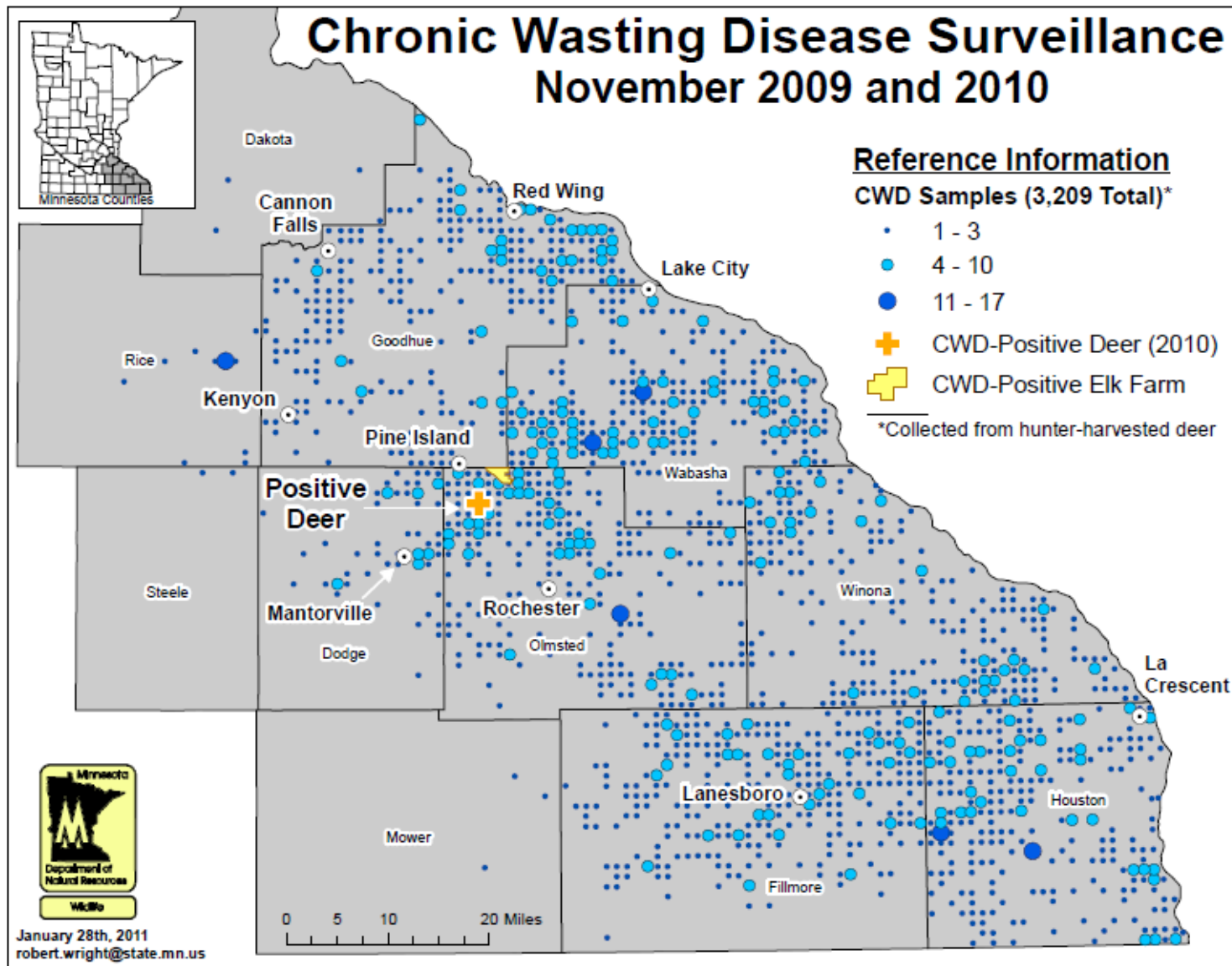


Figure 5. Sampling distribution for all hunter-harvested white-tailed deer ( $n = 3,209$ ) tested for chronic wasting disease (CWD) in southeastern Minnesota, falls 2009 and 2010, in relation to the location of CWD-positive deer.

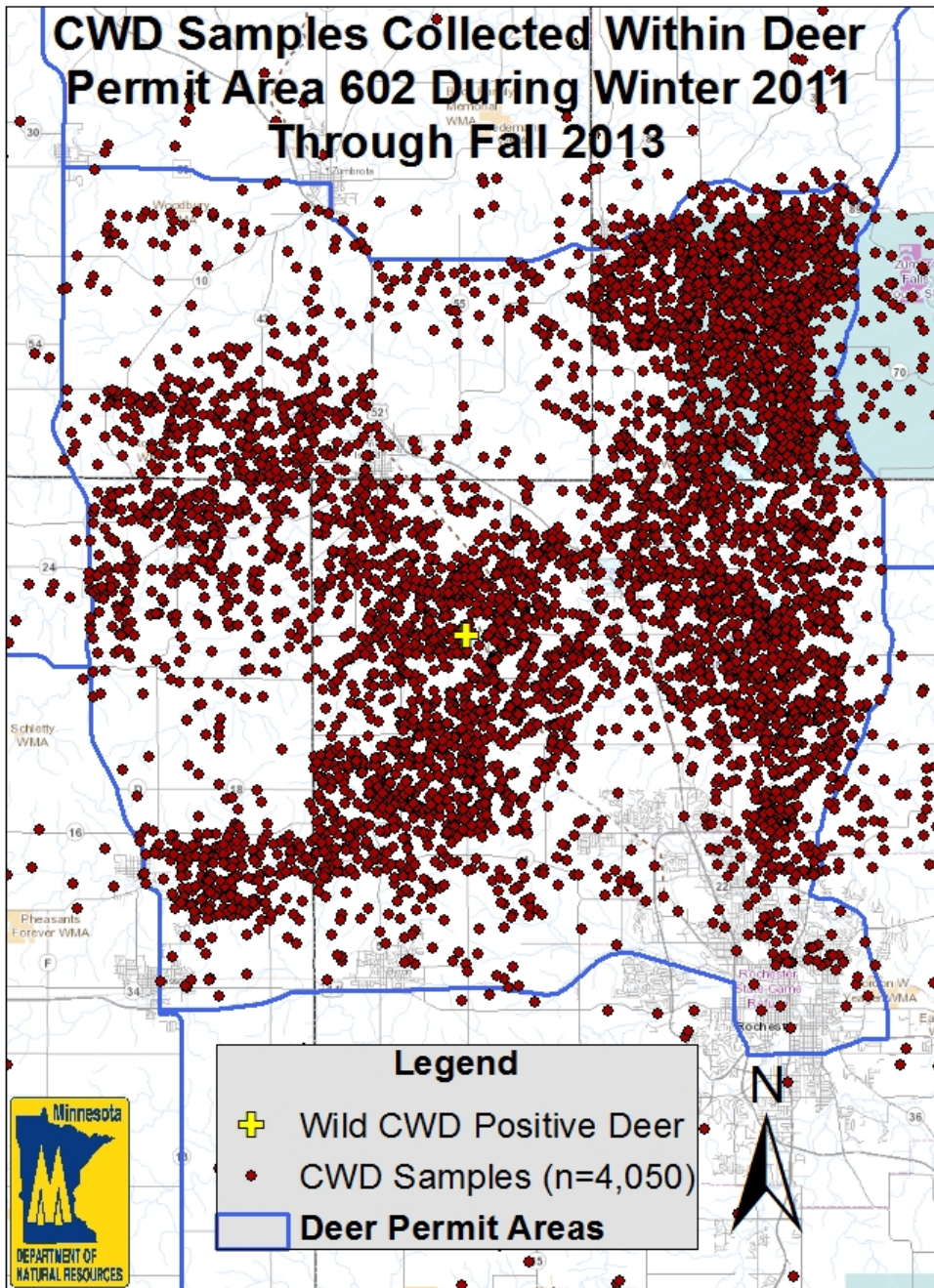


Figure 6. Sampling distribution of deer ( $n=4,050$ ) sampled for chronic wasting disease in deer permit area 602, winter 2011 through fall 2013.

# EPIDEMIOLOGY OF TOXOPLASMOSIS IN WHITE-TAILED DEER (*ODOCOILEUS VIRGINIANUS*): OCCURRENCE, CONGENITAL TRANSMISSION, CORRELATES OF INFECTION, ISOLATION, AND GENETIC CHARACTERIZATION OF *TOXOPLASMA GONDII*<sup>1</sup>

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## ABSTRACT

The prevalence of *Toxoplasma gondii* in white tailed deer (WTD) in the USA is high but little is known of the epidemiology of toxoplasmosis in this host. In the present study, we compared *T. gondii* seroprevalence from 749 WTD collected in 2012 and 2013 from a Metropolitan Park in Ohio and 487 WTD deer shot in Minnesota during 2008, 2009, and 2010. Serum samples were tested for antibodies to *T. gondii* by the modified agglutination test (cut-off titer, 25). Additionally myocardial samples from 123 seropositive WTD from Ohio were digested in pepsin and the digests were bioassayed for the isolation of *T. gondii*. Furthermore, to estimate transplacental rate of transmission, brains from 155 fetuses (included twins) from 148 deer from Minnesota were bioassayed in mice for the isolation of viable *T. gondii*. Seroprevalence of *T. gondii* varied with the year of collection, geography, and the age of deer. Of the Ohio deer sampled in 2012 and 2013 seroprevalences for the two years were similar (73.4% and 75.7%, respectively); remarkably 150 (66.1%) of 227 deer of <1 year of age were seropositive. Of the Minnesota deer, seroprevalence was lowest for the year 2008 (14.8%, 26/175) versus 2009 (27.7%, 59/213), and 2010 (25.2%, 25/99), thought to be related to environmental temperatures. Viable *T. gondii* was isolated in mice from the myocardium of four WTD from Ohio, and brain of one WTD fetus from Minnesota. Tachyzoites from infected mouse tissues were further propagated in cell culture. The DNA isolated from culture-derived tachyzoites of these five *T. gondii* isolates was characterized using 11 PCRRFLP markers (SAG1, 5\_- and 3\_-SAG2, alt.SAG2, SAG3, BTUB, GRA6, c22-8, c29-2, L358, PK1 and Apico). Four genotypes were found, including ToxoDB genotype no. 1 (Type II), no. 2 (Type III), no. 3 (Type II variant) and no. 146. Results indicate fluctuating seroprevalence, probably related to weather and warrant further epidemiological studies.

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<sup>1</sup> Veterinary Parasitology 202, 2014, pg. 270–275

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## CRANIAL AND BRAIN LESIONS OF A FREE-RANGING GRAY WOLF (*CANIS LUPUS*) CONFIRMED IN A HUMAN/WOLF CONFLICT INCIDENT<sup>1</sup>

M. Schwabenlander,<sup>2</sup> M. Carstensen,<sup>3</sup> K. Stepaniuk,<sup>4</sup> and A. G. Armién<sup>2</sup>

### ABSTRACT

We are describing significant brain, cranio-facial, and dental lesions in a wild wolf (*Canis lupus*) involved in the incident that occurred at Lake Winnibigoshish on August 23, 2013, which is the first confirmed wolf attack in Minnesota. DNA forensic analysis confirmed a positive match between the muscle tissue from the wolf and samples obtained from victim. On postmortem examination, the wolf presented asymmetric atrophy and bone remodeling affecting the incisive, maxilla, lacrimal, palatine, frontal, and ethmoid bones. There were dental abnormalities including malocclusion with teeth rotation and deviation, incomplete eruptions, and an odontogenic cyst with an unerupted tooth. Brain changes were bilateral loss and atrophy of extensive cortex regions including olfactory bulb and tract and frontal lobe. We highlight the relevance of a thorough post-mortem examination of wildlife to elucidate disease-based abnormal behavior as the reason for human/animal conflict. In contrast to the majority of cases of this nature, the current case signifies strong, biological, causative evidence for the abnormal behavior of a wild wolf that led to human injury.

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# SPATIAL AND TEMPORAL PATTERNS OF AVIAN PARAMYXOVIRUS-1 EPORNITICS IN DOUBLE-CRESTED CORMORANTS (*PHALACROCORAX AURITUS*) IN THE UNITED STATES<sup>1</sup>

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## ABSTRACT

Newcastle disease is caused by an avian paramyxovirus-1 (APMV-1) virus and is one of the most economically important diseases of poultry worldwide. Outbreaks of APMV-1 in double-crested cormorant (DCCO; *Phalacrocorax auritus*) nesting colonies in the United States and Canada have been sporadically documented in the literature. In this study, we describe the occurrence of APMV-1 associated epornitics in DCCO in the U.S. from first reported occurrence in 1992 through 2012. The frequency of APMV-1 epornitics has increased in the U.S. over the last decade, but the majority of events have occurred in DCCO colonies in the Midwestern U.S. Although morbidity and mortality in co-nesting species has been frequently reported during APMV-1 epornitics, disease caused by APMV-1 was uncommon in species other than DCCO. DCCO populations do not appear to have been significantly affected by this disease. However, since at least 49% the APMV-1 epornitics DCCO in the US have involved APMV-1 strains classified as virulent to poultry (virulent Newcastle Disease virus), its persistence and increased occurrence in DCCO warrants continued surveillance.

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## NECROPSY FINDINGS IN 62 OPPORTUNISTICALLY COLLECTED FREE-RANGING MOOSE (*ALCES ALCES*) FROM MINNESOTA, USA (2003 – 2013)<sup>1</sup>

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### ABSTRACT

The Minnesota moose population has declined dramatically since the 1990s. All 54 carcasses of moose that died of unknown cause or were euthanized by gun shot by tribal or Department of Natural Resources personnel because of perceived signs of illness between 2003 and 2013 and eight carcasses of moose that died due to vehicular accidents between 2009 and 2013 were submitted to the Minnesota Veterinary Diagnostic Laboratory and included in our study. The majority of the animals were underweight or cachectic ( $n = 53$ ; 85%). Neural migration presumably by *Parelaphostrongylus tenuis* was the most common finding related to disease in the examined animals ( $n = 28$ ; 45%). Moderate to marked *Dermacentor albipictus* (“winter tick”) ectoparasitism with widespread alopecia was the cause or a contributing cause of death in 14 (22%) cases in which grossly apparent anemia was associated with exhaustion of hepatic iron stores. Hepatic lesions associated with *Fascioloides magna* were common ( $n = 37$ ; 60%) but were unlikely to be the cause of death. Environmental factors favoring winter tick survival, habitat expansion of white-tailed deer (*Odocoileus virginianus*) and the survival of terrestrial and aquatic snails, serving as intermediate hosts for *P. tenuis* and *F. magna*, may contribute to the seemingly severe parasitic burden in Minnesota’s moose population.

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## INVESTIGATION OF TREMATODES AND FAUCET SNAILS RESPONSIBLE FOR LESSER SCAUP DIE-OFFS

Charlotte Roy

### SUMMARY OF FINDINGS

The spring of 2013 was very late, with lakes being ice-covered when migrating lesser scaup (*Aythya affinis*) arrived, forcing the birds onto rivers for open water. Ice-off on Winnibigoshish occurred after scaup departed, but the edges were opening and available to the birds just before their departure the second week of May. We were able to access Winnibigoshish from resorts on the Mississippi River, but could not access Bowstring or Round lakes while the birds were here. Five lesser scaup were suspected to have trematodiasis at Winnibigoshish in the spring of 2013, but the birds could not be captured for confirmation.

During the fall of 2013, few birds were suspected of trematodiasis at Winnibigoshish, Bowstring, and Round lakes. Five birds were collected from Winnibigoshish and confirmed to have had trematodiasis. The number of sick and dying birds in 2013 was very low this year but was not due to a lack of use by migrating scaup.

We sampled faucet snails at all sites where they were known to occur in interior Minnesota; Lake Winnibigoshish, Upper and Lower Twin lakes, the Shell River, First Crow Wing Lake, the Crow Wing River, Second Crow Wing Lake, Bowstring Lake and several ponds on the White Earth Nation. We also sampled Round Lake for faucet snails, because it is known to be important to migrating lesser scaup and sick birds have been previously reported here.

We are currently analyzing data. A final report will be prepared in September.

### INTRODUCTION

During the autumns of 2007 and 2008, thousands of lesser scaup and hundreds of American coots died on Lake Winnibigoshish in north-central Minnesota. These deaths were attributed to trematodiasis caused by non-native intestinal trematodes (*Cyathocotyle bushiensis*, *Sphaeridiotrema spp.*, and *Leyogonimus polyoon*) and concerned both waterfowl hunters and non-consumptive users.

The trematode species responsible for the die-offs have a complex life cycle that involves two intermediate hosts. The faucet snail (*Bithynia tentaculata*), a non-native species from Europe (Sauer et al. 2007), is the only known first intermediate host of these trematodes in the Midwest and also serves as the second host for *C. bushiensis* and *Sphaeridiotrema spp.* The second host of *L. polyoon* is one of a variety of larval aquatic insects, including damselflies (Zygoptera) and dragonflies (Odonata) (US Geological Survey, National Wildlife Health Center (NWHC), unpubl. data). Adult trematodes develop in waterfowl after they consume infected snails and in American coots (*Fulica americana*) and common moorhens (*Gallinula chloropus*) after consumption of infected insects. Parasite eggs are then defecated by sick birds and later ingested by snails, continuing the cycle. Because of this complex life cycle, the dynamics of faucet snail distribution and transmission of these parasites to lesser scaup and other birds are poorly understood.

The first U.S. detection of the faucet snail was in Lake Michigan in 1871 (Mills et al. 1993). It has since been documented in the mid-Atlantic states, the Great Lakes Region, and

Montana, and undoubtedly will continue to spread (Sauer et al. 2007). In 2002, the faucet snail was detected in the Upper Mississippi River. Since then, trematodiasis has killed an estimated 52,000-65,000 waterbirds, primarily lesser scaup and American coots, but also dabbling ducks such as blue-winged teal (*Anas discors*), Northern shoveler (*Anas clypeata*), mallard (*Anas platyrhynchos*), American black Duck (*Anas rubripes*), and Northern pintail (*Anas acuta*); diving ducks such as ring-necked ducks (*Aythya collaris*) and redheads (*Aythya americana*); and other waterfowl such as ruddy ducks (*Oxyura jamaicensis*), buffleheads (*Bucephala albeola*), and tundra swans (*Cygnus columbianus*, R. Cole, NWHC, pers. comm.).

The faucet snail was detected in Lake Winnibigoshish in the spring of 2008, following the loss of 7,000 Lesser Scaup and a few hundred coots to trematodiasis the previous fall (Lawrence et al. 2008). In 2008, 2,000 more birds died (Lawrence et al. 2009). The severity of the outbreaks seems to have lessened in Lake Winnibigoshish over time. This may be because fewer birds are stopping over on the lake during migration or there may be another explanation related to the disease cycle. In any event, these outbreaks are highly visible and attract the media, which can spur public concern and a desire for action.

In recent years, new areas have been designated as infested with faucet snails in north central Minnesota. The faucet snail was first detected in Upper and Lower Twin lakes and the Shell River in 2009. In 2010, the Crow Wing River was designated as infested with faucet snails, and in 2011, First Crow Wing Lake and Second Crow Wing Lake were added to the list of waters infested with faucet snails. In 2012, several new ponds were designated as infested on the White Earth Nation. These newly designated sites may afford us additional opportunities to learn about this disease cycle.

We examined the factors associated with faucet snail abundance and distribution, parasite prevalence within snails, and the influence of snail densities and site attributes (e.g., water depth, distance from shore, substrate composition) on lesser scaup foraging. For example, depth influences the amount of work that scaup have to do against buoyancy. Shallow depths are thus important to foraging scaup (Jones and Drobney 1986, Mitchell 1992). If such depths are also preferred by faucet snails, then the potential for exposure will be much higher than if snails prefer dissimilar water depths. The profitability of food items will vary as a function of depth, density, and prey type among other things (Lovvorn and Jones 1991, Lovvorn et al. 1991, Beauchamp et al. 1992, de Leeuw and van Eerden 1992, Lovvorn 1994).

## **OBJECTIVES**

- 1- Improve understanding of lesser scaup foraging as it relates to faucet snail and other food source distribution and density, including water depth, distance from shore, and substrate composition
- 2- Examine factors (e.g., temperature, substrate, vegetation, other snail species) that are associated with the distribution and movement of faucet snails
- 3- Examine the factors that influence the prevalence of the parasites in faucet snails (e.g., snail density, temperature, microhabitat, time of year)
- 4- Examine how faucet snail distribution varies during spring, summer, and fall

## METHODS

During 2013, we sampled faucet snails at the same locations sampled in 2012 (Roy 2013). We sampled during spring, summer, and fall at the same points within a lake or river (Table 1a,b). In small lakes (<405 ha), we used transects that traversed the entire length of the lake and across a range of depths. In large lakes, we used index areas with points stratified by depth for sampling. In Lake Winnibigoshish, we had 2 index areas, the West Winni Index Area and the East Winni Index Area, which were 5-6 km along the longest dimension and approximately 2 km in width. In rivers, we sampled points at regular intervals (500 m) along the infested corridor for a maximum length of 10 km. In small ponds, we placed sample points ~100 m apart in such a way as to attempt to maximize the number of sampling locations in each pond (diameter 75-320 m).

We used 2 sampling methods; we used a bottomless sampling cylinder (0.2 m<sup>2</sup>) at 30 and 60 cm depths for comparisons with an ongoing study on the Upper Mississippi River, and we also sampled with a benthic sled to standardize our protocol for all depths. We dragged the sled a distance of 1.2 m at deeper depths to examine how snail distribution varied within a water body. We collected data on microhabitat variables at each point to examine relationships to snail distribution, the snail community, and parasite prevalence. These included substrate (e.g., silt, rock, sand, vegetated, mud), temperature (C°), water depth (cm), and a secchi depth (cm) reading was taken 8 times (4 times on the way down and 4 times on the way up) from the shaded side of the boat and averaged. At each snail collection site, we determined pH, dissolved oxygen (mg/L), conductivity (μS/cm), and salinity (‰) with a Hach Company (Loveland, Colorado) HQd portable meter that was calibrated daily for pH and weekly for conductivity. Flow (mps) was measured at 60% of the total depth (from the surface) with a Global Water Instrumentation (Gold River, California) flow probe when flow was detectable and averaged over a 40 s interval (the USGS “6 tens method”).

Invertebrate samples were stored in the refrigerator until processed. We used a magnifying lens and microscope as needed to identify all invertebrates to Order and noted their presence in each sample. We identified all snails to genus and counted their numbers in each sample. We determined the size of *B. tentaculata* and similarly sized *Amnicola* spp. with calipers, as measured along the central axis from the apex. Parasite prevalence was determined for all samples possessing at least 50 *B. tentaculata* (R. Cole, NWHC, unpubl. data). For samples possessing 10-49 *B. tentaculata*, we collected additional snails while in the field from the same location at the same time to increase the number of samples for which we could do prevalence. These additional snails were not used in the determination of snail abundance at the site. Trematode stages (cercariae or metacercariae), species (*C. bushiensis*, *Sphaeriodotrema* spp., *L. polyoon*), and numbers were also recorded in the lab.

Each season, we collected a water sample at each sample pond, lake, or river and sent it to the Minnesota Department of Agriculture for analysis. Total phosphorus (ppm), nitrite plus nitrate nitrogen (ppm), chlorophyll a (ppb), total alkalinity (ppm), ammonia nitrogen (ppm), and calcium (ppm) were quantified.

We also attempted to identify sites where lesser scaup foraged and collected benthic samples at these locations. These sites were identified through observations of birds from shore or from a boat. We determined the location of rafts of scaup using a compass from 2-3 observation points, which was plotted in ArcMap version 10 (Environmental Systems Research Institute, Inc., Redlands, California) to determine the area occupied by the birds. We then placed a transect through this area and sampled at 100 m intervals. Food densities, water

depths, distance from shore, lake size, and substrate composition at these foraging locations were recorded using the same techniques as snail sampling.

We also collected scaup carcasses during die-offs at study lakes for confirmation of trematodiasis by the NWHC in Madison, Wisconsin. Additionally, Bowstring and Round lakes are known for having large number of scaup, particularly in the fall, and have been the sites of trematodiasis die-offs in the past. We monitored Bowstring and Round lakes for scaup die-offs during the fall only, because we were unable to access the lakes in the spring due to ice-cover. Staff from the Minnesota Department of Natural Resources-Grand Rapids office also made regular visits to Winnibigoshish, Round, and Bowstring lakes throughout the fall season to check for sick birds. Scaup sightings at focal lakes during DNR Fall Waterfowl Migration Surveys were also noted.

## RESULTS

### Faucet snails

We detected faucet snails at both index areas on Lake Winnibigoshish, Upper and Lower Twin lakes, the Shell River, First Crow Wing Lake, Crow Wing River, Bowstring Lake, and the White Earth Ponds (Tables 1a,b).

### Trematodes

Both *C. bushiensis* and *Sphaeridiotrema spp.* were detected on Lake Winnibigoshish, Lower Twin Lake, the Shell River, First Crow Wing Lake, the Crow Wing River, and the White Earth ponds. Samples from Bowstring and Upper Twin lakes did not include sufficient numbers of faucet snails to determine prevalence and intensity of infection this year. *Sphaeridiotrema spp.* has been detected at Upper Twin Lake in previous years of this study. Samples from Second Crow Wing Lake contained *C. bushiensis* only, but few samples contained faucet snails at this location. Prevalence (proportion of snails infected) of *C. bushiensis* was generally higher than that of *Sphaeridiotrema spp.* within a water body. Prevalence of *C. bushiensis* was generally highest at Winnibigoshish, Shell River, and the White Earth ponds. Prevalence of *Sphaeridiotrema spp.* was highest at West Winni Index Area and the Shell River. The intensity (number of parasites in infected snails) of parasite infections was also very high at these locations.

### Scaup

The spring of 2013 was very late, with lakes being ice-covered when migrating lesser scaup (*Aythya affinis*) arrived, forcing the birds onto rivers for open water. Ice-off on Winnibigoshish occurred after scaup departed, but the edges were opening and available to the birds just before their departure the second week of May. Five lesser scaup were suspected to have trematodiasis at Winnibigoshish in the spring of 2013, but the birds could not be captured for confirmation (Table 2). We were unable to monitor Bowstring and Round lakes in the spring of 2013 because the boat accesses were iced-up.

During the fall of 2013, few lesser scaup were suspected of trematodiasis at each of Winnibigoshish, Bowstring, and Round lakes. Five lesser scaup were collected from Winnibigoshish and confirmed to have had trematodiasis by the National Wildlife Health Center. An American coot was collected from Upper Twin Lake and also confirmed to have died of



trematodiasis. The number of sick and dying birds in 2013 was very low this year and was not due to a lack of use by migrating scaup (Table 2).

## DISCUSSION

This report summarizes activities for the third year of field work (spring, summer and fall 2013). Fall 2010, a pilot season, and the 2011 and 2012 field seasons were included in earlier reports. Data entry and analysis are preliminary and still underway. A final report will be prepared in September for the funding agency, with publications to follow in peer-reviewed journals.

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Table 1a. Regular sampling sites for faucet snails in infested northern Minnesota water bodies during spring, summer, and fall 2013. Number of points refers to the number of points sampled each season of sampling.

<b>Location</b>	<b>No. seasons sampled</b>	<b>No. sample points</b>	<b>Faucet detected</b>	<b>snails</b>
East Winnibigoshish Index Area	3	80	Yes	
West Winnibigoshish Index Area	3	80	Yes	
Upper Twin Lake	3	24	Yes	
Lower Twin Lake	3	39	Yes	
First Crow Wing Lake	3	37	Yes	
Second Crow Wing Lake	3	18	Yes	
Crow Wing River	3	18	Yes	
White Earth Ponds	3	24	Yes	
Shell River	3	22	Yes	
<b>Total</b>		<b>1026</b>		

Table 1b. Sampling sites for faucet snails associated with lesser scaup in northern Minnesota water bodies during spring and fall 2013. Number of points refers to the number of points sampled each season of sampling.

<b>Location</b>	<b>Season sampled</b>	<b>No. sample points</b>	<b>Faucet detected</b>	<b>snails</b>
Lake Winnibigoshish	Spring	9	Yes	
Bowstring Lake	Fall	7	Yes	
Round Lake	Fall	7	No	
<b>Total</b>		<b>23</b>		

Table 2. Reports of scaup observed by Minnesota Department of Natural Resources staff on lakes in northern Minnesota during spring and fall 2013. Scaup that failed to escape approach or had drooping heads were considered to be sick; dead birds were typically found along the shoreline.

Location	Date	Total no. of scaup observed	No. of sick or dead scaup observed
<b>SPRING</b>			
Mississippi River near west Winnibigoshish	4/29/13	100	None
Mississippi River near west Winnibigoshish	5/3/13	330	None
Mississippi River near west Winnibigoshish	5/6/13	585	~5 suspect <sup>a</sup>
Dam on Winnibigoshish	5/8/13	168	None <sup>a</sup>
Winnibigoshish	5/16/13	0	0
<b>FALL</b>			
Kenogama	10/8/13	200	Lake not infested, from flight <sup>b</sup>
Twin Lakes	10/16/13	350 coot	1 dead coot <sup>c</sup>
Round	10/18/13	1,450	None
Bowstring	10/18/13	3,000	Sick hen scaup <sup>a</sup>
Bowstring	10/28/13	2,000	7 suspect <sup>d</sup>
Round	10/28/13	2,000-5,000	One sick hen scaup <sup>d</sup>
Winnibigoshish	10/29/13	600-800 coot	Two dead scaup, 5 dead coot <sup>d</sup>
Winnibigoshish	10/29/13	1,000	Determined from flight <sup>b</sup>
Bowstring	11/2/13	8,700	None <sup>a</sup>
Round	11/2/13	4,100	None <sup>a</sup>
Bowstring	11/5/13	3,100	Sick birds noted <sup>d</sup>
Round	11/5/13	3,000-4,000	None <sup>d</sup>
Winnibigoshish	11/7/13	100-200	2 sick, 5 dead, other sick birds noted <sup>c,d</sup>
Bowstring	11/7/13	2,600	None <sup>a</sup>
Winnibigoshish	11/14/13	3	None, but hunters left 3 dead scaup on DNR vehicles <sup>d</sup>
Round	11/15/13	Frozen	None
Bowstring	11/15/13	200	2 sick, unable to fly

<sup>a</sup>Benthic samples collected below scaup.

<sup>b</sup>From flight during DNR Waterfowl Migrations Surveys.

<sup>c</sup>Collected and sent to the National Wildlife Health Center to be tested. Trematodiasis confirmed.

<sup>d</sup>Reported by Grand Rapids Area Wildlife staff.

## SHALLOW LAKES: ASSESSING QUALITY AND PREDICTING FUTURE CHANGE

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### SUMMARY OF PROPOSAL

Lake managers face unprecedented challenges to surface and groundwater quality. Minnesota's 4000 shallow lakes are especially vulnerable to degradation because they often occur in close proximity to intensive agriculture, they are highly interconnected allowing nutrient/chemical inputs and exchange of aquatic species, and because they can quickly transition from clear-water to turbid-water conditions with accompanying poor water quality. Future conservation and management practices for shallow lakes will require special attention as resource managers respond to increasing pressures from agriculture, shoreland development, expanding drainage networks, invasive species, and changing precipitation and temperature patterns. With the recent advent of dedicated funding through the Outdoor Heritage Account, funds are available for shallow lake management, but tools are needed to help prioritize management projects. Prior DNR-and LCCMR-funded studies and other research have identified factors associated with deterioration of water quality, wildlife habitat, and ecological characteristics of Minnesota shallow lakes. Our goal is to use this information to develop a modeling framework that links shallow lake quality to specific drivers of shallow lake condition, including characteristics of individual lakes and features of lake watersheds, along with future changes that may result from increased shoreland development, climate transitions, and other factors.

Previous research has identified important associations between fish, plants, nutrients, and shallow lake quality. Yet, ecological theory suggests that shallow lakes are dynamical systems, and that simple statistical models of associations may not be sufficient for predicting lake condition or for guiding management decisions. Rapid shifts from clear- to turbid-water conditions (or regimes) reflect complex non-linear relationships between predictors and water quality, with multiple interactions among key variables likely to be the norm. Further, thresholds that describe transitions away from clear-water states may vary regionally and conditions that describe transitions from turbid- back to clear-water regimes may differ from those favoring opposite shifts. We propose to develop a data- and model-based framework to guide decisions and management strategies for shallow lakes in Minnesota. This effort will use existing operational resources within the Shallow Lakes Program (DNR Wildlife), build on previous research done by DNR Fish and Wildlife staff and collaborators, and extend our current knowledge regarding the consequences of present and future conditions for shallow lakes in Minnesota. Our broad project goal is development of a modeling context for predicting future shallow lake conditions in Minnesota's changing landscapes which will be useful for managers as they try to identify where to invest existing and newly available resources.

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## INTRODUCTION

Minnesota has approximately 4,000 lakes characterized by mean depth  $\leq 15$  ft and surface area  $> 50$  acres (Nicole Hansel-Welch, personal comm.) and many thousands of smaller waters technically classified as large “prairie wetlands” yet functionally indistinguishable from the larger analogues. These shallow lakes are an international resource, providing critical waterfowl habitat and ecological benefits within Minnesota and the Mississippi Flyway. Currently, only about 48 of these lakes ( $> 50$  acres) are formally designated for wildlife management, yet many others are focus areas for lake rehabilitation and other habitat and conservation practices. Motivated by concerns over shallow lake water quality, seasonal duck abundance and habitat use, and hunter satisfaction, MN DNR recently proposed a collaborative plan to Recover Ducks, Wetlands, and Shallow Lakes ([http://files.dnr.state.mn.us/outdoor/activities/hunting/waterfowl/duck\\_plan\\_highlights.pdf](http://files.dnr.state.mn.us/outdoor/activities/hunting/waterfowl/duck_plan_highlights.pdf)), with a future goal of rehabilitating 1,800 shallow lakes in Minnesota. Lake rehabilitation efforts are logistically difficult, extremely costly, and time consuming. In addition, lake responses are sometimes disappointing and usually short-lived ( $< 5-10$  years, for example see Hobbs et al. 2012). Conservation planning for shallow lakes will require improved tools that managers can apply to many questions such as the following: Which management actions have the highest probabilities of improving lakes in a given region (inducing shifts to a clear state)? Which lakes within a given area have highest rehabilitation potential? What are the specific management actions that optimize the chances for successful rehabilitation given features of an individual lake in a particular watershed? Managers would also benefit from planning tools that can identify management actions that are likely to increase stability or resilience to change in shallow lakes presently in the clear regime, as well which clear lakes that might be candidates for ongoing management efforts. Anecdotal evidence indicates that it is far less costly to preserve clear-regime lakes with high water quality than to respond to deteriorated conditions once they develop, and tools we describe here should help managers develop such forward-looking approaches.

Unfortunately, data on typical baseline conditions and regional variability are not always available, especially for northern Minnesota lakes. Managers are often unsure of the current shallow lake status and whether ecological characteristics of these areas may be limiting use by waterfowl and other wildlife. Despite major advances in shallow lake monitoring, inventory, and research, managers still receive little guidance for improving condition of shallow lakes on a regional scale, or for predicting consequences of future changes such as decreasing CRP, increased drainage, accelerated shoreland development, and other anthropogenic influences in these areas.

Ecological characteristics of shallow lakes, along with their suitability for ducks and other wildlife, result from influences of climate, hydrology, and in-lake biological processes operating simultaneously at multiple scales. Lakes may respond to factors operating at considerable distance from affected basins, and lake responses to these influences probably vary regionally and throughout the state. Ecologists have shown that prairie wetlands (and “shallow lakes”) are strongly influenced by gradients of hydrology (or hydrogeomorphic setting) and climate (especially precipitation) (Euliss et al. 2004). Yet within these boundaries biological interactions in a given lake, especially those resulting from planktivorous and benthivorous fish communities, also exert major structuring influences on lake communities and characteristics of shallow lakes (Hanson et al. 2005). Previous research in Minnesota and elsewhere indicates that water clarity and quality in shallow lakes often differs dramatically in response to fish presence, density, and community structure in these sites. Fish - especially planktivorous and benthivorous species such as fathead minnows and black bullheads - influence

invertebrate community structure, nutrient dynamics, water transparency, and other lake features in Minnesota's Prairie Pothole Region (PPR) and elsewhere (Bendell and McNicol 1987; Hanson and Riggs 1995, Zimmer et al. 2000, 2001 and many others). Recent work shows that fish presence in shallow lakes has greater influence on major groups of aquatic invertebrates than does abundance or taxonomic composition of fish communities (Hanson et al. 2012). Herwig et al. (2010) recently showed that surface connectivity among lakes, rivers, and other waters is a major determinant of fish presence in shallow Minnesota lakes. Along with influences of fish on aquatic communities, this illustrates potential consequences of increased ditching, flooding, and factors influencing surface water connections among shallow lakes, and points toward importance of managing both site- and landscape-scale factors for maintaining and improving quality of shallow lakes.

Shallow lakes are distinct from their deeper counterparts because limited average depth allows greater light penetration to the substrate over most of these basins. With relatively clear water, these lakes often "fill" with lush stands of submerged vascular macrophytes (SAV), contributing to further reductions in turbidity, lower water-column nutrient levels, lower phytoplankton biomass, and favorable conditions for waterfowl and other wildlife (Moss et al. 1996, Scheffer 2004, Moss et al. 2013). Healthy SAV communities may be the most ubiquitous goal of shallow lake management worldwide (Moss et al. 1996, Scheffer 2004), pointing to the central role of these plant communities in maintaining high water quality and wildlife suitability in these sites.

Ecologists have proposed conceptual models to explain dynamics in which shallow lakes alternate between two alternative conditions, either clear-water (macrophyte- dominated) or turbid-water (phytoplankton-dominated) regimes (Scheffer and Carpenter 2003, Scheffer 2004). These shifts are known for shallow lakes worldwide and have been specifically shown to occur in Minnesota lakes (Hanson and Butler 1994; Zimmer et al. 2001; Zimmer et al. 2009). Here and elsewhere, shallow lake management activities usually strive to achieve and maintain clear-water regimes, and stable clear-water conditions with abundant SAV are usually the goal of lake rehabilitation projects.

A major goal of our previous shallow lake studies was to develop conceptual and empirical models linking watershed and site-level features to ecological characteristics of shallow lakes. An overarching finding of the prior work was that regional gradients were major sources of variance in characteristics of shallow Minnesota lakes. Regional differences in nutrient levels are most obvious, yet extent of surface-water connectivity, fish community composition, dissolved organic matter and other processes may also vary consistently among ECS regions in Minnesota and may affect lake-regime responses. For example, we know that combinations of increased benthivorous fish mass and/or decreased abundance of submerged macrophytes will often induce regime shifts in shallow lakes, and these changes probably portend shifts to turbid-water states. However, we speculate that increased fish mass is much less likely to induce turbid-states in north-central Minnesota lakes, and turbid states may not even be possible in northern lakes where low ambient nutrient levels prevail. Additional data synthesis is needed to document extent and patterns of regional variation in occurrence and frequency of turbid regimes in shallow lakes, and to assess how regime dynamics respond to key influences such as surface connectivity, fish community characteristics, stability of phytoplankton- and macrophyte-dominated states, and proportion of lakes in clear- vs. turbid-water states.

Models for quantifying transient, nonlinear transitions in response to major drivers have been developed for grasslands and other ecosystems, but rarely for shallow lakes, despite the fact that these approaches seem to hold great promise as forecasting

tools for managers who need frameworks for relating present and future changes to quality of shallow lakes. We believe understanding of lake-regime dynamics, data availability, and analytical methods has reached a point where the modeling framework we propose is possible and it would improve understanding of factors associated with shallow quality. Such a model framework would provide a useful conceptual and practical tool for lake managers in Minnesota and elsewhere.

## GOALS AND WORKING HYPOTHESES

Our overall project goal is two-fold; the first is hypothesis oriented, but the second is mostly predictive. Based on results of previous studies, we have already constructed environmental variable models testing various hypotheses about factors that influence shallow lake communities and characteristics. In a sense, the proposed effort extends this hypothesis testing, but also refines earlier efforts to solidify the basis for the second modeling framework described below. Broadly, we have two objectives. First, we propose to test region-specific hypotheses using models relating shallow lake regime dynamics to fundamental drivers, these being environmental variables responsible for variation in lake characteristics in Minnesota (and elsewhere depending on data availability). For example, we hypothesize that seasonal temperature patterns, increased sedimentation and nutrient levels, fish abundance and community type, loss of grass cover in watersheds, and lake morphometric features all influence probability of lakes being in clear-water, turbid-water, or transitional regimes in a given landscape setting. Lake responses to combinations of these factors are non-linear, synergistic, and will vary regionally based on ambient nutrient levels and other factors. Models must incorporate these multiple factors to identify isoclines delineating combinations of variables and their associated values where lake-regime characteristics, resilience, and transitions are most likely to occur. Other factors such as inter-annual variability in precipitation and temperature will have strong influences on shallow lakes, and this inter-annual variability may need to be incorporated in predictive models.

Our second project goal is to develop a 2-state (clear/turbid) Markov model (hereafter transition model), where the transition probabilities between states are informed by results of environmental variable models mentioned above, but also using published relationships from shallow lakes in North America and worldwide. For example, probabilities of transitioning from clear to turbid may depend on fish colonization rates (or lake connectivity), increases in nutrient loads, or changes in other key drivers. Initially, we expect to model transitions between states using rule- or frame-based formats described in various applications by Starfield et al. (1989), Starfield (1990), and Starfield and Chapin (1996). An example of a simple rule could be:

$$\begin{aligned} P(\text{transition from clear to turbid}) &= 0.1 \text{ if total phosphorus (TP) is } < x_1 \\ &= 0.4 \text{ if } x_1 \leq \text{TP} < x_2 \\ &= 0.9 \text{ if } \text{TP} \geq x_2. \end{aligned}$$

More complex rules may be defined using multiple state variables (e.g., TP and fish abundance). These additional state variables may themselves be influenced by other state variables (e.g., fish abundance may be influenced by lake connectivity) as well as additional rules. An advantage of rule-based models is that they require only reliable qualitative (not quantitative) understanding of variables influencing complex relationships (in our case, factors influencing whether shallow lakes are in turbid- or clear-water regimes). Results from our earlier studies of Minnesota shallow lakes, along with data compiled by DNR Shallow Lakes Program staff, should provide a solid foundation to help

guide development of rules governing transitions between states and also how watershed variables may influence these transition probabilities.

Our proposed framework can be viewed as a derivative of the dynamical system models suggested by Scheffer and Carpenter (2003) and Scheffer et al. (2001) which are now well known in aquatic ecology and do an excellent job of describing (and making sense of) catastrophic responses of shallow lakes to high densities of undesirable fish, increased nutrient loading, and perhaps other environmental influences. Such shifts are known from shallow lakes across North America and Europe, but occur in coral reefs, other ocean populations, and terrestrial ecosystems including grasslands and deserts (Scheffer et al. 2001). Dynamical systems models are useful not only as theoretical constructs, but have very practical applications helping managers understanding historical changes in Lake Christina and other shallow waters in Minnesota (Hanson and Butler 1994, Zimmer et al. 2009, Hobbs et al. 2012).

In the dynamical systems models developed for shallow lakes (Scheffer and Carpenter 2003, and others), transitions between states are not explicitly defined; rather they are an emergent property of these models. These transitions and also the equilibrium state of the system depend on how the model is formulated, including the number of key state variables (fish, nutrient levels, SAV, etc), the types of interactions among these state variables, and the specific parameters that govern the strength of these interactions. By contrast, we propose to explicitly model transitions between states using empirically derived rules. The main advantages of our proposed approach is that: a) the model will be considerably easier to parameterize and apply; 2) it will be easier for managers to understand; and 3) it will provide a simple framework for linking individual shallow lakes and within-lake variables to characteristics of adjacent watershed uplands or regional landscapes; these links are currently lacking in the dynamical systems models developed for shallow lakes.

## **Model Building Process**

We intend to use rapid prototyping (Starfield et al. 1994, Nicholson et al. 2002) to quickly explore a variety of simple models from start to finish (where, “finish” includes a summary of conclusions as well as a careful examination of assumptions and a sensitivity analysis to evaluate the effect of parameter uncertainty); a rule-based approach seems ideally suited to this goal. This important first step should help to further define key research objectives that focus efforts on the most pressing questions and problems facing managers, while also helping to elucidate the relative importance of different model components. Additional complexity can then be introduced as needed to capture missing features or to better fit the qualitative dynamics of the system. We expect significant regional variation in system dynamics, and this regional variation (and data) can provide a rich medium for adaptive model building. For example, models may be initially ‘tuned’ using data from one region and then tested by comparing model predictions to empirical data from another region. Lastly, we expect that the iterative process of model-building and testing may serve to motivate the need additional data collection efforts; if so, we anticipate that the participating graduate student, staff from the DNR Shallow Lakes Program, and our research team would work together to address these data needs.

## **Objectives and Hypotheses**

Presently, dynamical shallow lakes models (Scheffer and Carpenter 2003 and others) provide only generalized views of shallow lake behavior and, in a sense, stop



short of linking shallow lakes to characteristics of adjacent watershed uplands or regional landscapes. For example, it is not yet possible to estimate likelihood of lake-regime shifts in response to changing extent of native upland cover (grass or CRP) in lake watersheds, to increased ditching in lake watersheds, or to increased mean annual precipitation and temperature. Such refinements should be possible with sufficient data linking lake-regime to watershed and landscape characteristics, lake-level variables, and regional variation. For example (and as a step in this direction), Zimmer et al. (2009) linked inter-annual changes in fish mass and ambient nutrients levels (total phosphorus in lake waters) to regime shifts in Minnesota lakes, suggesting that these dynamics may be predicted in more detail using factors not entirely accounted for in original catastrophic models. We believe a major information gap exists between Scheffer's dynamical systems models of lake-regime behavior and various environmental "predictors", or factors that may affect these regime dynamics and are known for shallow lakes in Minnesota landscapes. Our approach points to a qualitative framework that relates site- (lake) and watershed-scale variables to regime status of shallow lakes (clear- and turbid-water states) in Minnesota. Empirical data from research in Minnesota lakes, along with other appropriate information, will provide an underlying basis for qualitative rules required for the model building process. Conceptually our project includes two general aspects:

- Part I: Develop statistical models linking factors associated with turbid, clear, and shifting lakes, to environmental variables, and (if possible) evaluate whether these key drivers of lake regimes vary across the state (Figure 1A,B). This requires a series of refinements to earlier analyses (and publications) relating lake- and watershed-scale variables to shallow lake communities and characteristics. Examples of previous efforts include reports linking surface water connectivity to fish presence in shallow lakes (Herwig et al. 2010), importance of fish abundance and guild composition on phytoplankton abundance (Friedrichs et al. 2010), comparing influences of lake- and watershed-scale variables on aquatic invertebrate community abundance patterns among lakes (Hanson et al. 2012), and predicting phytoplankton abundance in shallow lakes in response to watershed cover types, background nutrients, and in-lake fish abundance (Gorman et al. 2014). We propose further modeling with additional data (developed since these publications) to clarify relationships, better account for variability in lakes among ECS regions (Figures 1A, 2), and to fill in data gaps where they exist. As indicated above, this step will emphasize modeling responses of lake characteristics to environmental factors. Relationships are almost certain to be non-linear, synergistic, and variable according to regional gradients in ambient nutrient levels and other factors. Models must identify relationship isoclines (such as indicated in Figure 3), and delineate combinations of variables and their associated values having greatest influence on lake-regime characteristics and transitions.
- Part II: Formulate rules and develop a frame-based transition model for predicting status and changes in water-clarity regimes in shallow lakes in response to present lake- and watershed-scale conditions; model implications to shallow lake regimes resulting from future changes in Minnesota (e.g. climate change, increased agriculture in watersheds, more extensive ditching) (Figure 1C). We propose an approach that melds catastrophic-regime models of Scheffer et al. (2001, 2003) and Scheffer (2004) with rule- and frame-based modeling efforts such as those developed by Starfield and Chapin (1996). Preliminary

discussions with Dr. Tony Starfield confirmed that frame-based models (Starfield and Chapin 1996, Hahn et al. 1999) seem especially appropriate for predicting regime shifts in shallow lakes in response to changes in key environmental variables. Subsequent analyses may suggest better modeling approaches, but we anticipate using rule- or frame-based models as a starting point. Final models will incorporate environmental variable relationships (described in approach 1 above). This requires complex modeling approaches so as to incorporate influence isoclines, account for likely hysteresis in relationships between lake regime and environmental variables, incorporate influences of regional gradients (Figure 2), and include interactions among important variables. An example of theoretical frame-based lake model is shown in Figure 1C.

## **Logistics and Approach**

Our effort consists primarily of integrating existing data resources, using these to model (and refine) relationships between environmental variables and regime dynamics in shallow lakes, then incorporating these relationships into frame-based models to predict lake regime status and transition. Initial modeling will be based primarily on data resources derived from shallow lake studies conducted in Minnesota during 2005-2011 by research staff from DNR Fish and Wildlife Research Units, Dr. Kyle Zimmer (University of St. Thomas, St. Paul), and other collaborators and funded by DNR, the Natural Resources Environmental Trust fund (through LCCMR), Ducks Unlimited, National Science Foundation (through a grant to Dr. Zimmer), and other sources. In practice, this will require a combination of several approaches including evaluation of existing data from Minnesota studies and elsewhere, possible data gathering to meet additional information needs (especially if this is needed to clarify regional patterns), and actual model development. Data summary, evaluation, and possible data gathering will be conducted by primarily by Hanson, Herwig, Zimmer, and Hansel-Welch, along with staff from DNR Shallow Lakes Program, DNR Wildlife staff, and perhaps managers from other resource agencies in Minnesota. Model development will be primarily the responsibility of Fieberg, Starfield, and Johnson, along with a graduate student enrolled in Fish, Wildlife, and Conservation Biology at UM (with Fieberg serving as major advisor).

First, along with collaboration of DNR Shallow Lakes Program staff, we will review and synthesize data resources available to assess extent of shallow lake regime status by major ECS regions (Figure 1A). Previous LCCMR-funded research helped to clarify relationships between environmental variables and shallow lake quality, but also showed a need for a more extensive geographic sampling across the state. We anticipate utilizing Shallow Lake Program staff and other collaborators to fill in existing data needs, especially in north eastern and north western areas of Minnesota where relatively few shallow lake surveys have been completed. Expanded regional data gathering may be necessitated by extreme variability among shallow lake conditions across ECS regions (Figure 2) and because this variability is likely to influence complex responses of lake regimes to environmental gradients (Figure 3).

Second, we propose to expand and refine efforts to connect shallow lake characteristics known to reflect lake regime status (turbidity, phytoplankton biomass, extent of submergent macrophytes, and other variables) to key environmental variables, those being factors that are especially likely to influence lake regime status. This will include variables measured at lake- and watershed (landscape)- scales. We expect to expand environmental variable modeling at a large geographic (ECS Section) scale. This activity will improve our understanding of the factors responsible for lake quality,

improve understanding of regional patterns, and aid in conservation planning. Ultimately, data availability may determine extent to which regional analyses are plausible and broader geographic modeling may be required.

Third, collaborating biometricians and modelers (Fieberg, Starfield, Johnson) will work toward development of transition models useful for forecasting regime status and future changes in Minnesota shallow lakes in response to the suite of factors identified as key drivers of lake regimes (or identified as important environmental variables in our second objective, above). We anticipate that likely drivers of regime status and changes in shallow lakes will be loss of grasslands and riparian areas (such as CRP) in lake watersheds, and changes in temperature, precipitation patterns and hydrologic conditions (climate fluctuation), and other anthropogenic factors such as shoreland development, ongoing ditching among surface waters, further loss of temporary and seasonal wetlands and other factors.

## **MANAGEMENT IMPLICATIONS**

We anticipate that our products (transition models) will be useful on two levels. First, proposed “future lake-condition models” should be extremely useful for DNR Area Wildlife Managers, Shallow Lake Program staff, and other lake managers and conservation planners responsible for making informed policy decisions that impact water quality at regional scales in Minnesota. Our expectation is that this broad transition model framework will be especially useful as a conservation tool for those who need to forecast influences of complex changes in Minnesota’s landscape including transitions in upland cover types (such as loss of CRP), extent of ditching (or surface-water connectivity), transitions in precipitation and temperature (climate change), shoreland development, or other likely trends. We know from shallow lake survey data that over half of the shallow lakes surveyed in the prairie are poor condition. The transition models will help us forecast how many more lakes may be in jeopardy of decline with potential changes in landscapes or climate and serve as planning tools to potentially try to mitigate for some of these changes.

Second, the modeling framework described above will also help clarify expected lake-specific outcomes, integrating the best available information for assessing the relative effectiveness of alternative management actions as a function of within-lake conditions, landscape setting, connectivity, current conditions, and expected changes to the landscape/climate. In a sense, this means a manager should be able to refine expectations for individual lakes based on results of our transition models. If possible, we will also develop a lake manager’s model such as those developed previously by Starfield within Excel (unpublished data) that allows predictions of likely outcomes for a specific lake, given landscape setting, connectivity, current conditions, and expected changes to the landscape/climate.

## **PLANNED OUTREACH**

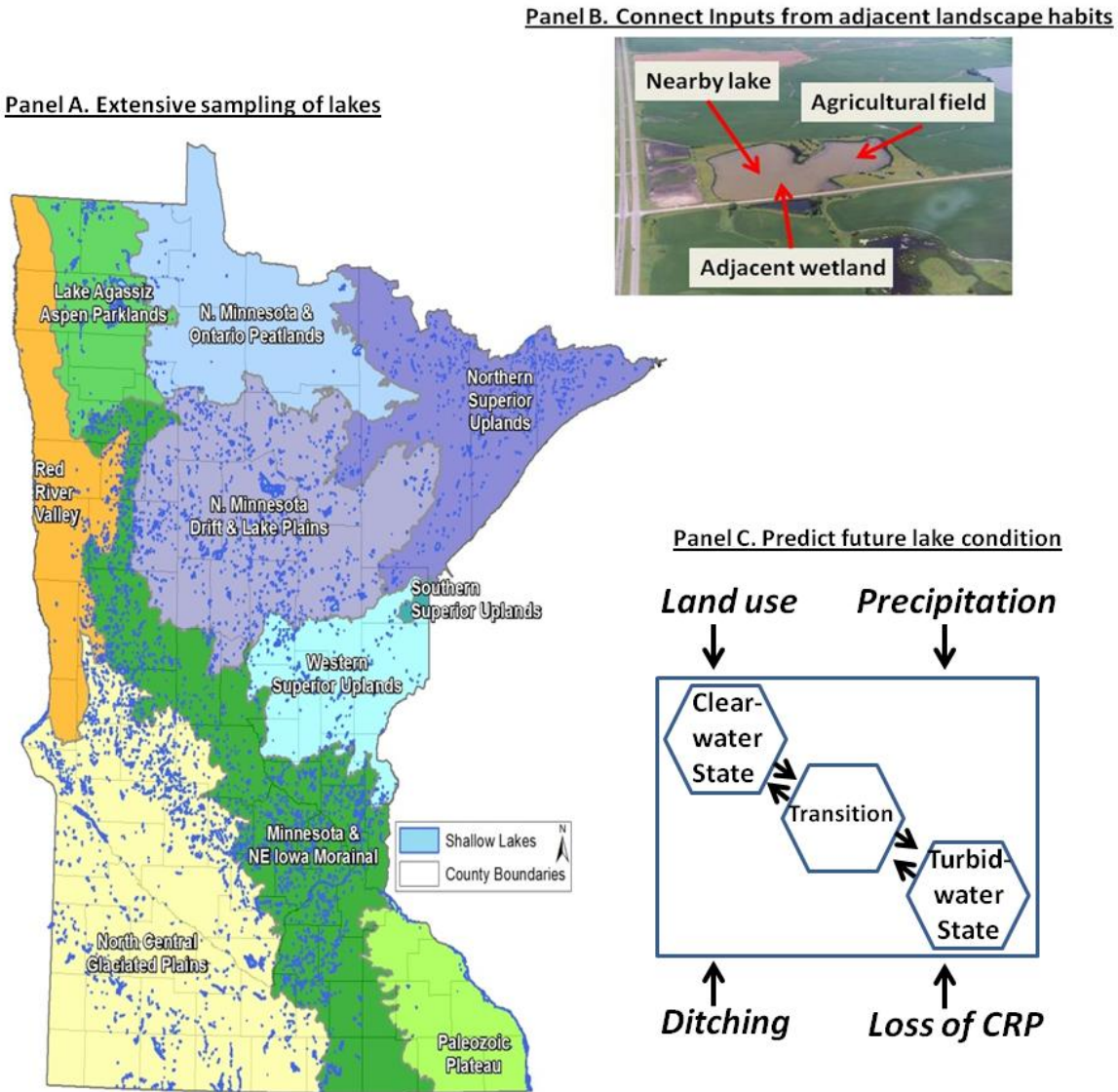
Models will help policy makers and lake managers identify primary causes of poor shallow lake water quality and health, more clearly evaluate future threats to shallow lakes, and determine where management actions have most potential to mitigate or minimize impacts and favor high water quality in these sites. The proposed frame- (or rule-) based modeling, while linked to empirical data, is qualitative and will be far easier to understand than more quantitative approaches. We expect our modeling framework will lead to original insights from new data analyses, but will also lend itself to practical applications by DNR lake managers and others. Frame-based models we describe should lead to observable, discrete lake outcomes (turbid- or clear-water lakes) based on sets of rules that will be easily applied to the specific conditions and

anticipated changes. If appropriate, and to encourage development of these applications, we plan to meet with DNR managers and others to distribute and discuss project models and results.

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**Figure 1.** Overall study strategy. **Panel A** shows extent of Ecological Sections and distribution of shallow lakes in Minnesota. **Panel B** indicates connections between landscapes inputs and imbedded lakes; these connections (arrows) will be modeled in **part I**. **Panel C** illustrates frame-based modeling approach (**Part II**) we plan to use to predict predominant future lake conditions. Predictions will be based on lake responses to potential landscape modifications and other changes within the Ecological Sections.

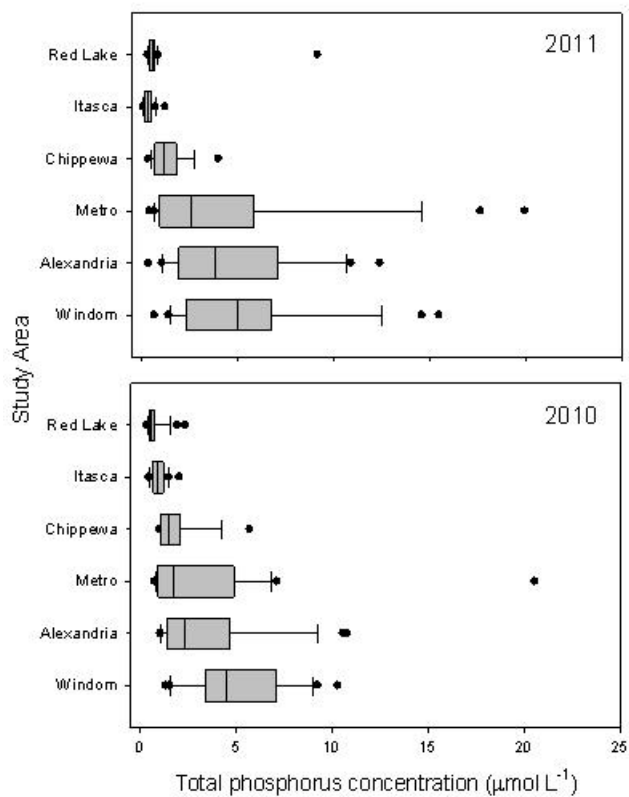


Figure 2. Box plots showing mean abundance of total phosphorus (TP) for 127 shallow lakes sampled within 6 study regions during 2010 and 2011. Vertical lines within boxes depict median TP values for each study region; extent of shaded boxes depict 25<sup>th</sup> and 75<sup>th</sup> percentiles. Whiskers show 10th and 90th percentiles, with dots indicating more extreme values.

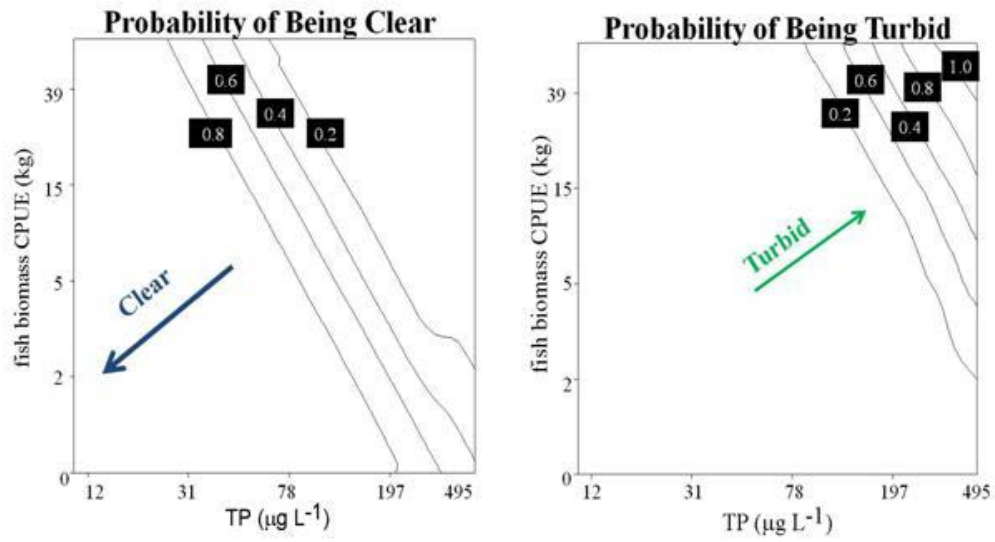


Figure 3. Isobars in panels indicate probability of lakes exhibiting characteristics of clear (left) or turbid regimes. Probabilities of turbid regime show positive relationship to TP concentrations and are based on data from 127 Extensive Lakes



## **REHABILITATING SHALLOW MINNESOTA LAKES: EVIDENCE FROM SHORT-TERM RESPONSES**

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### **SUMMARY OF FINDINGS**

Shallow lake management is complex and rehabilitation efforts often produce unexpected-even disappointing-results. Relatively few strategies are available to lake managers and general strategies have changed little during the past 50 years. Most common approaches are drawdown, removal of undesirable fishes, or combinations of both. Both the application of these efforts and the interpretation of lake responses are complicated by multiple factors. For example, inducing drawdown can be confounded by high precipitation, extensive surface water connectivity, and expectations of recreational lake users. Fish removal is logistically difficult, often requires in-lake use of chemical toxicants, and fish typically re-colonize lakes within a few years after treatment. To maximize project results, managers often apply multiple treatments, but this makes it nearly impossible to identify causal relationships that would inform future projects. Restoration of grasslands or other native vegetation within lake watersheds also favors improved water quality in shallow lakes, but these projects are extremely costly and benefits are slow to develop. We reviewed results of 12 shallow lake rehabilitations in Minnesota and compared communities of these lakes to those of regional shallow lakes in clear- and turbid-regimes. Results indicated that rehabilitated lakes showed a pattern of increased similarity to clear-regime lakes in terms of phytoplankton, nutrients, and submerged aquatic plants. In general, lakes treated by drawdown and fish removal appeared to show stronger rehabilitation responses than did sites where only fish removals were applied.

### **INTRODUCTION**

Shallow lake management is especially challenging because these sites are often highly altered and frequently occur in close proximity to intensive agriculture, roadways, and other unnatural features. Combined effects of increased nutrients, sedimentation, and agricultural chemicals along with altered hydrology may directly limit ecological integrity and wildlife suitability of these lakes. Watershed- and local-scale factors, along with mild winters, favor high densities of planktivorous and benthivorous fishes, and these populations contribute further to water quality and habitat declines. For lake managers, reducing nutrient inputs is often difficult or impossible. In the past 50 years, controlling dense, undesirable fish communities has been a core strategy of shallow lake managers in Europe and the United States (Lammens 1999). Research in recent decades shows potential improvements resulting from controlling undesirable fish communities (including species like bullheads, fathead minnows, and others) rather than

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just eradicating carp (Shapiro and Wright 1984, Hanson and Butler 1994, Moss et al. 1996, Zimmer et al. 2001, Scheffer 2004, Potthof et al. 2008, and many others).

Research in North America and Europe has shown that water-level fluctuations (WLF) influence virtually all aquatic vascular plants in freshwater wetlands (Squires and van der Valk 1992, Wilcox and Meeker 1992, van der Valk et al. 1994, Murkin et al. 2000, Euliss et al. 2004, van der Valk 2005, Wantzen et al. 2008, and many others). It is not surprising that WLF also affect submerged macrophytes (SAV), nutrient cycling, and other properties of shallow lakes, and that natural WLFs are essential for maintaining lake processes and communities (Coops et al. 2003, Chow-Fraser 2005, Leira and Cantonati 2008). Sustained high-water levels are almost certain to favor loss of SAV and may induce transitions to turbid regimes, especially in lakes with high nutrient levels and benthivorous or planktivorous fish (Blindow et al. 1992, Coops et al. 2003, Scheffer and Jeppesen 2007, Hobbs et al. 2012). Alternatively, intermittent periods of lower water levels increase light availability at sediments and favor increased abundance and diversity of submerged aquatic plants, ultimately favoring transitions to clear-water regimes.

In Minnesota, drawdown and fish removal are often used simultaneously (or sequentially) to improve water clarity, light available for SAV, and to enhance zooplankton and other invertebrates, usually for the benefit of waterfowl and other wildlife species (Hansel-Welch and Kudelka 2010). Efforts combining drawdown with fish removal are intended to maximize favorable responses by eliminating undesirable fish populations, favoring re-establishment of SAV, and stimulating germination of emergent macrophytes in lake margins. Rotenone is often applied during late fall or winter periods after lake volumes are reduced, and target fish populations may be concentrated and stressed by seasonal hypoxia. In nearly all cases, shallow lake rehabilitation aims to improve lake water clarity and ultimately induce shifts to clear-regime conditions (Moss et al. 1996, Scheffer 2004, Zimmer et al. 2009). Combination approaches certainly improve likelihood of favorable outcomes, but factors responsible for lake improvements are extremely difficult to identify. In addition, lake management may induce unexpected disturbances and surprising responses in shallow lake communities. For example, equivocal – even contrasting - responses may result from sediment desiccation, physical disturbance by wind and ice, re-colonization and variable survival of fish species following rotenone treatment, or other factors. In a sense, use of combined management techniques creates a conundrum; even if lake rehabilitation is successful, managers may not be able to identify which activity worked best and favored lake improvements.

To better understand responses to current management, we evaluated characteristics of 12 shallow lakes in Minnesota following rehabilitation of these sites during 2007-2011. We gathered post-treatment data from rehabilitated (managed) lakes and then assessed lake responses by comparing features of managed sites to those of similar untreated lakes within adjacent areas. We used two approaches to compare managed and untreated lakes. First, we summarized data on fish communities, SAV, and average total phosphorus, nitrogen, and phytoplankton abundance (as chlorophyll a, Chla) comparing these levels in rehabilitated and untreated sites. Second, we used ordination methods to compare limnological variables and SAV mass in managed sites with values from similar untreated lakes exhibiting clear- and turbid-water regimes. Our approach illustrates patterns among managed and unmanaged lakes, identifies challenges associated with evaluating shallow lake rehabilitation efforts, and we summarize what we believe are important considerations for shallow lake managers.

## **METHODS**

### **Selection of lakes**

During 2010-2011, we gathered data from 12 shallow lakes in Minnesota, all of which were managed between 2007 and 2010 (Table 1). Lakes were rehabilitated using combinations of water-level reduction (drawdown) and fish removal with 4 lakes managed by drawdown, 4 treated with rotenone, and 4 sites subjected to combinations of both approaches. In several cases, piscivorous walleye were also added (as fry) to suppress fathead minnows (*Pimephales promelas*) and increase zooplankton (Potthoff et al. 2008). In all cases, the management objective was re-establishment of clear-water conditions as described by Scheffer (2004), Zimmer et al. (2009). We included only managed lakes for which we could obtain at least one year of pre-management data, and we required that rehabilitation had been completed within the past 4 years. All lakes (managed and untreated comparison sites) lay within two ECS Ecoregions, Northern Glaciated Plains (N=7) and Western Corn Belt Plains (N=5, Omernik 1987). We made no attempt to evaluate cause-and-effect relationships, only to assess lake characteristics following management (most often including both fish removal and drawdown).

### **Fish Communities**

All fish sampling was done during July and August in 2010 or 2011 to estimate composition and abundance (biomass per unit effort) using a combination of gear deployed overnight. Three mini-fyke nets (6.5 mm bar mesh with 4 hoops, 1 throat, 7.62 m lead, and a 0.69 X 0.99 m rectangular frame opening into the trap) were set overnight in the littoral zone of each lake. One experimental gill net (61.0 mm multifilament net with 19.0, 25.0, 32.0, 38.0, and 51.0-mm bar meshes) was deployed along the deepest depth contour in lakes less than 2 m deep or along a 2 m contour in lakes with sufficient depth. The approach has been effective in shallow lakes in Minnesota (Herwig et al. 2010) and in small lakes from other regions (Tonn and Magnuson 1982; Rahel 1984). Fish were sorted by species, rated (counts per unit weight), and weighed in bulk. Data were summarized as the total biomass of each feeding guild (planktivores, benthivores, piscivores) collected in all four nets.

### **Nutrients, phytoplankton, and submerged macrophytes**

Surface (dip) water samples were taken from central locations of each lake once during July and concurrent with other sampling. Samples were frozen and transported to the University of St. Thomas (St. Paul, MN) for analysis of phytoplankton abundance (Chla), total nitrogen (TN), and total phosphorus (TP) using methods of APHA (1994) and Zimmer et al. (2009). Abundance of submerged macrophytes (SAV) was measured using modified techniques of Jessen and Lound (1962), and Deppe and Lathrop (1992). During July (2010) or August (2011) SAV was sampled at 15 stations in each lake, equidistant along four transects running the width of each basin. A single cast with a weighted plant rake was made at each location, and plant samples were gathered along 3 m of lake bottom. Plants recovered on each cast were weighed using spring scales (wet weight) and percent composition was recorded for each species. SAV data were summarized as average total mass of all rake samples (mean mass across the total number of rake casts) for each lake.

## **Framework for Evaluating Lake Responses**

Management activities at each of the 12 study lakes are summarized in Table 1 along with lake names, sizes, and other project information. Our evaluation was limited by lack of pre-rehabilitation data from 8 of these 12 sites. We were able to obtain a single year of pre-treatment data from an additional 4 managed lakes because these sites were subjects of another study. Also, as part of concurrent research in Minnesota and within these ECS regions, we gathered identical data from 48 similar but unmanaged shallow lakes during comparable periods in 2010 and 2011. We reasoned that this larger group of unmanaged lakes would provide biological benchmarks for comparing characteristics of managed sites, allowing us to illustrate similarity and patterns among lakes with contrasting management histories. Going one step further, we assigned unmanaged lakes to clear- or turbid-regime categories using Chla threshold values following classification of Zimmer et al. (2009). Use of these defined groups allowed us to assess whether characteristics of managed lakes resembled clear- or turbid-water regional counterparts. While less informative than pre- and post-treatment comparisons, data from unmanaged regional lakes did provide a means of broadly assessing lake rehabilitation success if positive outcomes could be inferred from similarity with clear-regime regional lakes (supporting dense SAV, relatively lower levels of Chla, TP, and TN).

We used non-metric multidimensional scaling (NMS; McCune et al. 2002) to compare characteristics of 12 managed and 48 unmanaged regional lakes. NMS is useful for exploring data patterns in cases like this because resulting plots emphasize similarity relationships among groups of study units (lakes in our case). The method also relaxes requirements for normal data distributions and linear relationships to environmental gradients. To identify factors responsible for underlying relationships, we calculated correlations between final NMS axes and variables in the original ordination matrix. We used Sorenson (Bray-Curtis) distance measures and preliminary models based on a 6-dimensional solution, with significance of axes assessed by Monte Carlo permutation (250 permutations). Axes in final ordinations models were based on stress reduction and were adjusted with varimax rotation to align axes and aid interpretation of patterns (McCune et al. 2002). All NMS procedures were conducted using PC-ORD v. 5 (McCune and Mefford 1999) and followed guidelines of McCune et al. (2002).

## **RESULTS AND DISCUSSION**

### **Trends in Fish Populations, SAV, and limnological features**

In general, details of drawdown responses varied widely among lakes and ranged from relatively brief and partial-, to full-basin de-watering lasting more than a single growing season. Use of drawdown and/or application of rotenone was only marginally effective in removing fish populations from managed lakes and planktivorous and benthivorous fish were well established in managed lakes by 2010 and 2011. Abundance of planktivores (such as fathead minnows) and benthivores (bullheads, carp) varied dramatically among rehabilitated lakes, but standard errors (SEs) indicated that mean mass levels could not be distinguished from those observed for unmanaged turbid-water sites (Figure 1A). Presence of fish in Levenson and Froland by August

suggested that fish either survived rotenone treatments during fall 2010, or quickly immigrated back into the lakes following management.

Mass of SAV remained relatively low in all managed lakes during 2010-2011 and values fluctuated only modestly between these study years. In only one managed lake (Teal lake) did SAV reach levels within the range of our regional unmanaged clear-water sites in 2010, and here SAV declined sharply by the following year (2011, data not shown). SAV data seemed to indicate marginal responses and low abundance in most managed lakes, with levels similar to those observed in turbid regional sites (Figure 1B). An important point is that variability in fish and SAV abundance (mass) among lakes and classes (rehabilitated, regional clear- and turbid-regime) were so high (during 2010-2011) that general trends among these categories were difficult to discern.

Chla (phytoplankton abundance), along with levels of TP and TN, also varied widely among lakes, but mean values in managed sites were intermediate between managed lakes and regional turbid-state sites (Figure 2A,B,C) although SEs overlapped among lake classes. TN levels showed higher overall variability than TP, but average TN values in managed lakes were generally similar to those in regional clear-regime lakes (Figure 2B,C) and this may be related to modest improvements in SAV (and resulting increases in denitrification rates) in managed sites.

### **Similarity Patterns Between Managed and Unmanaged lakes**

Overall patterns identified using NMS suggested that rehabilitated lakes had improved since management, but stopped short of transitioning solidly to conditions observed in regional clear-regime sites. Two significant dimensions (axes) in NMS explained approximately 95% of the variability in Chla, TP, TN, and SAV mass among lakes classified as rehabilitated, regional turbid-, and regional clear-regime sites (Table 2). Broadly, this suggests that managed lakes showed greater similarity with regional clear-regime lakes, yet managed sites remained somewhat intermediate between regional clear and turbid counterparts (Figure 3).

To clarify possible responses to “fish removal only” management, we included four lakes for which we had both pre- and post-treatment data. Comparing patterns for these lakes before and after management (rotenone treatments only) shows improvement, but scores remained in close proximity to those of regional turbid-regime lakes (Figure 3). We also examined correlations between Chla, TP and TN, and SAV and values showed that TN and Chla were most strongly associated with significant underlying NMS axes ( $R^2$  values on axis 1 = 0.82 and 0.54, respectively; Table 2).

### **CONCLUSIONS**

Our analysis indicates that shallow lake management (rehabilitation) via drawdown and/or fish removal often favors short-term improvements in water quality in shallow Minnesota lakes. Patterns also suggest that management favors conditions more like those noted for lakes in clear-water regimes, but we think these lake improvements should be viewed cautiously. First, results here depict only constellations among lake characteristics during one or two seasons within 3-5 years following lake management. Second, planktivorous and benthivorous fish were widespread in these lakes shortly after management and increases in these fish are known to trigger transitions to turbid-water regimes (Zimmer et al. 2009). Finally, while ordinations show evidence of modest lake improvements in terms of limnological variables and SAV, overall patterns suggested that lake responses typically stopped short of solid transitions to clear-water regimes.

Experiences of lake managers with these and other projects in Minnesota show that shallow lake improvements often persist no more than 5-10 years after management. Recent paleolimnological studies at shallow Minnesota lakes (Lake Christina and Blakesley WPA) help clarify reasons for this and indicate that sediment nutrients may be responsible for resilience of turbid-water conditions (Hobbs et al. 2012, 2014). In both Lake Christina and Blakesley WPA, historical lake sediments showed evidence of greatly increased nutrient loading since European settlement. While restoration of upland cover within lake watersheds has many beneficial effects, shallow lake characteristics are only loosely linked with present-day watershed land cover (Bayley et al. 2012, Hanson et al. 2012). This means that efforts to improve shallow lakes by conversion of uplands from row crops to grass may not be enough to trigger lake transitions to clear-water regimes, or to improve conditions for waterfowl and other shallow lake wildlife, at least on short time scales. Given the present uncertainty about triggering and sustaining long-term improvements in shallow lakes, we suggest lake managers consider the following. First, accelerated monitoring of shallow lakes is critical if we hope to improve understanding of what does (and does not happen) in response to lake management in Minnesota and elsewhere. Second, lake management approaches should continue to include methods favoring short-term improvements (like drawdown and fish removal) along with efforts to eliminate non-point nutrient inputs (by runoff from adjacent row crops). Third, high priority should be given to preserving clear-water shallow lakes with intact communities of SAV and limited surface-water connectivity. Fourth, lake managers and policy makers must realize that favorable lake responses to management are categorically successful, even if improvements persist for only several years. Managers should expect that drawdown, fish removal, or other efforts will need to be repeated if shallow lakes are to be maintained in clear-water regimes in highly altered landscapes. Finally, we need to refine models and forecasting tools to allow lake managers to anticipate implications of changing land-use patterns, temperature and precipitation, and economic fluctuations. An initial attempt to develop such a modeling approach is described in the accompanying research summary entitled: *Shallow lakes: assessing quality and predicting future change*.

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Table 1. Summary narrative of management activities applied to 12 shallow lakes in Minnesota during 2007-2011.

<b>Lake</b>	<b>Size (acres)</b>	<b>Rehabilitation Strategy</b>	<b>Evaluation Chronology</b>
Augusta	499	Full drawdown in 2008; lake re-flooded in 2009. Water control structure and fish barrier on adjacent waters.	2 yrs post-trt
Froland	15	Rotenone treatment during fall 2010	1 yr pre-& post-trt
Hjermstad	60	Partial drawdown implemented in 2008; rotenone-treated under the ice during 2008-09. Stocked with piscivores (walleye fry) in 2009 to reduce planktivores. Water control and fish barrier in place.	2 yrs post-trt
Leverson	27	Rotenone treatment during fall 2010.	1 yr pre-& post-trt
Maria	425	Full drawdown implemented from fall 2006 through fall 2007. Rotenone applied to remaining flooded areas under the ice in winter 2007. Water level remained low through 2010. Water control weir with stop logs and electric fish barrier in place.	3 yrs post-trt
Nora	60	Full drawdown implemented in 2007, reflooded by 2009. Began to refill in 2008, 40-50% open water by 2009. Metal half-riser structure with stoplogs functions as a fish barrier.	3 yrs post-trt
Sedan	62	Partial drawdown in 2007, full drawdown in 2008. Began to refill in 2009. Water control structure in place.	2 yrs post-trt
Spellman	300	Drawdown from 2006-08; full reflooding by 2009. Water control structure and fish barriers in place.	2 yrs
Teal	91	Partial drawdown during 2008; rotenone-applied under the ice during winter 2008-09. Water control structure in place.	2 yrs
Todd N	35	Rotenone treatment during fall 2010	1 yr pre-& post-trt
Todd S	23	Rotenone treatment during fall 2010	1 yr pre-& post-trt
Wilts	55	Natural partial drawdown during 2008; rotenone-treat of remaining standing water during fall 2008.	2 yrs post-trt

Table 2. Summary of  $R^2$  values from linear associations between limnological variables in shallow Minnesota lakes during 2010 and 2011. Both axes significant ( $P < 0.05$ ) and cumulative  $R^2$  for both = 0.95. Values show relationships between variables in main matrix and final fitted axes following non-metric multidimensional scaling with varimax rotation (details discussed in McCune et al. 2002).

	$R^2$ values	
	Axis 1	Axis 2
<b>ChlAve</b>	<b>0.535</b>	<b>0.217</b>
<b>TPAve</b>	<b>0.368</b>	<b>0.002</b>
<b>TNAve</b>	<b>0.817</b>	<b>0.002</b>
<b>SAVAve</b>	<b>0.327</b>	<b>0.006</b>



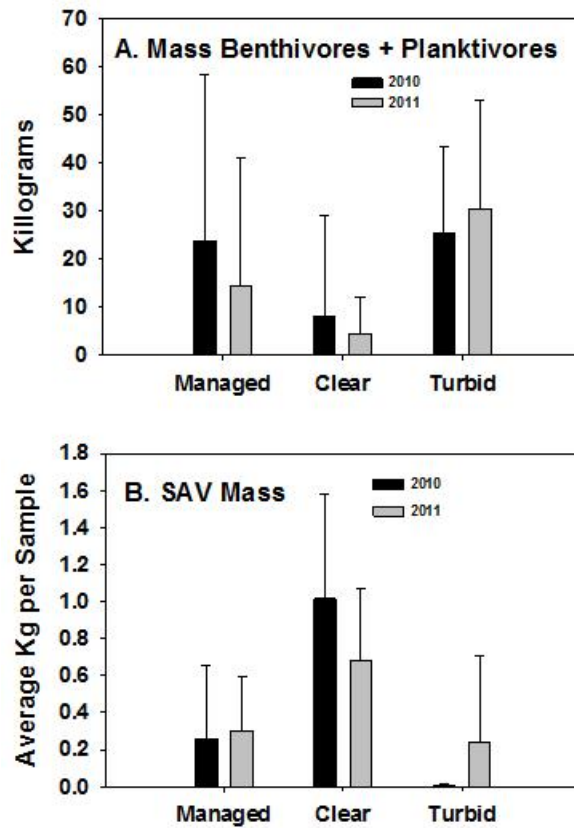


Figure 1. Relative abundance of planktivorous and benthivorous fish (average mass per lake) in study lakes during 2010 and 2011. Bars indicate means and SEs in regional unmanaged regional lakes in clear- and turbid-water regime, along with managed (rehabilitated) lakes as described in text (lake categories based on threshold values established following approach of Zimmer et al. 2009).

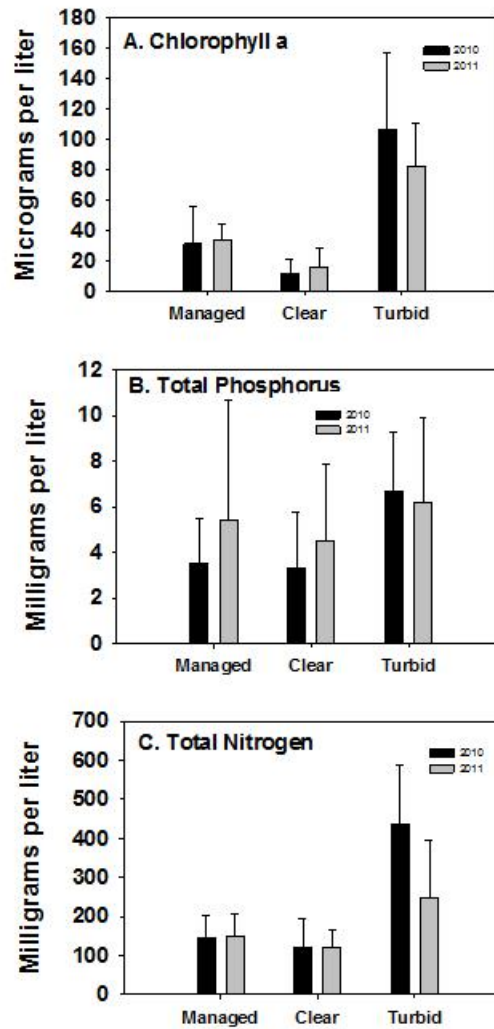


Figure 2. Bars depict chlorophyll a, total phosphorus, and total nitrogen concentrations in study lakes during 2010 and 2011. Values indicate means and SEs in regional unmanaged regional lakes in clear- and turbid-water regime, along with managed (rehabilitated) lakes as described in text (lake categories based on threshold values established following approach of Zimmer et al. 2009).

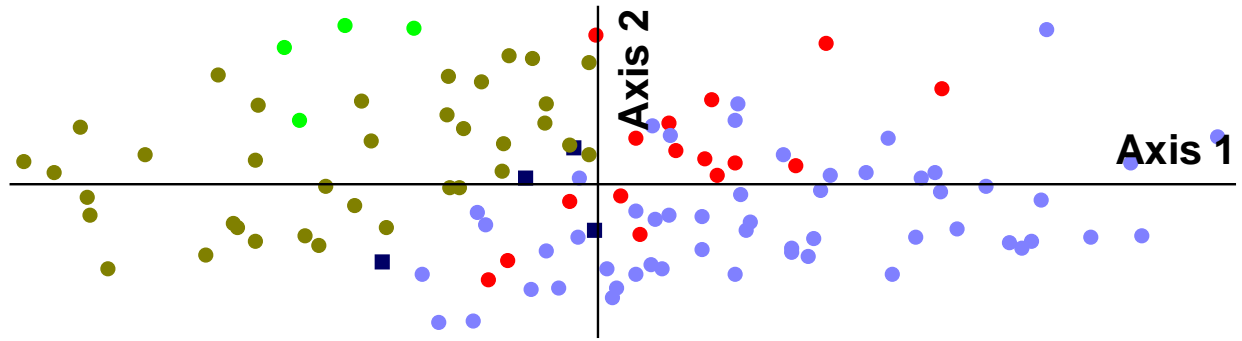


Figure. 3. Two-dimensional NMS summary of final non-metric multidimensional scaling model with varimax rotation using Chlorophyll a, total phosphorus, total nitrogen, and total SAV mass measured in 112 shallow Minnesota lakes during 2010 and 2011 (details discussed in text). Symbol color indicates lake rehabilitation status: regional clear lakes (blue), regional turbid lakes (olive), rehabilitated within 3-4 yr (red) lakes, 1-yr pre-rehabilitation (green), 1-yr post-rehabilitation (black box).

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