Summaries of Wildlife Research Findings

2016

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

SUMMARIES OF WILDLIFE RESEARCH FINDINGS
2016

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MOOSE CALF SURVIVAL, CAUSE-SPECIFIC MORTALITY, AND HABITAT USE

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SUMMARY OF FINDINGS

Adult survival and recruitment are important drivers of large herbivore population dynamics. The moose (*Alces alces*) population in northeastern Minnesota has exhibited a downward trend from 2006 to 2017. Our research was initiated because neonatal and seasonal survival rates and specific causes of mortality (e.g., predation, undernutrition, disease) of calves were largely unknown. Also unknown was habitat use of calves and their dams during vulnerable and energetically taxing life stages. We remotely monitored global positioning system (GPS)-collared adult female moose during the calving season to locate and GPS-collar neonates in 2013 and 2014 (n = 49 and 25, respectively). In 2015, we used behavioral cues of existing GPS-collared cows to identify calving behavior and calf mortality due to predation. We surveyed and compared habitat characteristics of pre-calving, calving, peak-lactation, and mortality sites to investigate important factors that predicted use at a fine and broad scale. Survival of calves dropped precipitously to 58% by 30 days of age in 2013 and 2014, and then to 34% by 9 months of age. Median age at death of calves that died before 1 year of age was 18.3 days. Hazard started low at birth and spiked at about 20 days old. Similar patterns were observed in 2015, with a 30-day survival rate of 63% and 10-month survival rate of 40.5% of uncollared calves. Over-winter survival was generally high in all 3 years. Predation was the leading cause of mortality in 2013 and 2014, with 84% of mortalities due to wolves (*Canis lupus*) or black bears (*Ursus americanus*). Predation was an important cause of mortality in 2015 as well, but the relative certainty in assigning cause was low. Calves were generally preyed upon once the dam and calves departed their calving sites. At a fine scale, canopy closure, total available forage, and calf concealment were important variables when distinguishing site types. Cows tended to move from areas of little concealment cover to areas of greater concealment but less forage to calve. During peak milk production, cows and their calves used areas with abundant forage and high concealment. At a broad scale, the amount of mixed and deciduous forest was lower around calving sites when compared to pre-calving and peak-lactation sites. Calf mortality sites also had less deciduous forest. Identifying specific causes of calf mortality and understanding their relations to various landscape characteristics 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

Ungulate population declines have been attributed to poor juvenile survival (Pinard et al. 2012, Forrester and Wittmer 2013). Large herbivore population growth is most sensitive to variation in adult survival, but differences in temporal variation of juvenile survival may be important in

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accounting for between-year variation in growth rates (Gaillard et al. 1998, 2000, Lenarz et al. 2010). 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, Severud et al. 2015*a*). Less is known about other specific ultimate and proximate sources of moose (*Alces alces*) calf mortality or contributing factors. 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 population-wide calf survival rates depends on the extant predator guild and relative densities of predator and prey (Eriksen et al. 2011, Patterson et al. 2013). The moose population in northeastern Minnesota has declined 55% from 2005 to 2016 (DelGiudice 2016). Survival and cause-specific mortality of calves was largely unknown in this area, but recruitment had been estimated at 0.40 mostly before the population began its decline (Lenarz et al. 2010).

Selection of calving sites by ungulates may influence neonatal survival. Often females tradeoff access to forage for predator avoidance during this vulnerable life stage (Bowyer et al. 1999, Poole et al. 2007). Cover may affect vulnerability to predation (Griffith and Youtie 1988). As nutritional demands for lactation increase and calves begin to incorporate browse into their diet, forage becomes more important. Lactation is an energy-demanding phase of reproduction, requiring 2–3 times more energy than gestation (Robbins 1993). Milk production peaks 21–31 days post-parturition for moose cows (Schwartz and Renecker 2007).

OBJECTIVES

1. Estimate neonatal (30-day), seasonal, and annual survival of moose calves
2. Quantify cause-specific mortality of moose calves
3. Identify potentially important covariates that influence survival or cause-specific mortality
4. Compare habitat characteristics at pre-calving, calving, peak-lactation, and mortality sites at 2 different spatial scales

METHODS

Our study area is the same as that of the Environmental and Natural Resources Trust Fund (ENRTF)-supported study in the Arrowhead region of northeastern Minnesota focused on survival and cause-specific mortality of adult moose (Carstensen et al. 2015). White-tailed deer populations occurred at pre-fawning densities of ≤4 deer/km^2 (Grund 2014). Major predators of moose in the area included gray wolves (*Canis lupus,* 3 wolves/100 km^2, Erb et al. 2016) and black bears (*Ursus americanus,* 23 bears/100 km^2, Garshelis and Noyce 2011). Moose had not been harvested in the state since 2012 (DelGiudice 2014).

As part of the adult moose mortality study, 84, 25, and 20 female moose were captured and fitted with Iridium GPS collars (Vectronic Aerospace GmbH, Berlin, Germany) during January 2013, February 2014, and February 2015, respectively (Butler et al. 2013; Carstensen et al. 2014, 2015). Blood was collected and analyzed for serum progesterone; ≥2.0 ng/mL was indicative of pregnancy. We monitored cow movements during pre-parturition and calving, with particular attention given to pregnant cows, looking for calving movements (Bowyer et al. 1999, McGraw et al. 2014, Severud et al. 2015*a*).

In 2013 and 2014, calves were located and fitted with an expandable Globalstar GPS Calf Collar (Vectronic Aerospace, Berlin, Germany). Details of calf captures, handling protocols, and mortality investigations can be found in Severud et al. (2015*a*,*b*). In response to capture-induced abandonment of calves and capture-related mortality of adults (DelGiudice et al. 2014, 2015; Carstensen et al. 2015), the Governor of Minnesota issued Executive Order 15-10 (28 Apr 2015), barring state agencies from conducting or permitting any collaring of moose in the state. We then monitored existing collared adult females for calving movements, and tracked dam behavior for indications of mortality movements.

We estimated birth-dates of all calves (2013–2015) based on dams' calving movements. We assumed calves were born 12 hours after the cow localized. In 2013 and 2014, time of death was estimated using the mortality mode of collars, and calf and dam locations relative to the mortality site. In response to a high rate of collar slippage in 2014, we conducted an apparent survival check flight in March 2015. In 2015, dam behavior was used to indicate calf mortality (time and location from which the dam initially fled was also the estimated time of death). We conducted flights via helicopter to assess seasonal apparent survival rates in 2015 during late November and early December (about 190 days old) and late March 2016 (about 320 days old).

We calculated Kaplan-Meier survival, hazard, and Cox proportional hazard using the R packages *survival, KMsurv,* and *muhaz*. Since calf births were tightly synchronized (Severud et al. 2015*b*), we calculated survival by calf age, with day 0 meaning birth. For smoothed empirical hazard curves, we used a global bandwidth and the product-limit method. We calculated cause-specific mortality rates with a cumulative incidence function using the R package *wild1*. Dam and calf location data were screened for locations that were thought to be erroneous fixes. We then calculated proximity between cows and calves. Summer field tests demonstrated mean linear error (± standard error, range) of locations for adult collars of 3.7 m (± 0.3, 0–17) under open canopy and 7.0 m (± 0.3, 1–36) under dense canopy (≥80% closure), and for calf collars of 24.9 m (± 2.7, 1–274) under open canopy and 40.3 m (± 1.3, 0–367) under dense canopy (Obermoller et al., unpublished data).

We collected site characteristics at the pre-calving site (location immediately preceding the calving movement) and presumed calving site (averaged coordinates over a 40- to 48-hour time period immediately following the calving movement, adjusted on site as confirmed by calving evidence; Figure 1). We similarly surveyed locations where calf mortalities were indicated by GPS locations of the dam (2013 and 2014) or confirmed by site evidence (e.g., calf bone fragments, hooves, hair, or predator sign in 2015). Calf mortalities occurring at the calving site were treated as having identical habitat conditions. When calf mortalities occurred outside of the calving site, new habitat data were collected.

Peak lactation of moose dams occurs 21–31 days postpartum (Schwartz and Renecker 2007). In 2015, when we observed evidence indicating a calf had survived ≥26 days (pellets and tracks), we collected site characteristics at the corresponding cow's peak-lactation location. We used the nearest GPS location from each collared cow with a known calf at 26 days post-calving. If the location was in the middle of a long distance movement, we used the center of the nearest grouping of ≥3 locations, which were usually 1 hour apart. We conducted all habitat work to match phenological conditions (i.e., leaf off and leaf on) to the time the initial location was recorded.

Habitat plots were centered at each cow's GPS location closest to the time of interest, unless that location was refuted by visual evidence. This typically occurred at calving and mortality sites, when we were able to see where a cow had calved or where a calf had died. In these cases, plot centers were placed in the middle of the cow's calving bed or at the primary location of calf remains or sign of a struggle. In the center of each plot, we collected an averaged waypoint using a handheld GPS unit, recorded the elevation from the unit's base map, and used a spherical convex densiometer to estimate canopy density. We also measured the prevailing slope and aspect using a clinometer and compass.

Canopy density (in addition to being measured at the plot center) and horizontal visibility were recorded 15 m from the plot center in each cardinal direction. We used a 2-m cover pole to determine horizontal visibility, recording the visible percentage (0, 25, 50, 75, or 100%) of each of 19 bands from the center (Poole et al. 2007). To estimate calf hiding cover, we held a cardboard cut-out of a standing moose calf silhouette at the center of the plot and recorded the percentage of the cut-out that could be seen from 15 m away at a 1-m height in each cardinal direction. The observer then moved towards the cut-out, maintaining a 1-m height, and recorded their distance from the calf when visibility reached 25, 50, 75, and 100%.

We recorded trees, saplings, and shrubs within an 11-m radius from the central point. Trees were defined as any upright (<45° lean) woody plant with a DBH ≥10 cm. Saplings and shrubs were defined as DBH <10 cm. We determined the species and DBH of each tree, alive or dead, within the plots, and counted number of stems of saplings and shrubs by species. Trees <18 cm DBH and shrubs were further classified as forage or non-forage species (Peek et al. 1976, Portinga and Moen 2015).

We used binary logistic regression to compare pairs of pre-calving, calving, peak-lactation, and mortality sites. The response variable was coded as 1 of these 4 site types. To avoid multicollinearity caused by correlated habitat covariates, we conducted pairwise correlation analysis on all variables. We removed single variables from a highly correlated pair ($|r| > 0.5$), retaining the most parsimonious set of variables. We developed *a priori* models using all possible combinations of remaining variables, and evaluated model support using Akaike's Information Criterion corrected for small sample size (AIC_c); models within 2 AIC_c units of the best approximating model (i.e., $\Delta AIC_c \leq 2.0$) were considered to have strong support.

To investigate broad-scale patterns, we buffered pre-calving, calving, peak-lactation, and mortality sites from 2013–2015 (sites defined using the same criteria listed above) with a 565-m radius to yield polygons of about 100 ha (Poole et al. 2007, McGraw et al. 2012). We overlaid these polygons on a land cover classification layer (Minnesota Land Cover Classification and Impervious Surface Area by Landsat and Lidar) and calculated the area (ha) of each class per polygon, or buffered location. We then compared land cover types by our defined site types using analysis of variance (ANOVA) with Tukey's Honestly Significant Difference (HSD) as a *post hoc* analysis.

RESULTS

We collared 49 calves from 31 dams in 2013 and 25 calves from 19 dams in 2014 (58% and 32% twinning rates, respectively). The sex ratio of collared calves was 36 females: 38 males. Seven dams abandoned 9 calves in 2013 and 6 dams abandoned 9 calves in 2014 (DelGiudice et al. 2014, 2015). These calves, as well as 2 additional calves that died during or shortly after capture from trampling by the dam and not nursing due to unknown causes (DelGiudice and Severud 2016), were not included in survival analyses, leaving 54 calves. Of these 54 calves, 4 slipped their collars in 2013 and 10 in 2014, allowing the study of survival and natural cause-specific mortality in 40 calves. In 2015, we observed calving movements or localization of 50 cows and tracked those dams for mortality movements. Assuming a 30% twinning rate (M. Schrage, Fond du Lac Natural Resource Management Division, unpublished data), this yielded about 65 uncollared calves under observation during 2015. Median calving dates for 2013, 2014, and 2015 were May 14, 19, and 10, respectively.

Blood profiles of calves sampled in 2013 were reported elsewhere (DelGiudice and Severud 2016). For the sample of all collared calves from 2013 and 2014, mean total body mass at capture was 15.8 kg (± 0.3, 12–20.5, $n = 38$) and mean hind foot length (HFL) was 45.9 cm (± 0.3, 42–49, $n = 42$). Body mass and HFL were weakly correlated ($r^2 = 0.31$, $P < 0.001$). There were no differences in mass or HFL by sex or between twins versus singletons. Mean rectal temperature was 101.6 °F (± 0.12, 99.9–103.4, $n = 43$). Mean dam age of all collared calves was 6.4 years old (± 0.5, 1–14, *n* = 43). Mean dam age of calves that died was 6.7 years old (± 0.7, 1–12, *n* = 23).

For pooled 2013 and 2014 collared calves, 30-day survival was 0.584 (95% Confidence Interval [CI] = 0.461–0.740, Figure 2) and declined to 0.341 (95% CI = 0.226–0.516) by 206 days of age (6–10 February 2014), when all remaining collars were removed (Figure 3). Incorporating data from winter survey flights to look for calves that slipped collars, survival is further adjusted to 0.285 (95% CI = 0.178–0.457). Nearly 80% of mortalities occurred by 1 July (about 50 days old) and 95% by mid-August (about 100 days old).

In 2015, we observed calf mortalities during the first 30 days of life, as indicated by mortality movements of dams. We were successful in confirming calf mortality in cases when the dam fled and made 1–7 return trips. Based on suspected and confirmed calf mortalities, 30-day survival was 0.632 (95% CI = 0.518–0.770, *n* = 54, Figure 2). For the uncollared 2015 cohort of calves, flights in early winter (30 Nov–3 Dec 2015) and late winter (28–29 Mar 2016) indicated an apparent survival rate of 0.442 and 0.405, respectively. In all 3 years, survival dropped dramatically from birth to age 50 days (Figures 2 and 3).

For collared calves in 2013 and 2014, dam age, HFL, mass, sex, and twin status did not meet the assumptions of proportionality, so we could not run Cox proportional hazard models. The empirical hazard function was low initially, and then peaked at about 15 days old before declining, with a second spike in hazard around 90 days of age (Figure 4). Mean age of death of calves that died before 1 year of age was 35 days old (± 7, 3–205, *n* = 31), but the median age was 18.3 days, very close to the peak in hazard. Mortalities from predation (*n* = 26) occurred 31.6 days (± 6.5, median = 17, range = 0–120.5) after leaving the calving site and occurred 1,553 m (± 289, median = 1,142, range = 107–5,788) from the calving site.

We documented 31 natural mortalities of collared calves in 2013 and 2014. Specific causes of mortality included 20 wolf-kills, 5 bear-kills, 2 natural abandonments, and 1 each of the following: drowning, abandonment of unknown cause, unknown predation, and an infection resulting from wolf bites (Figure 5). The cause-specific mortality curves rose rapidly from birth to 50 days of age. Over the first 9 months of age, the cumulative probability of being preyed upon by wolves was 50.2% (90% CI = 37.1–63.5), 11.7% (90% CI = 3.5–19.9) for bear predation, and 9.6% (90% CI = 2.9–16.3) for other causes. Predation accounted for 84% of all natural mortalities, with wolves having the greatest impact overall (77% of the predation events).

For uncollared calves born during 2015 we documented 11 natural mortalities, with 4 additional cases pending (no direct evidence of calf mortality, but predator scat [1 wolf, 5 bear] will be analyzed for presence of calf hair). We documented 8 wolf-kills, 1 bear-kill, and 2 unknown predator-kills (saliva evidence pending, calf remains located).

Most dams and their offspring (one outlier cow-calf pair excluded) were a mean of 101 m (± 1.5, 0–6,083) apart throughout the year. Much variation by individual and fate was apparent (Figure 6). The outlier was a twin that separated from its mother and twin in November. With this outlier included, the mean proximity of all dams and their offspring was 3,736 m (Figure 7).

We measured fine scale habitat characteristics at 34 pre-calving, 37 calving, 25 peak-lactation, and 5 mortality sites in 2015. For fine scale analysis, remaining habitat variables after removal of highly correlated variables included: slope, mean calf model visibility at 15 m (15 m vis), mean canopy closure, and total forage. We then evaluated 15 models of all possible combinations of variables.

There was high model uncertainty when comparing characteristics of pre-calving and calving sites; however, 15-m visibility was in 5 of 6 top models (Table 1). Median percentage of the calf model visible from 15 m was 40% less at calving sites when compared to pre-calving sites

(Figure 8). Canopy and forage were both significant predictors that distinguished calving from peak-lactation sites (Table 1). Peak-lactation sites had higher canopy closure and higher total amount of forage compared to calving sites (Figures 9 and 10). There was also high model uncertainty when predicting characteristics of calving and mortality sites, yet canopy or forage appeared in 5 of the top 7 models. Mortality sites had more open canopy and more forage than calving sites (Figures 9 and 10).

We analyzed 150 pre-calving, 155 calving, 73 peak-lactation, and 36 mortality sites from 2013 to 2015 at a broad scale. The discrepancy between pre-calving and calving sites is because not all cows made a calving movement. Only cows that still had a calf at heel 26 days postpartum were included. There were no differences by site type in the amount of open water, emergent wetlands, forested wetlands, conifer forest, regenerated forest, developed/urban, row crop, or grassland land cover types. However, pre-calving and peak-lactation sites both had more mixed and deciduous forest land cover compared with calving sites ($P < 0.03$; Figure 11). Mortality sites also had less deciduous forest compared to pre-calving and peak-lactation sites ($P < 0.03$; Figure 11).

DISCUSSION

We documented high mortality rates of moose neonates in this declining population. However, the mortalities tended to occur once the dams and their calves departed from calving sites. Peak energetic demands for dams due to lactation occur 21–31 days postpartum (Schwartz and Renecker 2007), which coincides with the highest hazard calves experienced. This suggests that dams seeking out high quality or quantities of forage to meet this demand may be travelling in risky areas or that movement to new foraging patches is itself risky, potentially exposing dams with young calves to predation. Our habitat surveys found that calving sites contained less forage, lower concealment, and decreased land cover types containing optimal foraging habitat than peak-lactation sites.

Our near-recruitment rates for 2013–2014 and 2015, although estimated in different ways (via collaring of calves versus observing cow movements and subsequent aerial surveys), were similar. Both methods required collars on adult cows, yet without calf collars extensive field searches and helicopter flight time were required. Tracking GPS-collared cow movements was a highly reliable way to estimate calving rates and to a lesser degree calf mortality. Due to the Governor of Minnesota's Executive Order 15-10, we were unable to confirm presence of calves shortly after birth, nor handle or collar calves in 2015. Without observing neonates at calving sites, we could not estimate twinning rates. We also did not know when a calf had died, but used dam movements as an indication of calf mortality. This also delayed site investigations, frequently making assignment of mortality cause difficult. Only in cases where the calf was ≤23 days old and the dam fled and made 1–7 return trips were we successful in confirming calf mortality. In a subset of those cases we could assign cause of death. This technique may serve as a method to estimate early neonatal mortality, but it has less power to detect mortality as calves age beyond 3 weeks (but see Obermoller et al. 2017). This method will not reliably detect calves that succumb to forms of mortality other than predation, because we have not documented cows fleeing from and returning to other mortality events (e.g., disease, drowning, abandonment, but see Obermoller et al. 2017).

Wolves accounted for the largest proportion of mortalities in all 3 years of the study. Wolf predation has been partially implicated in the decline of this population (Mech and Fieberg 2014) and has been shown to account for adult mortalities as well (Carstensen et al. 2015). However, adults have typically exhibited predisposing factors when preyed upon by wolves. The overall poor health of the northeastern Minnesota moose population (Carstensen et al. 2015, DelGiudice and Severud 2017) could potentially explain not only the high number of capture-induced abandonments we observed (DelGiudice et al. 2014, 2015), but also the high rates of predation on calves. Dams in other studies and study areas defended their calves less vigorously following harsh winters or if in poor nutritional condition (Keech et al. 2011, Patterson et al. 2013).

Dams and calves often were in close proximity throughout the first year of life. One outlier was a twin that did not follow its dam and twin across a large lake at about 175 days old. The lone twin returned to where the group had spent time and survived until mid-winter when she was captured to have her collar removed.

At a fine scale, pre-calving sites were relatively open (less concealment cover) with moderate levels of canopy closure and forage availability. Calving sites had more concealment cover but less forage, aligning with other findings that moose tradeoff forage for safety during calving (Bowyer et al. 1999, Poole et al. 2007). Peak-lactation sites had a more closed canopy and abundant forage, leading to high calf concealment cover. However, this cover also may cause dams to be unable to detect approaching predators (Poole et al. 2007). Mortality sites tended to be more open in both concealment cover and canopy, resulting in less forage available. Some of these habitat metrics could be influenced by phenology. Pre-calving and calving typically occurred pre-leaf-out, whereas peak-lactation and mortality sites occurred post-leaf-out. Indeed, horizontal cover and canopy closure increase dramatically in deciduous-dominated over- and understory after leaf-out.

At a broad scale, we observed calving sites surrounded by less mixed and deciduous forest cover types, which are important foraging habitat (Mabille et al. 2012), indicating again that cows are forfeiting forage availability when choosing calving sites. By examining land cover types as well as fine scale measurements, we can infer that the increased concealment and canopy closure we observed at peak-lactation sites were not only an artifact of phenology, but also a result of the habitat itself. Collared moose in Finland showed a similar pattern—cows calved in areas with minimal vegetation <5 m in height, but cows and their calves moved to areas with dense vegetation shortly thereafter, ostensibly to seek out high quality and quantities of forage (Melin et al. 2015).

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Table 1. Highest ranking *a priori* models for distinguishing pre-calving, calving, peak-lactation, and mortality sites of moose calves in northeastern Minnesota, May–July 2013. Statistically significant variables are marked with an asterisk (*). Only models within 2 units of the Akaike's Information Criterion for small sample size (AIC_c) for the best approximating model (i.e., $\Delta AIC_c \leq 2$) are presented. Canopy = mean canopy closure, Forage = total number of stems of forage species, 15m Vis = amount of calf silhouette visible from 15 m away from focal site (concealment cover).

Site comparison	Model	AIC_c	ΔAIC_c
Pre-calving vs. calving	15m Vis	99.185	0.000
	15m Vis + Forage	100.02	0.835
	15m Vis + Canopy	100.19	1.005
	Slope + 15m Vis	100.61	1.425
	Canopy	100.71	1.525
	15m Vis + Canopy + Forage	101.02	1.835
Calving vs. peak-lactation	Canopy* + Forage*	72.567	0.000
	15m Vis + Canopy* + Forage*	72.572	0.005
	Slope + Canopy* + Forage*	74.016	1.449
	Slope + 15m Vis + Canopy* + Forage*	74.131	1.564
Calving vs. mortality	Forage	33.172	0.000
	Canopy	34.196	1.024
	Slope	34.301	1.129
	15m Vis	34.641	1.469
	Slope + Forage	34.684	1.512
	15m Vis + Forage	35.104	1.932
	Canopy + Forage	35.117	1.945
Peak-lactation vs. mortality	Canopy*	23.042	0.000
	15m Vis + Canopy*	23.569	0.527
	Slope + Canopy*	24.165	1.123
	Slope + 15m Vis + Canopy*	24.27	1.228
	Canopy* + Forage	24.54	1.498

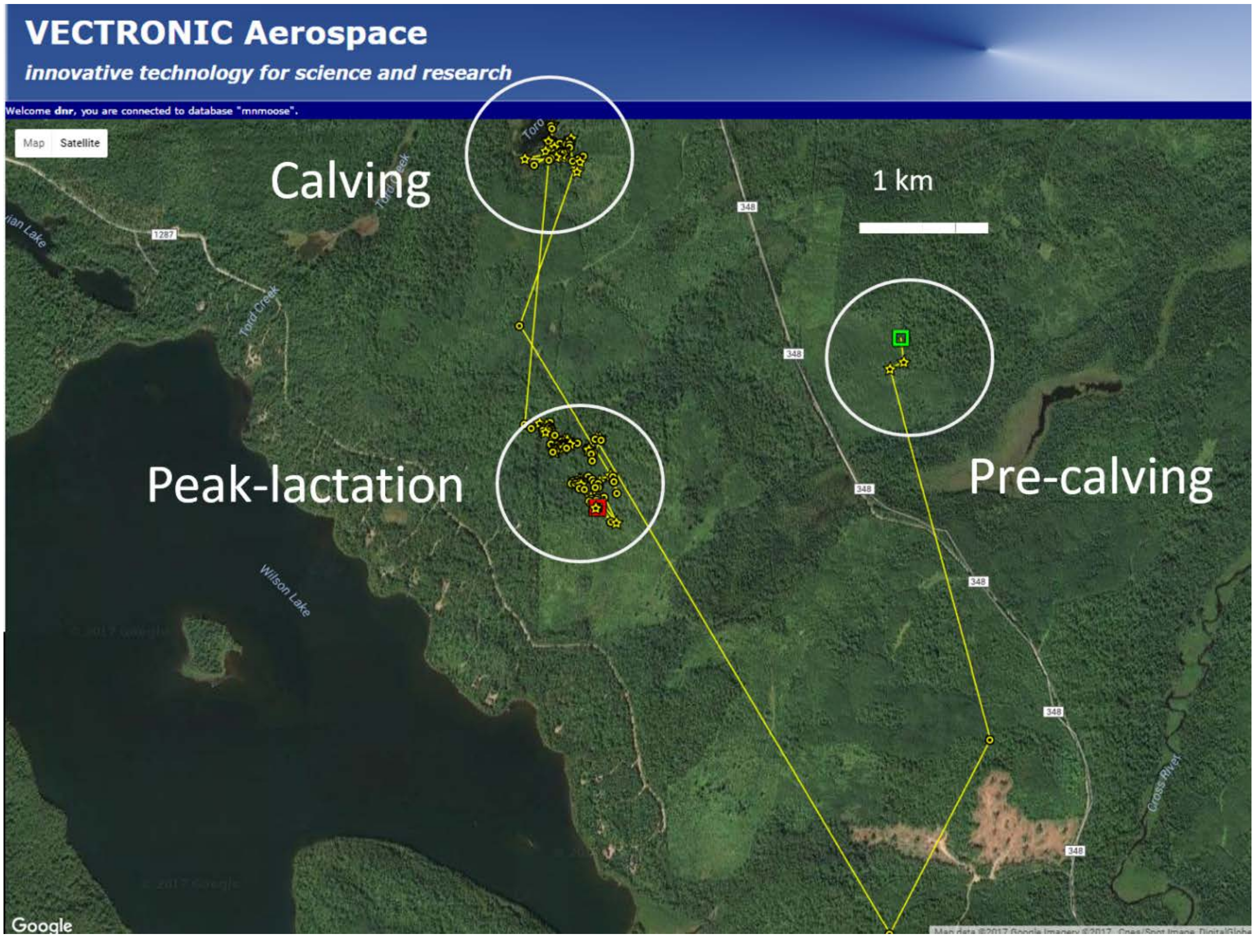

Figure 1. Example of moose pre-calving, calving, and peak-lactation sites, northeastern Minnesota, May–July 2013–2015. Pre-calving sites were defined as being where the calving movement originated. Peak-lactation sites were defined as locations of dams at 26 days from calving (milk production peaks 21–31 days postpartum).

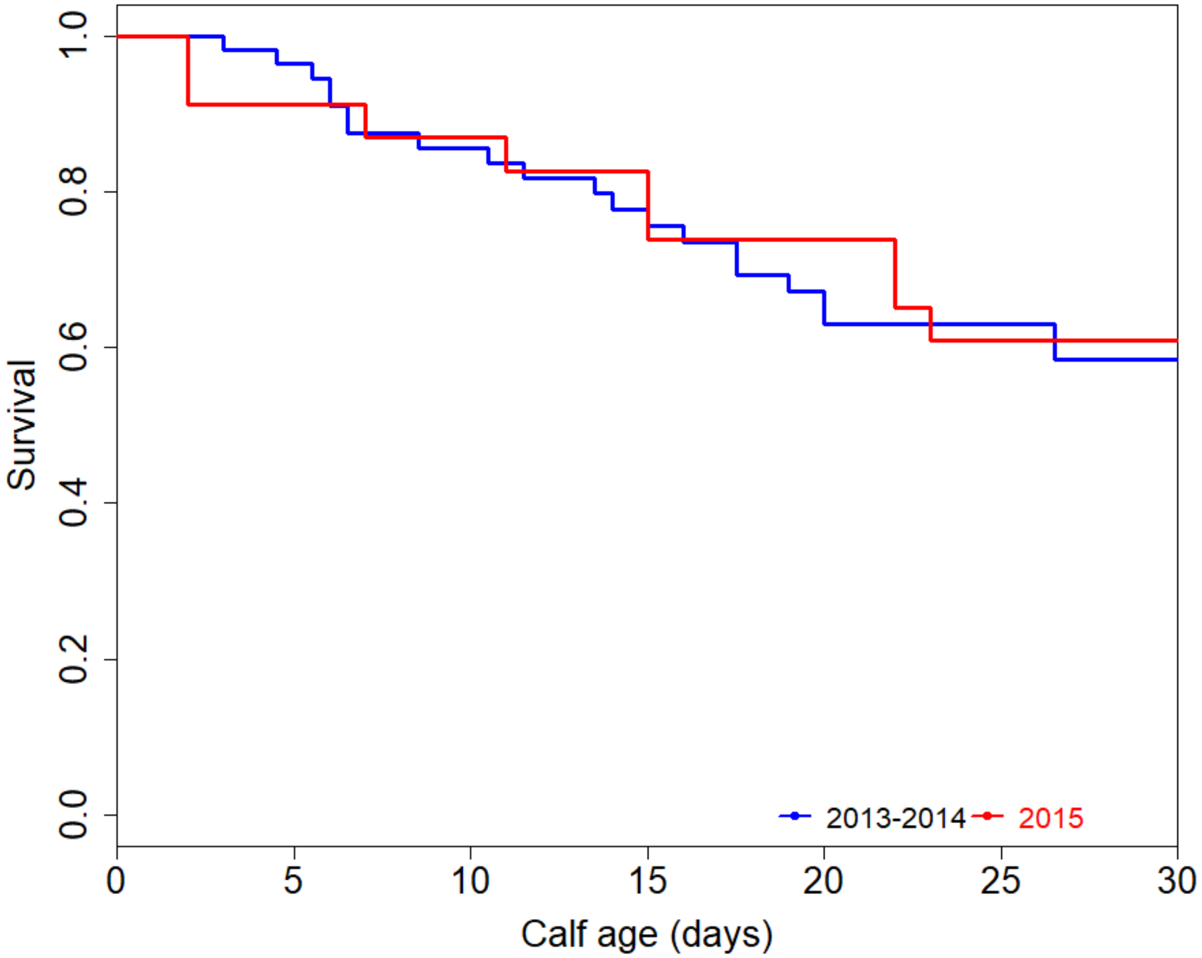

Figure 2. Kaplan-Meier 30-day survival for known moose calf mortalities, northeastern Minnesota, May–June 2013–2015. Mortality was confirmed by GPS collars (pooled 2013 and 2014, blue line, *n* = 54 calves) or through investigations triggered by dam movement patterns and observation of calf remains (2015, red line, *n* = 65 calves).

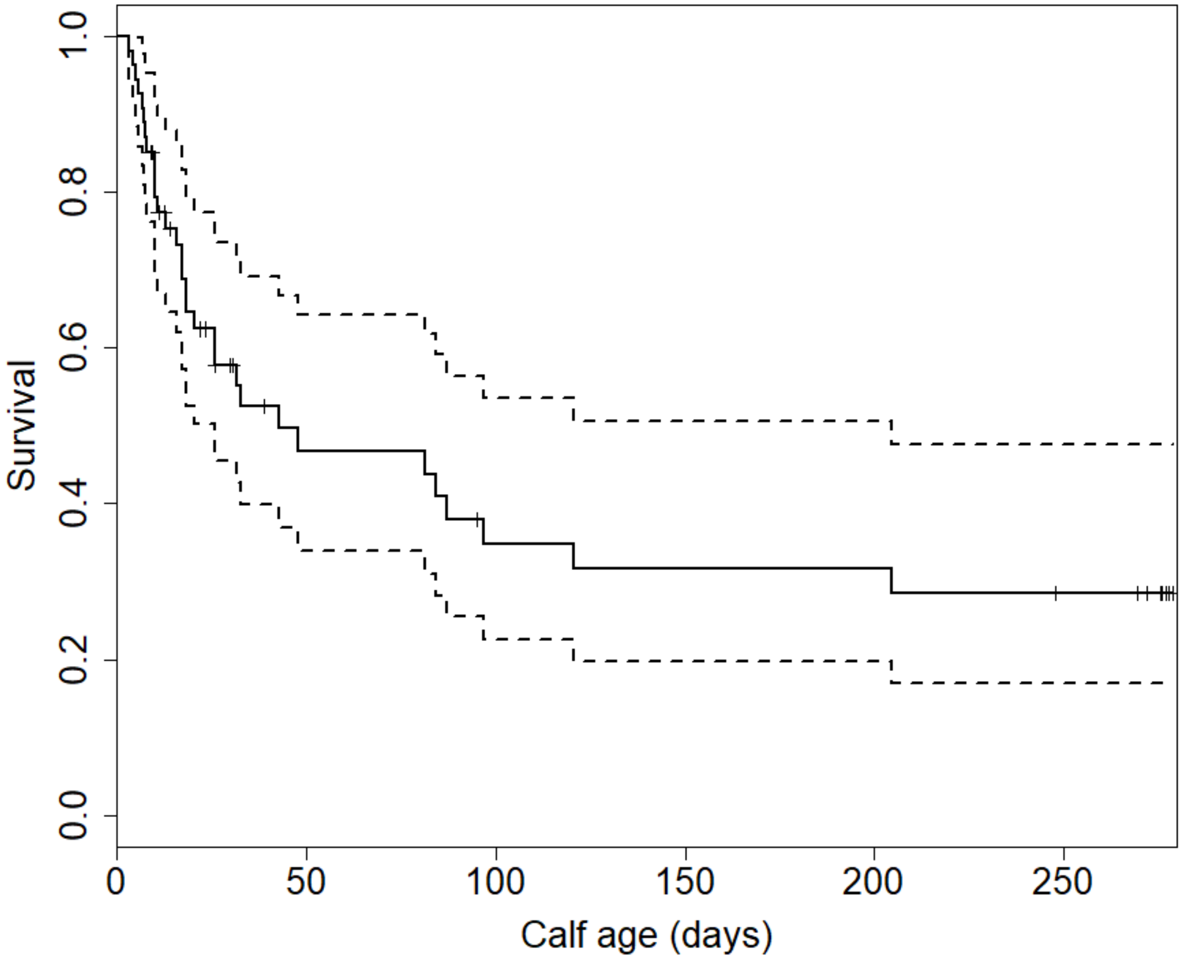

Figure 3. Kaplan-Meier 250-day survival for known moose calf mortalities (*n* = 54 calves), northeastern Minnesota, May–February 2013–2015. Tick marks indicate individuals censored due to slipped or removed collars. Dashed lines represent 95% confidence intervals.

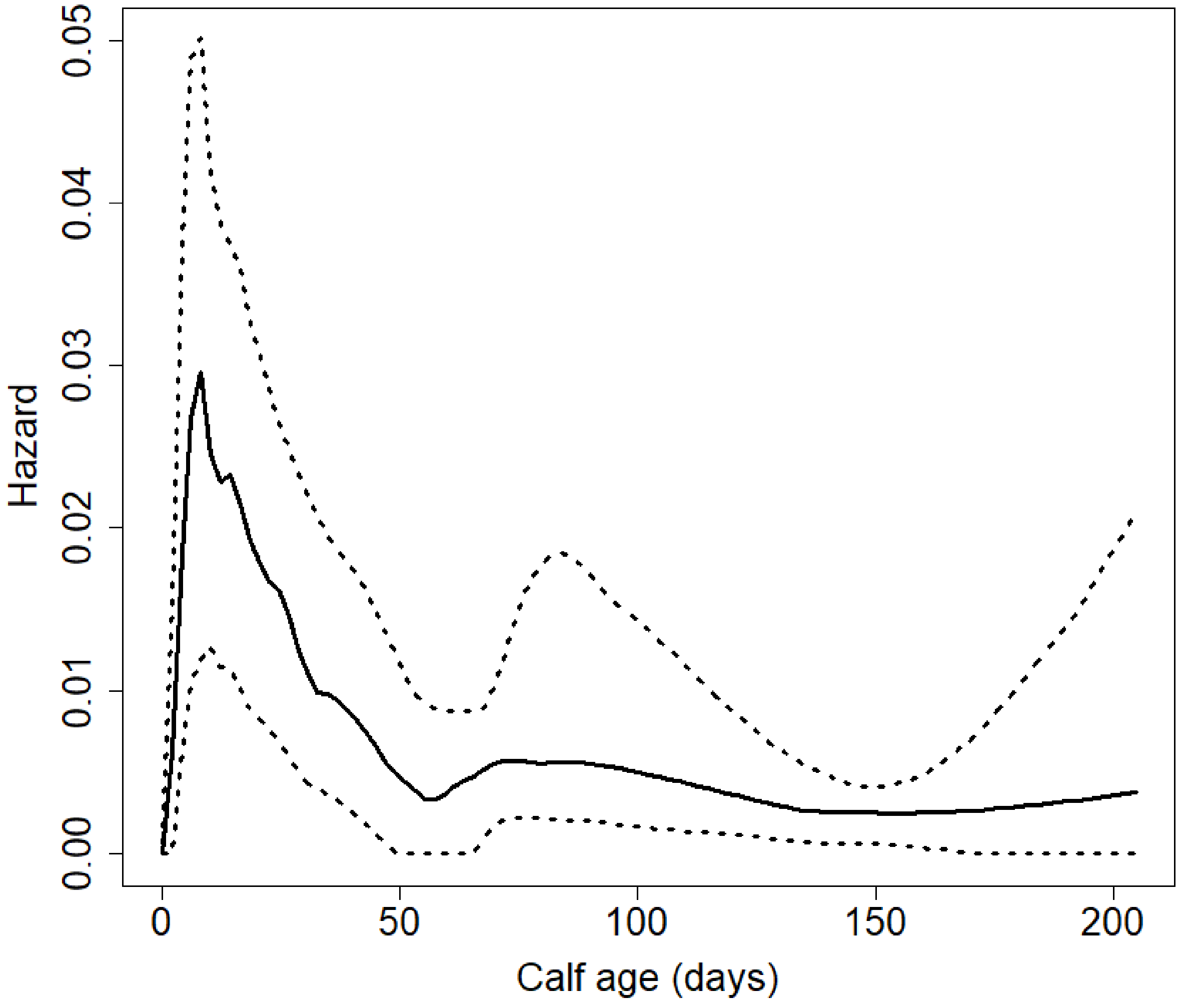

Figure 4. Empirical hazard function for known moose calf mortalities (*n* = 31 calves), northeastern Minnesota, May–February 2013–2015. Dashed lines represent 95% confidence intervals.

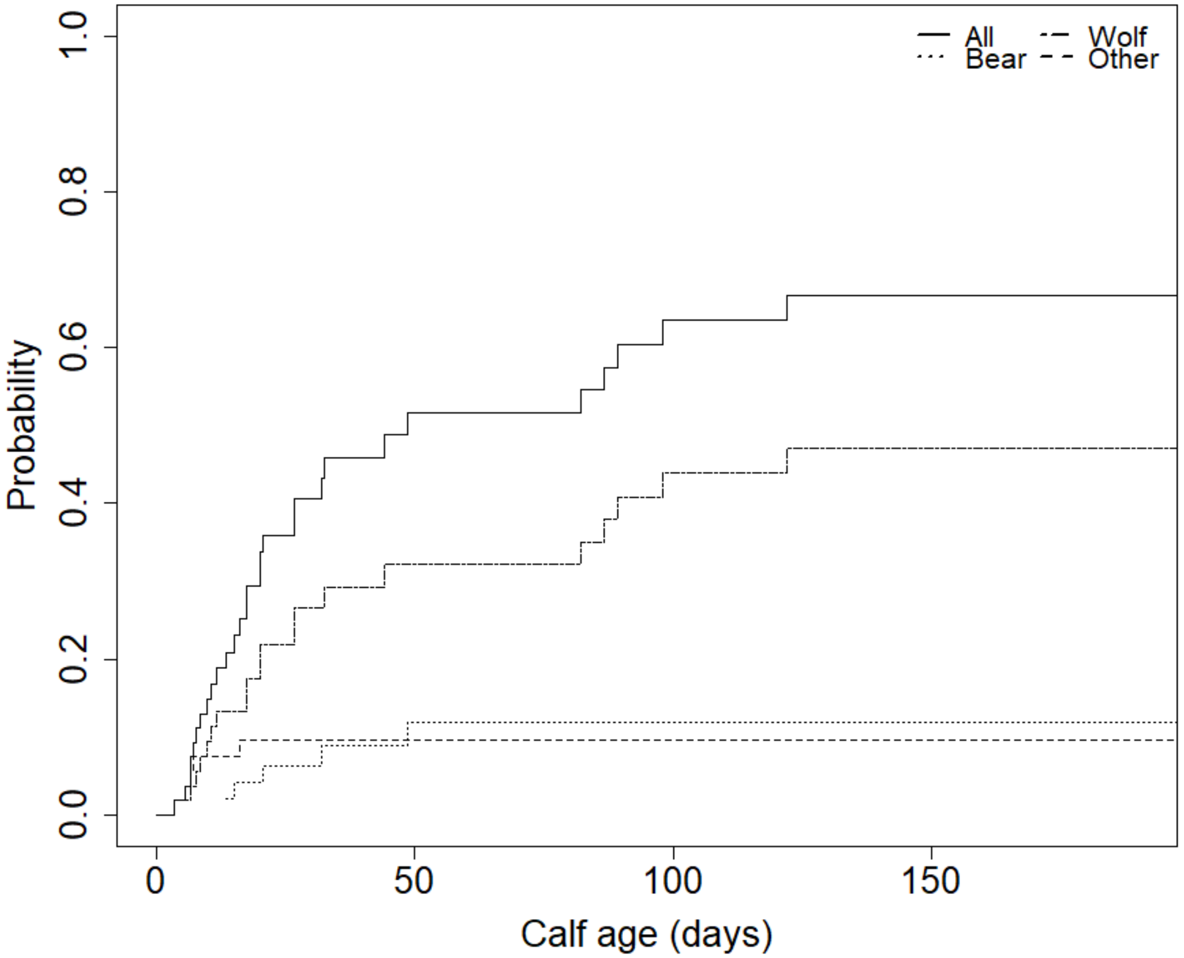

Figure 5. Cumulative incidence function for cause-specific mortality of moose calves in northeastern Minnesota (*n* = 40 calves), May–February 2013–2015. Causes of mortality were wolf predation (20), black bear predation (5), and other [natural abandonment (2), drowning (1), abandonment of unknown cause (1), unknown predator (1), and infection resulting from wolf attack (1)].

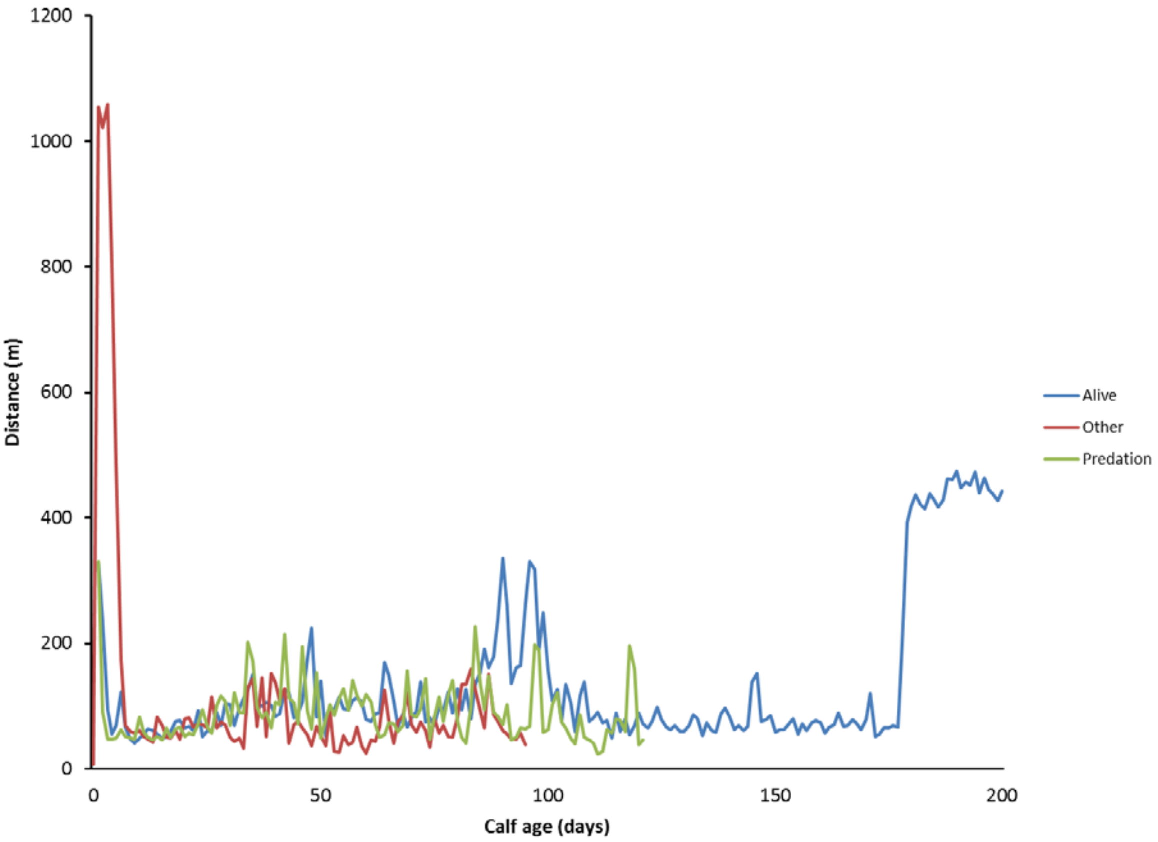

Figure 6. Mean daily distance between moose dams and their calves, excluding an outlier calf that moved up to 28,595 m from its dam, by calf age (up to 200 days old) and fate type (alive, other [non-predation mortality], and predation), northeastern Minnesota, May–February 2013–2015. Spikes in distance during the first 5 days were due to capture and handling.

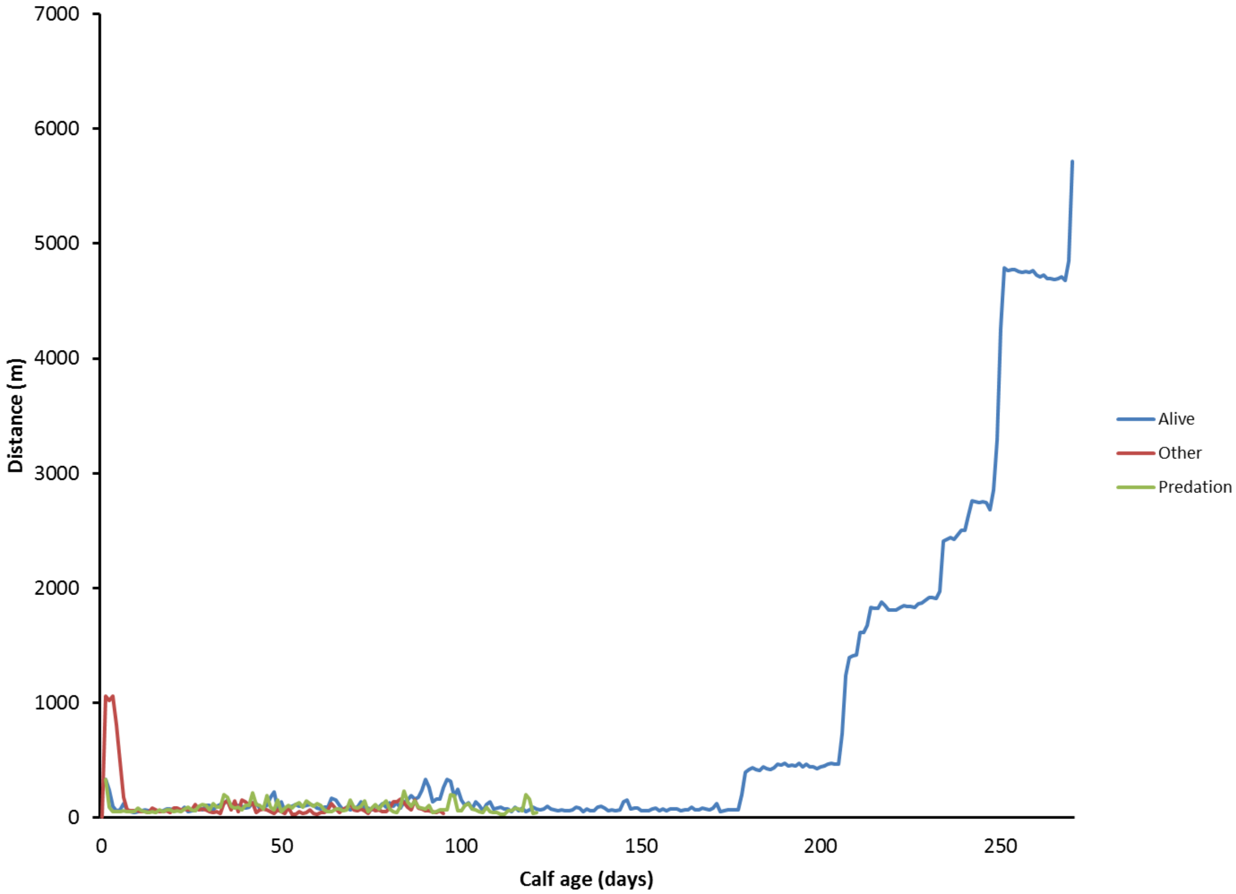

Figure 7. Mean daily distance between moose dams and their calves, including an outlier calf that moved much further from its dam than any other collared calf (up to 28,595 m), by calf age (up to 270 days old) and fate type (alive, other [non-predation mortality], and predation), northeastern Minnesota, May–February 2013–2015.

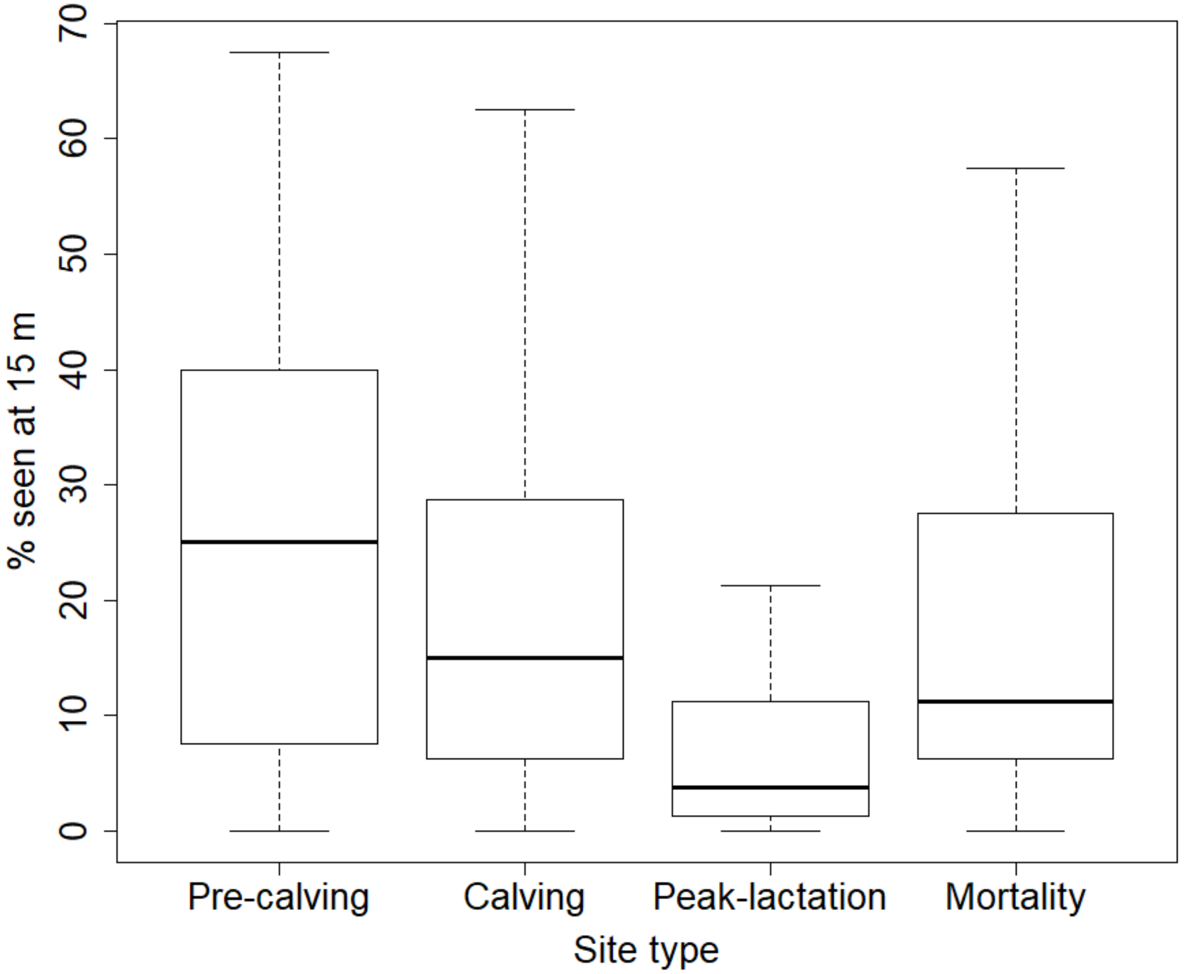

Figure 8. Percentage of calf silhouette visible from 15 m at pre-calving, calving, peak-lactation, and mortality sites (*n* = 34, 37, 25, and 5, respectively) of moose calves in northeastern Minnesota, May–July 2015. Boxes depict interquartile range and dark lines are median values.

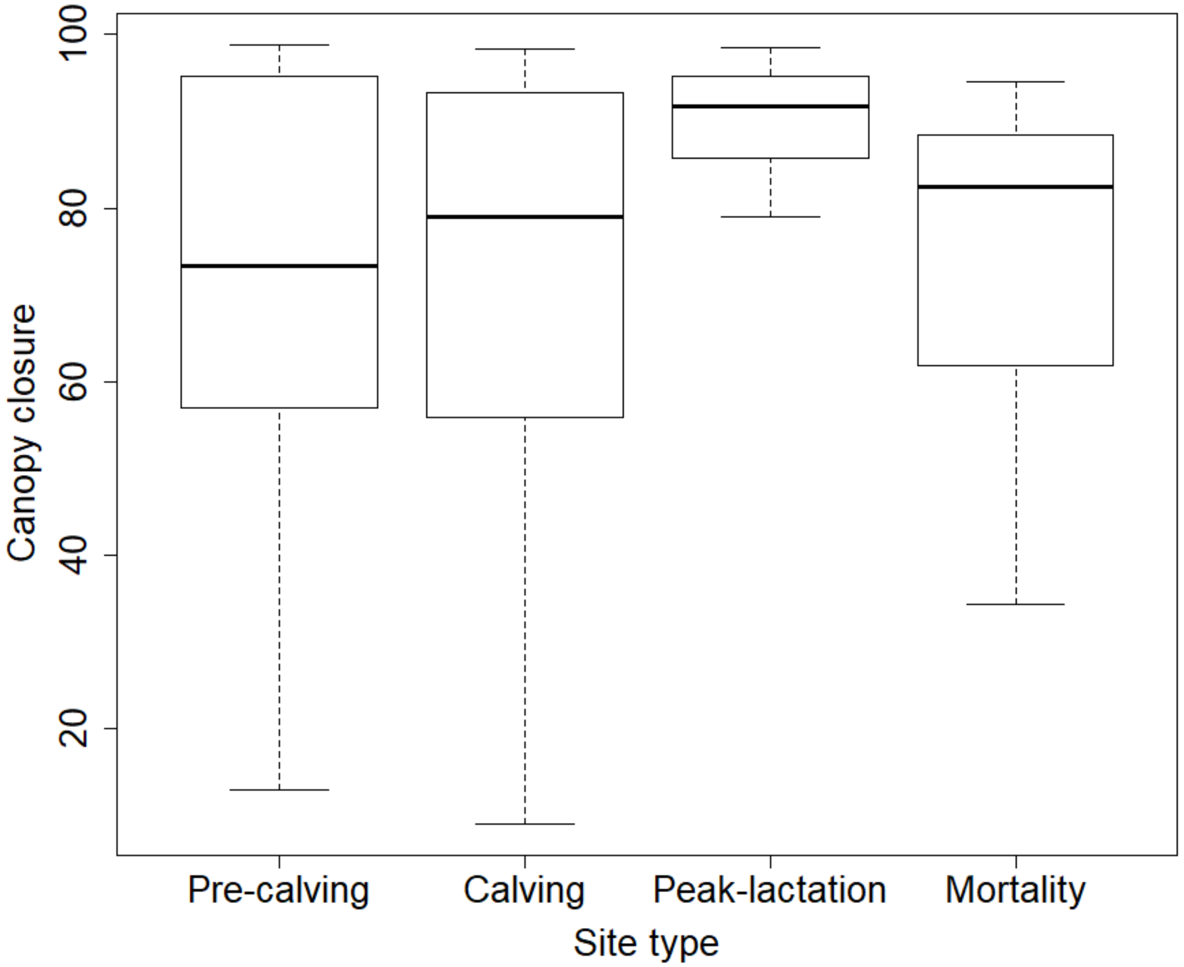

Figure 9. Canopy closure at pre-calving, calving, peak-lactation, and mortality sites (*n* = 34, 37, 25, and 5, respectively) of moose calves in northeastern Minnesota, May–July 2015. Boxes depict interquartile range and dark lines are median values.

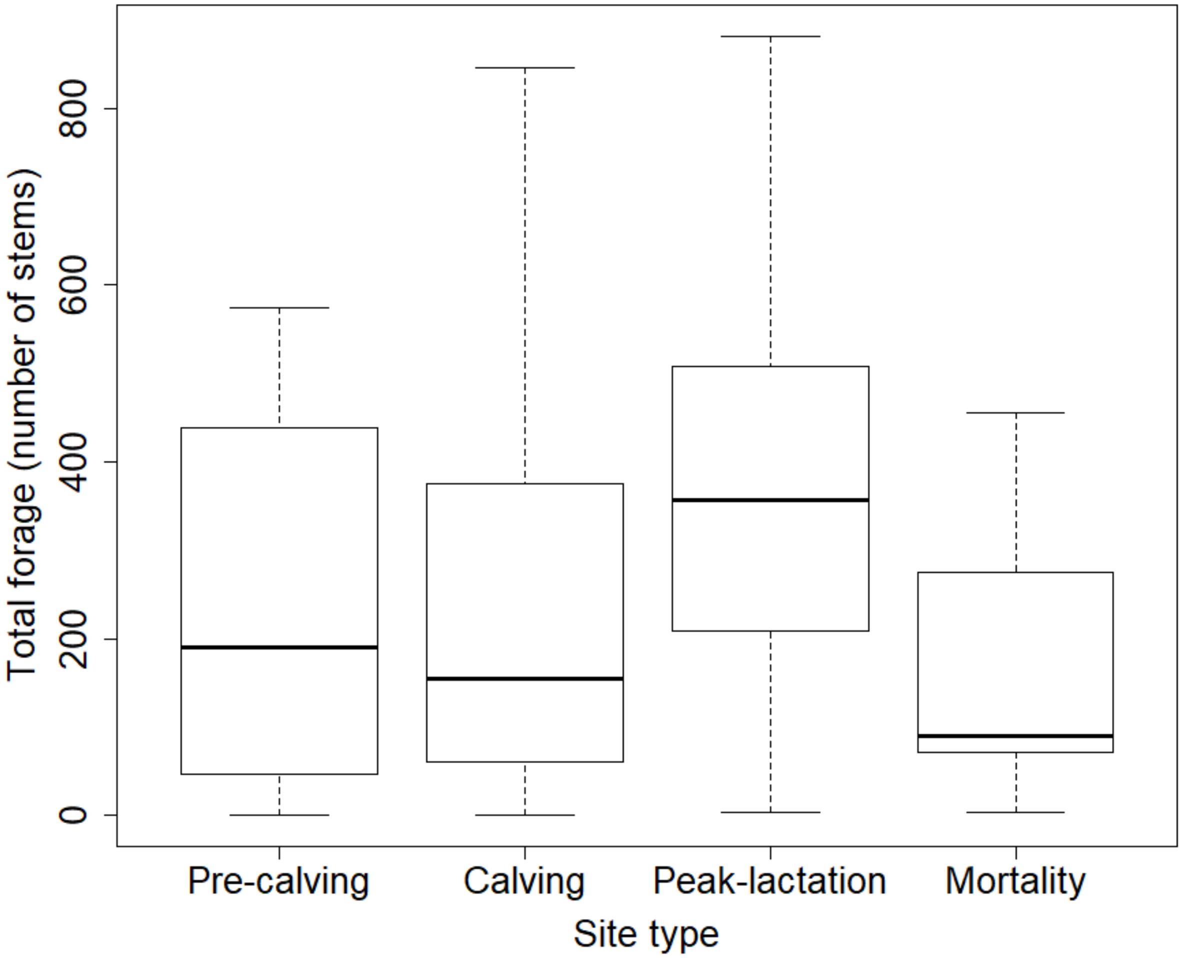

Figure 10. Number of forage stems at pre-calving, calving, peak-lactation, and mortality sites (*n* = 34, 37, 25, and 5, respectively) of moose calves in northeastern Minnesota, May–July 2015. Boxes depict interquartile range and dark lines are median values.

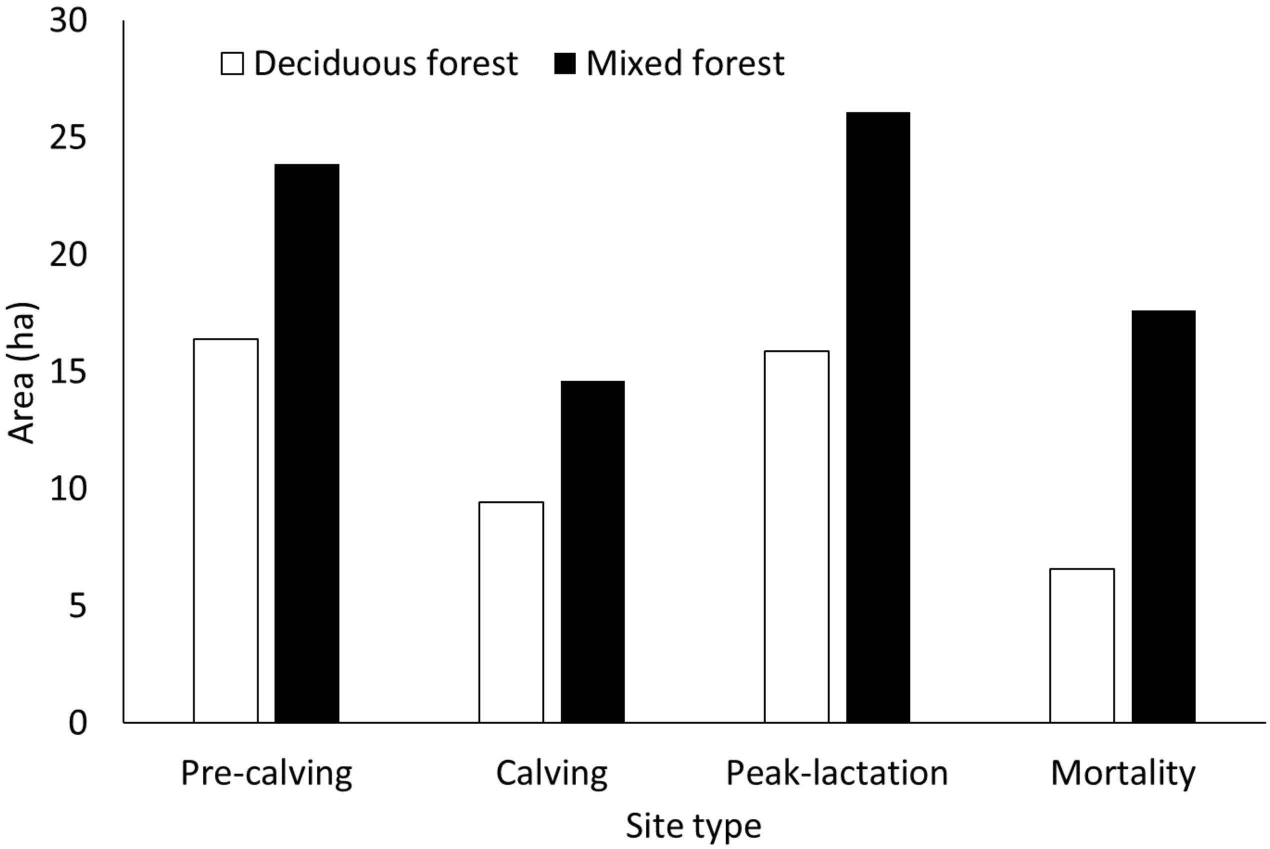

Figure 11. Amount (area in hectares) of deciduous forest (white) and mixed forest (black) within 100-ha buffers around pre-calving, calving, peak-lactation, and mortality sites of moose calves in northeastern Minnesota, May–July 2013–2015.

USING ADULT FEMALE MOOSE BEHAVIOR TO ESTIMATE CALVING AND MORTALITY OF CALVES

Tyler R. Obermoller[1], Glenn D. DelGiudice, William J. Severud[1]

SUMMARY OF FINDINGS

Continuing research on cause-specific mortality and annual survival of moose (*Alces alces)* calves in northeastern Minnesota is critical to understanding the long-term trajectory of the population. Thirty-five global positioning system (GPS)-collared adult females were computer-monitored beginning in late-April 2016 for calving movements, or a long distance movement followed by intense localization. We observed 28 of 35 (80.0%) make a calving movement, and along with additional visual observations of calves, determined 31 of the 35 (88.6%) cows were pregnant. Mean birth-date was 12 May 2016 (median = 11 May 2016, 24 Apr–10 June [range]). Following confirmation of calf presence (e.g., calf pellets, tracks, afterbirth), cows were monitored for a rapid, long-distance movement ("flee") followed by a return to the origin of the flee, indicating a possible predator attack. We observed evidence of 15 mortalities with a mean age at death of 30.6 days (± 15.5 [standard error], 2.5–243, *n* = 15). Specific causes of mortality included 9 wolf-kills (*Canis lupus*), 3 bear-kills (*Ursus americanus)*, 1 unknown predator-kill, and 2 deaths following vehicle collisions. Eight of 12 cows returned to the mortality site a mean 2.6 (± 0.5) times. Calf survival to 30 days of age was 66.7% (± 8%). Survival declined to 32.6% (± 8%) at 1 year of age, and the calf:cow ratio was 0.35. Understanding movement behaviors of cows can yield important insight into mechanisms driving the decline of the population in northeastern Minnesota and aid in future management decisions.

INTRODUCTION

The near disappearance of moose (*Alces alces)* in northwestern Minnesota since the mid-1980s and a dramatic decline of northeastern Minnesota's population since 2006 (Murray et al. 2006; Lenarz et al. 2009, 2010; DelGiudice 2016) prompted aggressive studies of survival and cause-specific mortality of adults and calves in 2013 in northeastern Minnesota (Figure 1) using cutting-edge global positioning system (GPS) collar technology (Carstensen et al. 2014, Severud et al. 2015*a*). Earlier work (2002–2008) in northeastern Minnesota focused on moose survival and employed very high frequency (VHF) telemetry (Lenarz et al. 2009, 2010). Current studies changed to GPS collars to facilitate more expeditious investigations of adult and calf mortalities (Butler et al. 2013, Severud et al. 2015*a*).

Adult survival and reproduction are the primary drivers of ungulate population performance (Gaillard et al. 2000, Raithel et al. 2007). Several studies have reported that low and highly

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variable juvenile survival contribute to population declines (Hatter and Janz 1994, Cooley et al. 2008, Forrester and Wittmer 2013). Ungulate calves are particularly vulnerable to predation within the first few months of life (Franzmann et al. 1980, Keech et al. 2011, Patterson et al. 2013). In the most recent study of cause-specific mortality of moose calves in northeastern Minnesota, 50% of the collared individuals died within 50 days of birth (Severud et al. 2015*a*). The majority of calves were killed by wolves (*Canis lupus*), and a smaller proportion by black bears (*Ursus americanus*). Other studies have reported similar findings (Ballard et al. 1981, Osborne et al. 1991, Keech et al. 2011, Patterson et al. 2013). Wolves may prey on calves throughout the entire year, whereas bears have their greatest impact closer to parturition when the calves are less mobile (DelGiudice et al. 2009, Basille et al. 2015).

A primary objective of 1 of the studies initiated by the Minnesota Department of Natural Resources (MNDNR) in 2013 has been to assess annual variation of cause-specific mortality of calves. Unique challenges to the study's protocol for capturing and handling neonates in 2013 and 2014 (e.g., capture-induced abandonment, DelGiudice et al. 2015) and adults in 2015 (e.g., capture-related mortality, Carstensen et al. 2015) resulted in a Governor's executive order that captures be discontinued. Consequently, since 2015, research has continued without the benefit of neonates fitted with GPS collars (Severud et al. 2015*b*, Obermoller et al. 2017). In 2015, we monitored 60 cows with functioning GPS collars for calving activity (i.e., calving movement), and subsequently for a "mortality movement" relative to a potential calf mortality. A mortality movement was described as a cow making a sudden long-distance movement ("flee"), followed by a return to the origin of the flee, often multiple times (Figure 2; Obermoller et al. 2017).

Dams with young calves display reduced movements (Testa et al. 2000), which allowed us to differentiate between cows with and without calves. The calves' limited mobility at an early age makes them particularly vulnerable to wolf and bear predation. Once dams lose their calf or calves, their movements may increase by approximately 12% within 48 hours (Testa et al. 2000). DeMars et al. (2013) applied a movement threshold (using a 3-day average) to caribou (*Rangifer tarandus*) dams. When the dam's movements exceeded 186.5 m/hour, the calf was assumed to be dead. "Normal movements" of females then resumed, because they were no longer limited by the mobility of a calf. These authors successfully detected calf survival up to 4 weeks of age using this threshold. We attempted to improve our understanding of the temporal and spatial aspects of maternal movement patterns relative to calf mortalities using 2013 and 2014 movement data of GPS-collared moose dams relative to known mortalities of their GPS-collared calves. We then applied that understanding to support detection and investigation of mortality events of calves born in spring 2016.

OBJECTIVES

1. Determine the pregnancy rate of GPS-collared adult females by intense computer-monitoring of movements associated with calving activity and field confirmation
2. Increase our understanding of dam movements relative to cause-specific mortality of calves
3. Identify, locate, and assign cause of mortality to moose calves with field confirmation
4. Determine seasonal and annual survival rates of moose calves

METHODS

Adult moose (128 females, 51 males) were captured and fitted with GPS collars (Vectronic Aerospace GmbH, Berlin, Germany) in winters 2013–2015 as part of a companion study examining survival and cause-specific mortality in northeastern Minnesota (Butler et al. 2013, Carstensen et al. 2015). Due to natural mortalities, malfunctioning GPS collars, and battery expiration, 35 adult females with functioning GPS collars were available for intense computer-monitoring during the 2016 calving season. Twenty-three cow collars were programmed to take locations every 4 hours and transmit these locations to our base station after 6 successful fixes. The remaining 12 cow collars were locked in “mortality mode,” programmed to take locations every hour, and transmit these locations after 11 successful fixes. We monitored 20 cows with functioning GPS collars for calving in May−June 2017: 12 collars took a location every 4 hours, while the remaining 8 recorded a location every hour. In early-May of each year, all cows were monitored for a calving movement, which is a long-distance movement followed by intense localization (McGraw et al. 2014, Severud et al. 2015*a*). Automated reports highlighting calving movements were generated twice daily based on a 4-hour fix rate for each cow (Severud et al. 2015*a*, Obermoller et al. 2017*;* J. D. Forester, University of Minnesota, unpublished data). In spring 2016, we verified calving by examining the calving site for calf presence (e.g., tracks, pellets, hair) or evidence of birth (e.g., scrape in the earth, afterbirth) after the cow left the area to avoid disturbing her or her calf. In a few cases, we confirmed the presence of a calf by a public-reported visual, camera traps, or by searching a subsequent localization. Following verification of a calf by 1 of these methods, we monitored each cow’s locations daily for a mortality movement (as previously described). All monitoring in 2017 was completed remotely, and calving was not verified by the aforementioned methods.

In 2013 and 2014, we captured and fitted 74 neonates with GPS collars (with mortality accelerometers) to monitor them for survival and mortality events (Severud et al. 2015*a*,*b*). We analyzed movement patterns of GPS-collared dams of GPS-collared calves that died of known causes in 2013 and 2014 to determine characteristics indicative of specific causes of mortality (e.g., wolf or bear predation, abandonment). Specifically, we used temporal and spatial analyses of mortalities of 2013 and 2014 to aid in identifying calf mortality events in 2015 and 2016. In 2015, a 50% success rate of detecting mortalities associated with a mortality movement was documented until moose neonates were approximately 3 weeks of age, after which, identifying mortality sites became increasingly difficult, because of increased movement rates of the cow-calf pairs (Severud et al. 2015*b*).

When we observed a presumed mortality movement we deployed a team to the site for an investigation. At the site, we searched in the immediate area for any sign of calf mortality. If no evidence was immediately found, we expanded our search to more efficiently and thoroughly cover the surrounding area. We conducted the search as a 3- to 4-person team; 1 person carried a handheld GPS and hiked in each cardinal direction, and the other team members spread out to the right of the first person in 10-m intervals. We hiked in this manner for about 200 m, returned to the origin, and repeated this process in the remaining cardinal directions. Due to various factors (e.g., calf age, habitat type, topographical limitations, and lack of cow or calf sign), we adjusted our search area as needed. We searched for carcass remains, sign of a specific predator, and other site evidence (e.g., broken vegetation, blood sprays) to lead us to a cause of death. Evidence indicative of a bear-kill included cached body parts, peeled or inverted hide, selective feeding on viscera or sensory organs, and claw marks across the body, whereas a wolf-kill would be indicated by long bones chewed on the ends, presence of the rumen and its contents, scattered remains over a large area, and puncture wounds on the head, neck, or hindquarters when present (Ballard et al. 1981, Severud et al. 2015*a*,*b*). The aforementioned information was used to assign specific causes of mortality. Following a mortality or unusual movements, we performed survival investigations to determine whether the cow had lost her calf or calves or had a remaining twin alive. We executed survival investigations by heading to localized areas previously occupied by the cow. Opportunistically, we placed camera traps in areas the cow frequently occupied to attempt to capture evidence of a calf.

We conducted surveys using a Bell Jet Ranger (OH-58) helicopter to locate all cows with functioning GPS collars and determine 8-month and near-annual calf survival (recruitment). We also determined which cows still had surviving calves to evaluate the success of our mortality and survival investigations from the previous summer. To calculate success, we had a predetermined expectation of whether each cow would have a calf or not based on their movements post-calving. We monitored the status of the cows via calf survival checks, mortality investigations, and movement rates. In cases where the cow was not located, we searched for a maximum 15 minutes before moving to the next animal.

We calculated Kaplan-Meier survival using the *KMsurv* package in Program R. A calf was assumed to be dead if not observed with its dam during survey flights; we assigned the mortality date at the median of the birth date and flight date. Cows not seen during the flights were censored from the analysis (Figure 4).

RESULTS

Thirty-one of 35 (88.6%) cows monitored in 2016 were determined to be pregnant via calving movement and site confirmation or by visual observation of a calf or calves. We observed 28 of the 35 (80.0%) cows, or 90.3% (28 of 31) of the pregnant cows make a calving movement. Mean duration of the calving movement was 24.5 hours (± 2.5 [standard error], 3.2–63.8 [range], n = 28), and mean total path length over this period was 5.3 km (± 0.8, 0.1–18.3, n = 28). Mean displacement from the start of the calving movement to the birth location was 2.3 km (± 0.6, 0.1–15.2, n = 28). Of the 3 remaining cows that were not observed making a calving movement, one came back “on air” during the calving season and a calf was confirmed via camera trap, another did not make a calving movement, but was seen with a calf by the public, and the remaining cow was killed by wolves with a calf *in utero* (Carstensen et al., unpublished data).

We confirmed evidence of a calf for 27 of 31 of our cows via calf pellets, tracks, afterbirth, or visualization of the calf (e.g., camera trap or seen by public). In the 4 remaining cases we were only able to confirm the presence of a calving bed, but subsequent evidence of reduced movements by the cow further increased our confidence a calf was still present. The mean birth-date was 12 May 2016 (median = 11 May, 24 Apr–10 Jun), with 75.0% of the localizations occurring during 4–14 May 2016 (Table 1).

The calving movement occurred over a mean 26.4 hours (± 6.0, 2.1–75.4, n = 16) in 2017. Subsequently, the cows localized for 6.4 days (± 0.9, 2.5–13.9, n = 15). Sixteen of 20 (80%) cows this spring (2017) made a calving movement. The mean birth-date was 11 May 2017 (median = 11 May, 1 May–17 May), with 81.3% of the localizations occurring during 7–13 May 2017 (Table 1).

We documented 15 calf mortalities from 28 mortality investigations, providing a 53.6% overall success rate. We observed mortalities at 12 of 21 (57.1%) investigations where a cow made a mortality movement. Following a mortality we checked and confirmed evidence (e.g., tracks, pellets, hair) of a surviving twin for 5 cows. The remaining cows were checked and had no confirmed evidence or had increased movement rates indicating a calf was not present. Based on the preponderance of evidence at each mortality site, we recorded 9 (60.0%) wolf-kills, 3 (20.0%) bear-kills, 1 (6.7%) unknown predator-kill, and 2 (13.3%) deaths following a possible vehicle collision (Figure 3). Accounting for both calf mortalities and confirmation of calf presence (e.g., calf pellets, tracks, afterbirth, or observation of calf), 30-day calf survival was 66.7% (± 8.0%, Figure 4). Calves died at a mean of 30.6 days (± 15.5, 2.5–243.3, n = 15) of age. We also had 5 cases where a cow made a mortality movement, but no evidence of a calf mortality was found. The cows’ behaviors (increased movements) following the mortality movement suggested a calf or calves had been lost; we believe the mortalities simply were not found within the searched area or the mortality occurred beyond this area. Winter survey flights confirmed these calves had been lost. Furthermore, during the flights we noted 4 calves had been lost, but no mortality movement had been observed.

The annual survival rate was 32.6% in 2016 (Figure 4). The mean distance cows fled following a mortality was 1,633.5 m (± 444, 126–5,805, *n* = 12). Cows that made return trips to the mortality site returned a mean of 2.6 times (± 0.5, 1–5, *n* = 8). Return trips were a mean of 106 m (± 25.9, 33.8–230, *n* = 7) from the mortality site.

On 17−18 January, we observed 14 moose calves from 25 of the original 35 cows monitored at calving. There were 10 moose not located during the winter survey (3 mortalities, 7 with non-functioning GPS collars). During the spring survey (19 Apr 2017), we located 6 moose calves from 17 cows; 8 additional moose were not located (3 cows were not found, 2 collars were remotely removed, 3 collars were non-functioning). The recruitment rate for the 2016 cohort was 20%, with 29.2% (7 of 24) of the cows giving birth to twins. Our calf:cow ratio from the spring survey (19 April 2017) was 0.35 (6 of 17).

DISCUSSION

Identifying parturition via the calving movement continued to be a reliable tool for estimating pregnancy rates. We had only 1 case where a cow did not make the calving movement and was subsequently seen with a calf. With this tool, recapturing GPS-collared cows to fit vaginal implant transmitters (VITs) each year is unnecessary, and it reduces cost and stress to the animals. Total path length and displacement associated with calving movements in 2016 were similar to movements of 2012 to 2015 (McGraw et al. 2014, Severud et al. 2015*a*,*b*). We noted that 80% of monitored cows (90% of our pregnant cows) made a calving movement; similarly, 82% of cows were observed making a calving movement in 2015. Our mean birth-date was 12 May, very similar to what was reported in 2013 and 2015 in northeastern Minnesota (Severud et al. 2015*a*,*b*). The mean birth-date of 19 May 2014 was much later and may have resulted from a severe and prolonged winter (Severud et al. 2015*a*). Normal birth-dates, as in 2013 and 2015–2017, may indicate relatively good health of adult females during the calving period and during the previous rut. Interestingly, the 2017 peak calving period was much shorter than in previous years, and so far, the range of birth dates follows a similar pattern (Table 1).

Compared to 2015, in 2016 we increased our success rate of locating mortalities using the mortality movement by 21% (Severud et al. 2015*b*). We also located 5 mortalities where the calf was ≥30 days of age at mortality; none were located past 21 days of age in 2015. This was likely most attributable to our addition of conducting formal searches (patterns versus casual searches) for mortality evidence and to more rigorous monitoring.

Our percentage of predator-kills was similar to those of the first 3 years of this study (2013–2015), increasing our confidence that predators, especially wolves, are the leading cause of calf mortality in northeastern Minnesota. Furthermore, as during 2013−2015, calf survival to 30 days in 2016 was low (66.7%), and less than half that (32.6%) by 1 year (recruitment), which highlights the contribution of poor reproductive success to the sluggish performance of the northeastern moose population (Gaillard et al. 2000, Raithel et al. 2007).

A wealth of valuable calf production (must assume twinning rates), survival, and cause-specific mortality data and information are missed when biologists must forego capturing and GPS-collaring moose neonates. However, identifying calf mortalities via the movements of GPS-collared dams provides researchers with an option for continuing, with limitations, assessments of the impacts of calf survival and cause-specific mortality on population performance.

ACKNOWLEDGMENTS

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Table 1. Calving date summary for GPS-collared cow moose in northeastern Minnesota, 2013–2017. Calving activity was concluded based on observation of a calving movement.

Year	Mean	Median	Earliest	Latest	"Peak" calving	% of calves during peak	WSI[a]
2013	14 May	14 May	2 May	2 June	6–17 May	73%	120–139
2014	19 May	18 May	5 May	16 June	11–22 May	75%	180+
2015	11 May	10 May	29 April	14 June	3–15 May	76%	100–119
2016	12 May	12 May	24 April	10 June	4–14 May	75%	67–105
2017[b]	11 May	11 May	1 May	17 May	7–13 May	81%	60–119

[a] Winter severity index (WSI) was calculated by accumulating a point for each day ambient temperature was ≤0° Fahrenheit (−17.8° Celsius) and an additional point for each day snow depth was ≥15 inches (38.1 cm)

[b] 2017 calving season not completed, values may change.

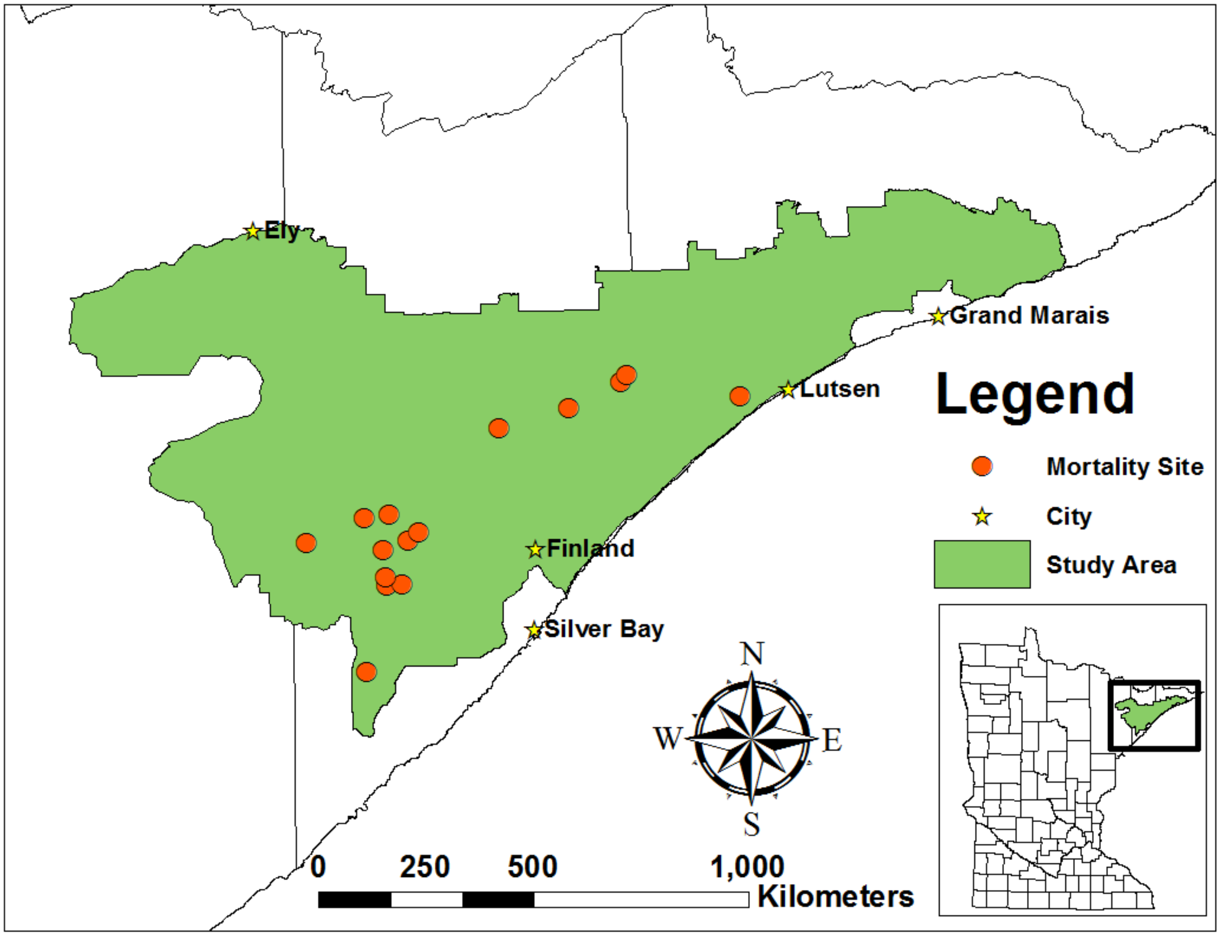
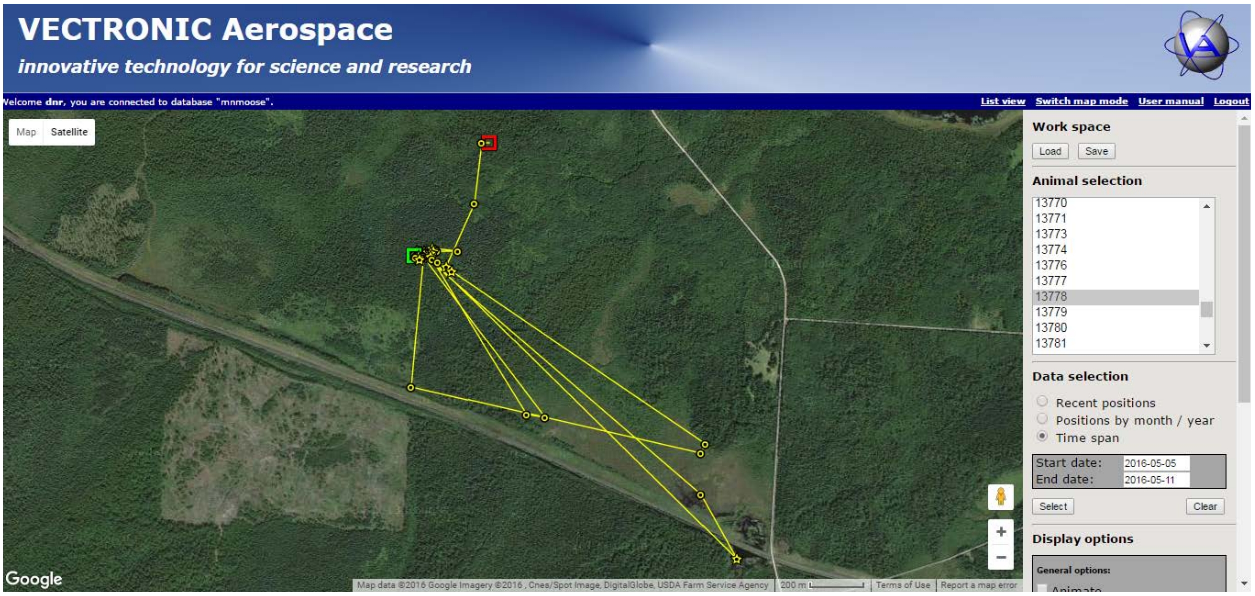


Figure 1. Moose calf study area (6,068 km^2) for examining survival and cause-specific mortality in northeastern Minnesota, 2016–2017. Solid orange circles depict mortality sites (*n* =15) of moose calves during May 2016–January 2017.



elcome **dnr**, you are connected to database "mnmoose".

○ Recent positions
○ Positions by month / year
● Time span

Start date: 2016-05-05
End date: 2016-05-11

Select
Clear

Figure 2. Vectronic Aerospace website (https://www.vectronic-wildlife.com) displaying the path of adult female moose 13778 in northeastern Minnesota, 5–11 May 2016. The green and red squares represent the beginning and end of the interval, respectively. The cow's movements show flees and return-visits to the green square; a mortality occurred on 5 May 2016. This cow made 3 return-visits before leaving the area. We found 3 wolf scats at the mortality site, which consisted of calf hair, teeth, vertebrae and other bone fragments. The estimated age at mortality was 2.7 days.

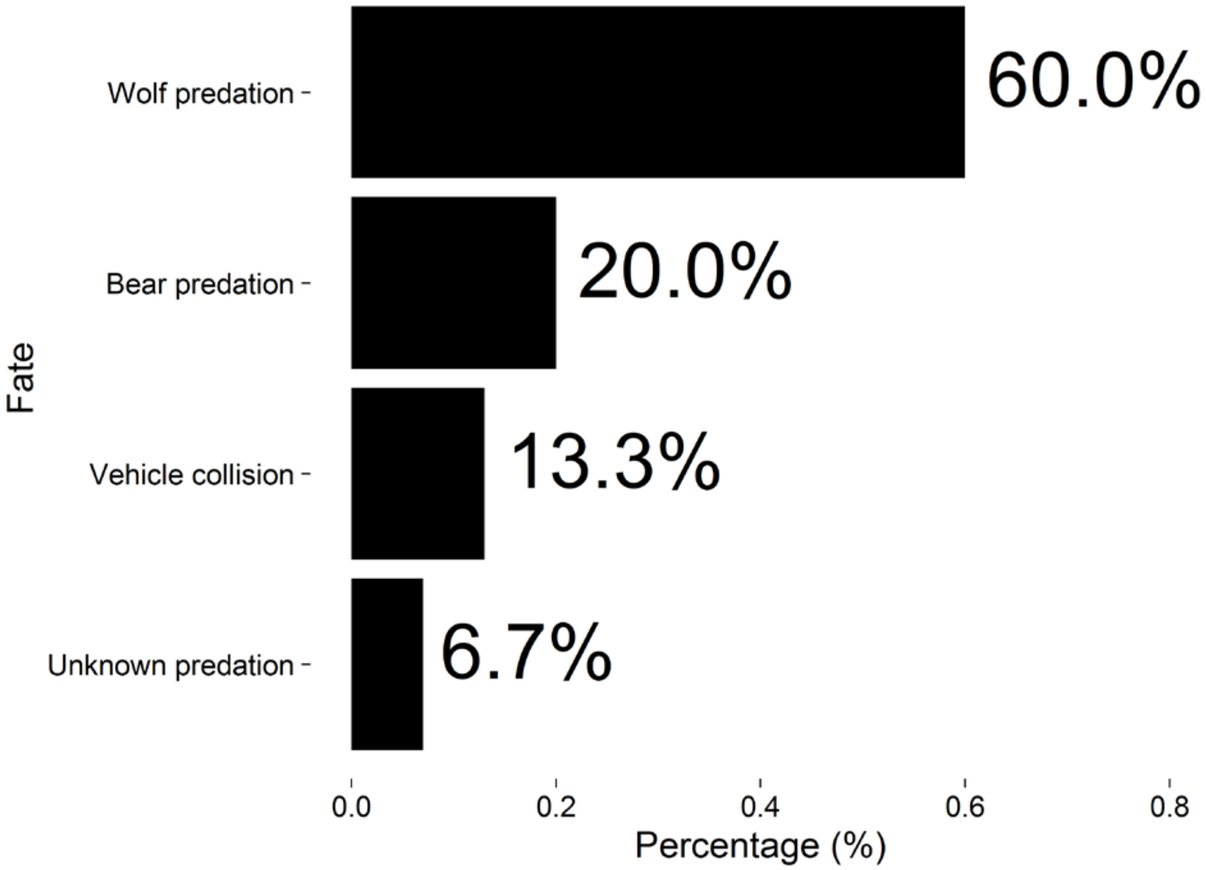


Figure 3. Cause-specific mortality of moose calves (*n* = 15) in northeastern Minnesota, May 2016–January 2017.

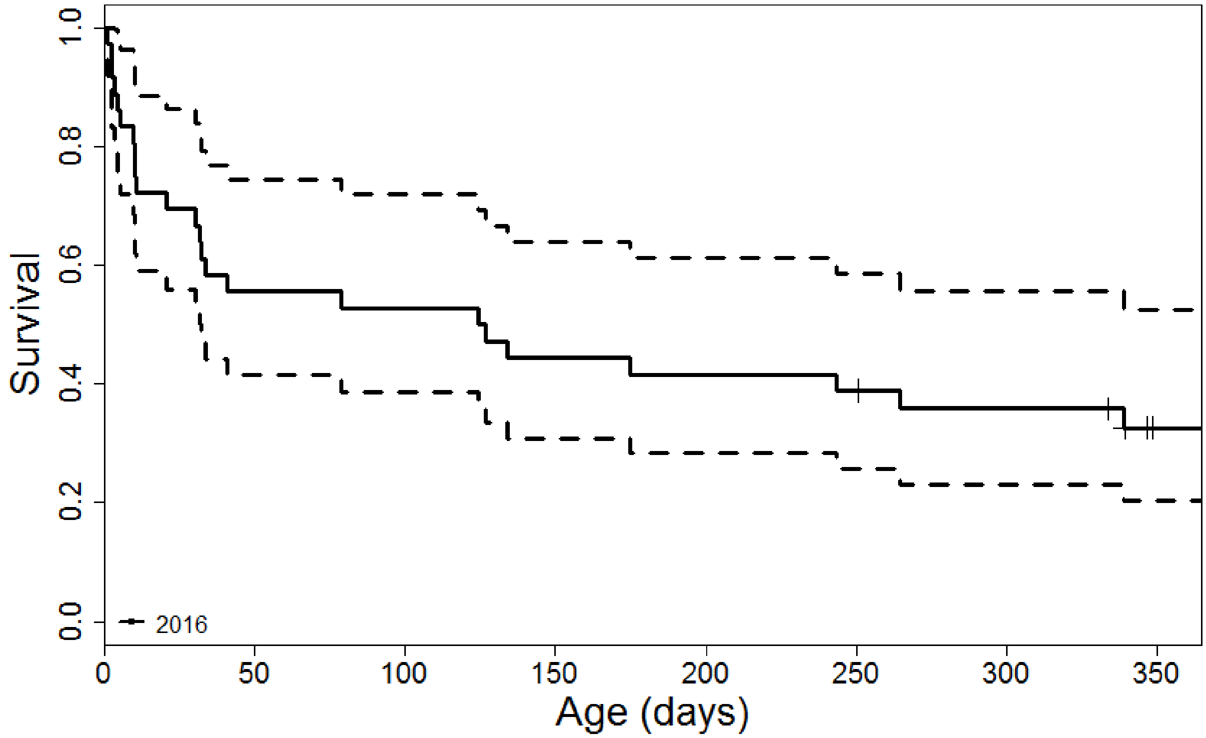


Figure 4. Kaplan-Meier annual survival (± 95% confidence intervals) of moose calves in northeastern Minnesota, 2016–2017.

CLIMATE CHANGE, WINTER NUTRITIONAL RESTRICTION, AND THE DECLINE OF MOOSE IN NORTHEASTERN MINNESOTA, WINTERS 2013–2017

Glenn D. DelGiudice, William J. Severud[1,] and Tyler R. Obermoller[1]

SUMMARY OF FINDINGS

The moose (*Alces alces*) population in northeastern Minnesota has declined an estimated 58% from 2006 to 2017. As in northwestern Minnesota, a number of complex ecological relationships between undernutrition, pathogens, predation, and environmental factors (e.g., habitat, temperature) are likely exerting pressure on moose and contributing to this recent decline. Nutrition is centrally related to our understanding of all other aspects of wildlife ecology, including population performance. Winter nutritional restriction of moose and other northern ungulates may be physiologically assessed by serial collection and chemical analysis of fresh urine in snow (snow-urine); urea nitrogen:creatinine (UN:C) ratios have shown the greatest potential as a metric of winter nutritional status with values <3.0, 3.0–3.4, and ≥3.5 mg:mg being indicative of moderate (normal), moderately severe, and severe nutritional restriction, respectively. During 6 January–28 March 2013–2017, we collected annual totals of 123, 307, 165, 189, 160 moose snow-urine samples, and mean seasonal UN:C ratios were 3.7, 2.9, 2.9, 3.5, and 3.7 mg:mg for the 5 winters, respectively. The mean population UN:C ratios for winters 2013, 2016, and 2017 were above the threshold indicative of severe nutritional restriction (i.e., a starvation diet) and accelerated body protein catabolism. During 2014 and 2015 the corresponding values reflected moderate nutritional restriction. Most indicative of the unique severity of nutritional restriction in 2013, nearly one-third of all samples collected yielded UN:C ratios ≥3.5 mg:mg.

Perhaps the ultimate value to management of nutritional assessments of free-ranging animals comes when the findings can be related to the performance and dynamics of the population and other ecological factors challenging that performance. Presently, our population-level nutritional assessments are closely tracking population estimates ($r^2 = 0.75$) and calf production ($r^2 = 0.85$) of moose in northeastern Minnesota. Although nutritional restriction varied among the 5 winters, elevated UN:C values suggested a level of deprivation not supportive of population stability or growth. Climate change, reflected by the heat stress index for moose, and variation in winter conditions, as indexed by the winter severity index (WSI), were not related to nutritional restriction of moose. For the 5 winters, we also have documented that the level of severe nutritional restriction is inversely related ($r = –0.86$) to variation of *natural* winter survival of global positioning system (GPS)-collared adult moose. While these relationships do not substantiate cause-and-effect, presently it provides the best preliminary empirical evidence that inadequate winter nutrition at the population level is intricately related to the declining trajectory of moose numbers in northeastern Minnesota.

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INTRODUCTION

Declines in regional populations of moose (*Alces alces*) along the southern periphery of their global range have been common in recent decades (Timmerman and Rodgers 2017). In northeastern Minnesota the estimated 2017 population (3,710 moose) is 58% less than in 2006 (8,840 moose, DelGiudice 2017), exhibiting a trajectory similar to that documented previously for moose in northwestern Minnesota, where the population decreased from ~4,000 in the mid-1980s to <100 moose in 2007 (Murray et al. 2006). Furthermore, mean annual mortality rates of collared adult moose were similarly high (21%) in the northwest and northeast during the declines (Murray et al. 2006; Lenarz et al. 2009; R. A. Moen, unpublished data). In northwestern Minnesota, malnutrition and pathogens were identified as important factors influencing the population's decreasing trajectory (Murray et al. 2006). In northeastern Minnesota a recent (2013–2016) aggressive study of global positioning system (GPS)-collared, adult moose reported a mean annual mortality rate of 15.3%, with health-related factors (e.g., parasites, disease) accounting for about two-thirds of the deaths, wolf (*Canis lupus*) predation for one-third, and complex interactions between the 2 categories well-documented (Carstensen et al. 2017). In the earlier studies, climate change (i.e., warming temperatures) was implicated in both population declines (Murray et al. 2006; Lenarz et al. 2009, 2010).

These temperature-survival relationships are complex, and indicate that climate change can directly and indirectly impact ungulate populations (Bastille-Rousseau et al. 2015, Davis et al. 2016, Street et al. 2016). Moose are particularly well-adapted to cold climates, but temperatures that exceed "heat stress" thresholds of 14 to 24^{o}C during summer and –5^{o}C during winter may increase metabolic rates, induce energy deficits, and hasten deterioration of body condition (Renecker and Hudson 1986, 1990; Broders et al. 2012; McCann et al. 2013). These thresholds may be influenced by exposure to solar radiation and wind (Renecker and Hudson 1990, McCann et al. 2013). Nutritional and health status (e.g., disease, parasites), behavioral responses (e.g., altering movement, foraging, and bedding patterns), and quality of available habitat have the potential to affect the animal's ability to mitigate negative impacts from heat stress (Van Beest et al. 2012, Street et al. 2016).

Energy balance is central to animal fitness, which is critical to survival and reproduction, the 2 drivers of population performance (Robbins 1993). The natural "nutritional bottleneck" of winter typically imposes the greatest challenge to the supply side of energy budgets of moose and other northern ungulates (Mautz 1978, Schwartz and Renecker 2007). Gestation at this time increases energetic and nutritional demands, particularly during late-winter and early-spring (Robbins 1993). Although moose are generally well-adapted to this seasonal nutritional deprivation, elevated ambient temperatures exceeding heat stress thresholds, coupled with the influence of other compromising extrinsic factors (e.g., pathogens, poor quality forage and low availability of thermal cover, densities of conspecifics or other nutritionally competing species) can exacerbate energy deficits and associated consequences for adult and juvenile survival, subsequent reproductive success, and population dynamics (Robbins 1993; DelGiudice al. 1997, 2001).

Winter nutritional restriction of moose and other northern ungulates can be physiologically assessed at the population level by serial collection and chemical analysis of fresh urine voided in snow (snow-urine; DelGiudice et al. 1988, 1997, 2001; Moen and DelGiudice 1997, Ditchkoff and Servello 2002). Urea nitrogen (interpreted as a ratio to creatinine, UN:C), the end-product of protein metabolism, is one of many chemistries investigated for its value as a physiological metric of the severity of nutritional restriction (DelGiudice et al. 1991*a*,*b*, 1994). In healthy moose, urinary UN:C values decrease (N conservation) in response to diminishing intake of crude protein and digestible energy, but as dietary restriction and negative energy balance become more severe and fat reserves are depleted, ratios increase to notably elevated values in response to accelerated net catabolism of endogenous protein. Snow-urine UN:C ratios exhibited differential effects of a winter tick (*Dermacentor albipictus*) epizootic and habitat differences on the severity of nutritional restriction of moose on Isle Royale, and were strongly related to dynamics of the population, including a pronounced decline and recovery to historically high numbers (DelGiudice et al. 1997).

OBJECTIVES

1. To determine how nutritional restriction varies annually and as winters progress
2. To examine potential relationships between the severity of nutritional restriction and the winter heat stress index (HSI) for moose, seasonal survival rates of GPS-collared adult moose, and annual population estimates of moose

We hypothesized that increasing winter ambient temperatures, exceeding the HSI threshold, are contributing to the severity of nutritional restriction and energy deficit of moose, decreases in survival by various proximate factors, and diminishing performance of the population. Findings will set the stage for additional work assessing nutritional relationships of moose to variations in habitat and other factors.

STUDY AREA

We assessed winter nutritional restriction of moose within a 6,068-km^2 study area located between 47°06'N and 47°58'N latitude and 90°04'W and 92°17'W longitude in northeastern Minnesota (Figure 1). Including bogs, swamps, lakes, and streams; lowland stands of northern white cedar (*Thuja occidentalis*), black spruce (*Picea mariana*), and tamarack (*Larix laricina*); and upland balsam fir (*Abies balsamea*), jack pine (*Pinus banksiana*), white pine (*P. strobus*), and red pine (*P. resinosa*), this region has been classified as Northern Superior Upland (Minnesota Department of Natural Resources [MNDNR] 2015). Trembling aspen (*Populus tremuloides*), white birch (*Betula papyrifera*), and conifers are frequently intermixed.

Wolves (*Canis lupus*) and American black bears (*Ursus americanus*) are predators of moose (Fritts and Mech 1981, Severud et al. 2015) with recent densities estimated at 3.4 wolves and 23 bears/100 km^2 (Erb and Sampson 2013, Garshelis and Noyce 2015). White-tailed deer (*Odocoileus virginianus*) are managed at pre-fawning densities of <4 deer/km^2, and are the primary prey of wolves in most of northern Minnesota (Nelson and Mech 1986, DelGiudice et al. 2002). The MNDNR assesses winter severity (1 Nov–31 May) by a winter severity index (WSI), calculated by accumulating 1 point for each day with a temperature $\leq 0^{o}$F (–17.8^{o}C, temperature-day) and 1 point for each day with snow depth $\geq$15 inches (38.1 cm, snow-day), for a potential total of 2 points per day. Maximum WSI values varied markedly across moose range, 35–160, 184–245, 54–152, 31–142, and 50−159 for winters 2012–13 to 2016–17, respectively (Minnesota State Climatology Office 2017). Mean daily minimum and maximum temperatures varied markedly during November–April from 2012–13 to 2016–17 at Ely, Minnesota (Midwestern Regional Climate Center 2017; Figure 2). The heat stress index (HSI_{Min} and HSI_{Max}) for moose during January and the "cold season" (Nov–Mar) was calculated by daily accumulation of degrees Celsius >–5^{o}C for the maximum and minimum ambient temperatures, respectively (Renecker and Hudson 1986).

METHODS

We collected fresh snow-urine specimens of moose during 6 January−28 March 2013− 2017. We conducted snow-urine sampling according to a random design. Our field team drove (by truck or snowmobile) a route of approximately 201 km (125 miles) to distribute the sampling throughout the study area (Figure 1). Field technicians were not restricted to this route, rather they could deviate, particularly on foot, as dictated by the presence of fresh moose sign (e.g., tracks, urine specimens, pellets). Each field team used handheld GPS units loaded with several land coverages (R. G. Wright, Minnesota Information Technology @ Minnesota Department of Natural Resources, Section of Wildlife) and a Superior National Forest map (U. S. Forest Service) to navigate in the field.

Generally, sampling was conducted within 7 days of a fresh snowfall, most often within 2–4 days, so that we could associate urine chemistry data and nutritional assessments with specific temporal intervals. Upon observing fresh moose sign, technicians tracked the individual(s) on foot as necessary until they found a fresh snow-urine specimen. The objective for the collections was to sample primarily adult (>1 year old) moose (indicated by track and bed size). This was not particularly challenging, because by this time of year calves comprised only 13–17% of the population (DelGiudice 2017). We focused primarily on the adult age class to facilitate optimum comparability of data.

Specimens were collected and handled as described by DelGiudice et al. (1991*a*, 1997). A GPS waypoint was recorded for each snow-urine specimen collected. Date of the most recent snowfall and comments describing the presence of moose or other sign in the area also were recorded.

Snow-urine specimens were analyzed for UN and C (mg/dL for both) by a Roche Cobas Mira auto-analyzer (Roche Diagnostics Systems, Inc., Montclair, NJ) in the Forest Wildlife Populations and Research Group’s laboratory. We used 0.1 and 3.0 mg/dL as reliable thresholds for accurate measuring of C and UN, respectively, for our auto-analyzer; samples with values below these thresholds were excluded (C. Humpal, MNDNR, personal communication). Data were compared as UN:C ratios to correct for differences in hydration, body size, and dilution by snow (DelGiudice 1995, DelGiudice et al. 1988).

Winter (Jan–Mar) was divided into 6, approximately 2-week sampling intervals (1–15 Jan, 16–31 Jan, 1–15 Feb, 16–28 Feb, 1–15 Mar, and 16–31 Mar). Sample sizes for the snow-urine collections varied by interval due to variability of weather (i.e., snow conditions), equipment availability, logistical challenges, and ease of finding samples. Most of the UN:C data are reported by the entire winter or by sampling interval as means (± standard error). Additionally, based on past work, urinary UN:C values were assigned to 1 of 3 levels of nutritional restriction: moderate or “normal,” <3.0 mg:mg; moderately severe, 3.0–3.4 mg:mg; and severe, ≥3.5 mg:mg (DelGiudice et al. 1997, 2001, 2010). We report the percentage of samples with UN:C values falling within each of these categories. We examined relationships between proportions of snow-urine specimens with UN:C values indicative of severe nutritional restriction (≥3.5 mg:mg) and populations estimates, seasonal survival, and HSI by simple linear regression analyses in Excel (Version 14.0.7153.5000, Microsoft Corporation 2010).

RESULTS AND DISCUSSION

During January–March 2013–2017, a total of 944 urine specimens from moose were collected to assess nutritional status at the population level. Specifically, annual totals of 123, 307, 165, 189, and 160 sufficiently concentrated moose snow-urine samples, respectively, were collected during 5–6, 2-week sampling intervals using our designated routes. The greater number of samples collected during 2014 was largely due to the early and prolonged deep snow cover.

Overall, mean UN:C ratios were 3.7, 2.9, 2.9, 3.5, and 3.7 mg:mg for winters 2013 to 2017, respectively (Figure 3). The mean population UN:C ratio for entire winters 2013, 2016, and 2017 were above the threshold indicative of severe nutritional restriction (i.e., a starvation diet) and accelerated body protein catabolism. The elevated mean UN:C of 2016 and 2017 were influenced largely by several collected samples that exhibited very high UN:C ratios indicative of a moribund condition (≥22.0 mg:mg), given that the proportion of samples in the lowest UN:C category was greatest that year (Figure 4). During 2014 and 2015 the population means were just below the defined moderately severe interval. Additionally, indicative of the unique severity of nutritional restriction in 2013, nearly one-third of all samples collected yielded UN:C ratios ≥3.5 mg:mg (Figure 4). The corresponding percentages of winters 2014 to 2017 were notably less than in 2013.

Mean urinary UN:C ratios by 2-week interval of winter 2013 indicated that nutritional restriction was normal or moderate during late-January, but became severe throughout February and early-March, and was still assessed as moderately severe in late-March (Figure 5). As severe nutritional restriction of moose progresses with winter, those animals may be under-sampled as some eventually die, and those still alive urinate less, which is a physiological mechanism to conserve water and electrolytes. Percentage of samples with urinary UN:C ratios indicative of severe nutritional restriction peaked (73.3%) in early-February and remained relatively high through late-March (36%) during 2013 (Figure 6). Such elevated values have been associated with long-term fasting in controlled nutrition studies of captive white-tailed deer and starvation of free-ranging elk (*Cervus elaphus*), bison (*Bison bison*), and moose (DelGiudice et al. 1991*a*, 1994, 1997, 2001). The percentage of snow-urine specimens in 2013 with UN:C ratios indicative of moderately severe to severe nutritional restriction throughout the winter was 45.5% (Figure 4).

During 2014 mean urinary UN:C ratios in all 2-week intervals, except early February, remained just below the moderately severe category (Figure 5), and the percentage of samples with ratios indicative of severe nutritional restriction gradually decreased as this winter progressed (Figure 6), either due to an easing of conditions restricting access to forage or because severely stressed individuals were being under-sampled, which may be most plausible as previously explained. Adverse effects of the late, but prolonged conditions of winter 2013, including warm temperatures, may have contributed to the high spring-summer calf loss and absence of the need for dams to lactate (Severud et al. 2015). This also may have allowed the surviving animals to rebound nutritionally more quickly and to fare better during winter 2014. This would not be unlike the documented effects on the nutritional status and survival of northern Minnesota deer during the consecutive severe winters of 1996 and 1997 (DelGiudice et al. 2006; G. D. DelGiudice, unpublished data). Overall in winter 2014, UN:C values of 64% of the collected snow-urine samples classified nutritional restriction as moderate (normal), whereas 36% reflected moderately severe to severe restriction, which was less than in 2013 (Figure 4). Similar to winter 2014, severe nutritional restriction of moose was not as prevalent in 2015 as in 2013, but it was up slightly compared to 2014 (Figure 4). However, a higher percentage of moose appeared to be experiencing moderate or normal restriction and a smaller percentage moderately severe than in 2013 and 2014 (Figure 4). Rapidly diminishing snow cover prevented collection of snow-urine samples or assessments during the last 2 weeks of March 2015, certainly a positive factor relative to moose nutrition at that time. Unexpectedly, both 2016 and 2017 had the greatest percentage of samples with urinary UN:C ratios indicative of moderate nutritional restriction (70.4% and 70.6%, respectively) and the smallest percentage indicative of severe nutritional restriction (Figure 4), despite having the highest HSI values calculated with daily maximum (958 and 833) or minimum (220 and 194) ambient temperatures.

According to maximum WSI values, winter 2014 was the most severe of the 5 in northeastern Minnesota moose range, followed by 2013, 2017, 2015, and 2016. Although the WSI numbers have value for annual comparisons of winter conditions, this WSI formula has far greater relevance to the size and energetics of white-tailed deer than for the much larger moose, which are not hindered as much by deep snow (DelGiudice et al. 2002, 2006; Schwartz and Renecker 2007). Furthermore, while the accumulation of snow-days and temperature-days has proven significant relative to the survival of white-tailed deer (DelGiudice et al. 2002), actual snow depth, its temporal occurrence, and duration may be of equal or greater importance for moose and deer (Telfer and Kelsall 1984, DelGiudice 1998, DelGiudice et al. 2002, Schwartz and Renecker 2007). During 2013, conditions became severe during mid- to late-winter; consequently, a high number of snow-days did not accumulate, but the season was prolonged. The severe nutritional restriction of moose in 2013 was most similar to that which occurred in moose during several winters (1988–1990) on Isle Royale associated with severe winter tick infestations and steep population decline (DelGiudice et al. 1997). Abundant evidence from the field in the MNDNR's ongoing studies similarly indicated that the winter tick infestation of moose in northeastern Minnesota was notably more severe during winter 2013 than in any of those that followed through 2017 (Carstensen et al. 2014; M. Carstensen, MNDNR, personal communication).

Perhaps the ultimate value to management of assessments of nutritional status of free-ranging animals comes when the findings can be related to the performance and dynamics of the population and other ecological factors challenging that performance (DelGiudice et al. 1997, Cook et al. 2004). Our population-level nutritional assessments continue to closely track (r^2 = 0.75) population estimates of moose from the annual aerial survey (Figure 7). What is most clear is that although restriction varies among the 5 winters, elevated UN:C values suggest a level of nutritional deprivation not supportive of positive population performance or growth. During 2013 to 2015, warming winter temperatures appeared to be having the most pronounced influence on the nutritional status of moose. As the January and winter HSI_{Max} values increased, the incidence of severe nutritional restriction of moose increased ($r^2 \geq 0.93$, DelGiudice and Severud 2017), which we believed may have led to many of these animals becoming more vulnerable to various health-related causes of mortality and predation (Carstensen et al. 2015). Similar relationships were noted between winter nutritional restriction, winter tick epizootics, and decreasing moose numbers on Isle Royale (DelGiudice et al. 1997). But, something unexpected occurred when data from winters 2016 and 2017 (the warmest winters since 2013 in northeastern Minnesota) were incorporated into the analyses. The strong relationships between HSI_{Max} of January and winters 2013–2015 and severe nutritional restriction collapsed; the incidence of severe nutritional restriction of moose was still noteworthy in 2016 and 2017, but was the lowest of the 5 winters. However, the incidence of severe nutritional restriction at the population level remained inversely related to variation of *natural* winter ($r = -0.86$) and winter-summer ($r = -0.88$) survival rates of GPS-collared adult moose (Figure 8). Importantly, because these latter relationships are consistent with our association of severe nutritional restriction with the population estimates, it suggests that the current study cohort of GPS-collared moose is indeed representative of the free-ranging population in northeastern Minnesota. While these aforementioned relationships do not substantiate cause-and-effect, a preponderance of the empirical evidence is suggesting that winter nutritional restriction is a critical factor influencing the performance and dynamics of the northeastern moose population. Clearly, there is still much to understand about these relationships. New to this understanding are the effects of variation in severe nutritional restriction and the loss of breeding females on annual calf production (Figure 9).

In addition to the multi-year occurrence of severe nutritional restriction of moose, preliminary analyses reveal a vast spatial distribution throughout moose range of collected snow-urine samples with UN:C ratios indicative of severe nutritional deprivation (Figure 10). The wide temporal and spatial distributions of severe nutritional restriction suggest that habitat deficiencies at the landscape scale may constitute a primary contributing factor. We continue to apply significant efforts into investigating the habitat-nutrition relationships, but habitat deficiencies related to forage availability and quality, vegetative species composition, or less-than-optimum arrangements of forage openings and forest stands affording seasonal thermal cover remain unclear. Data from future winter nutritional assessments are required to provide additional support for our conclusions or to refute them. But the current data set, in combination with data from other ongoing habitat and nutritional studies, should provide a basis for formulating management recommendations that may be implemented and evaluated in the near future.

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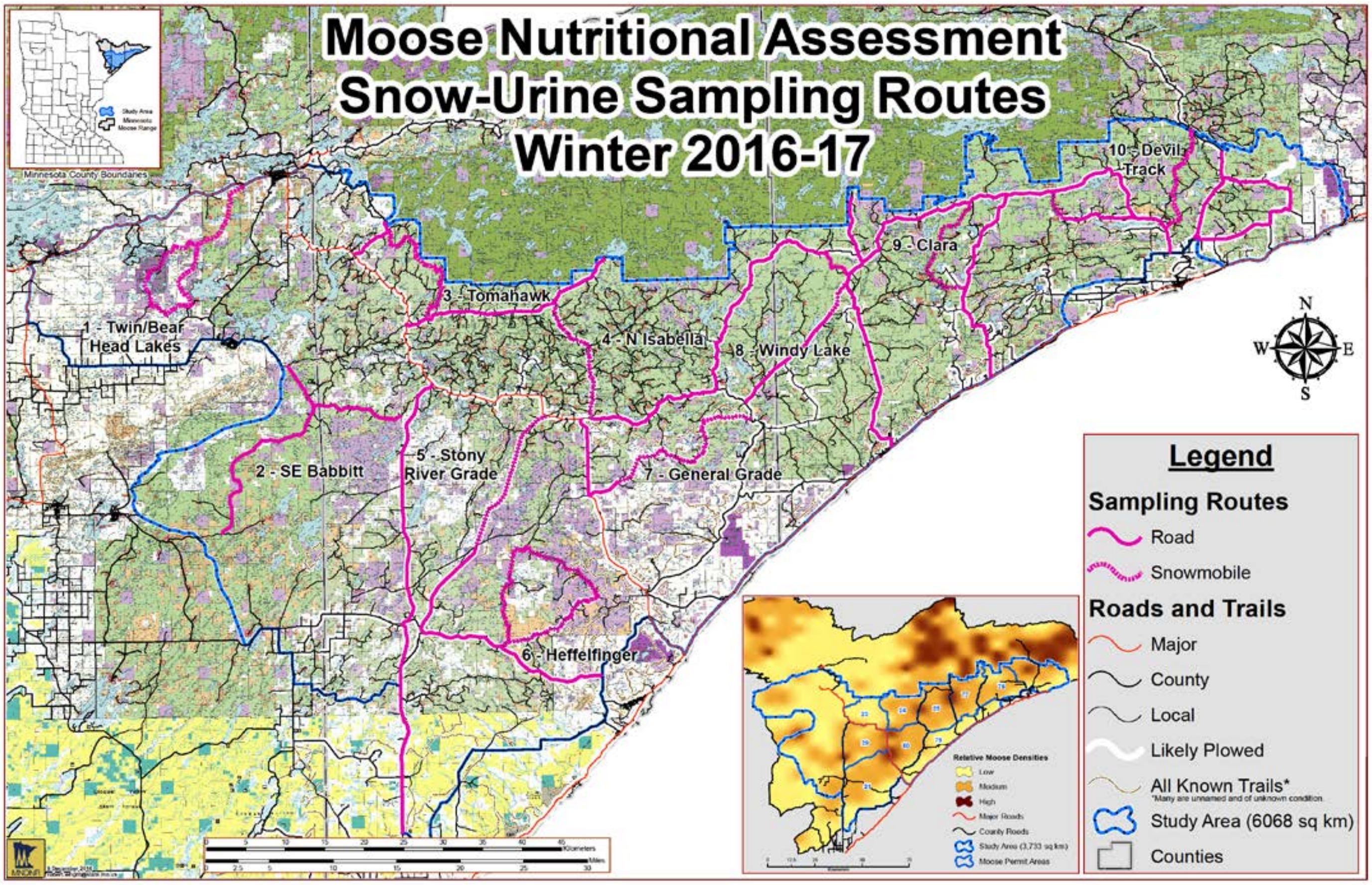
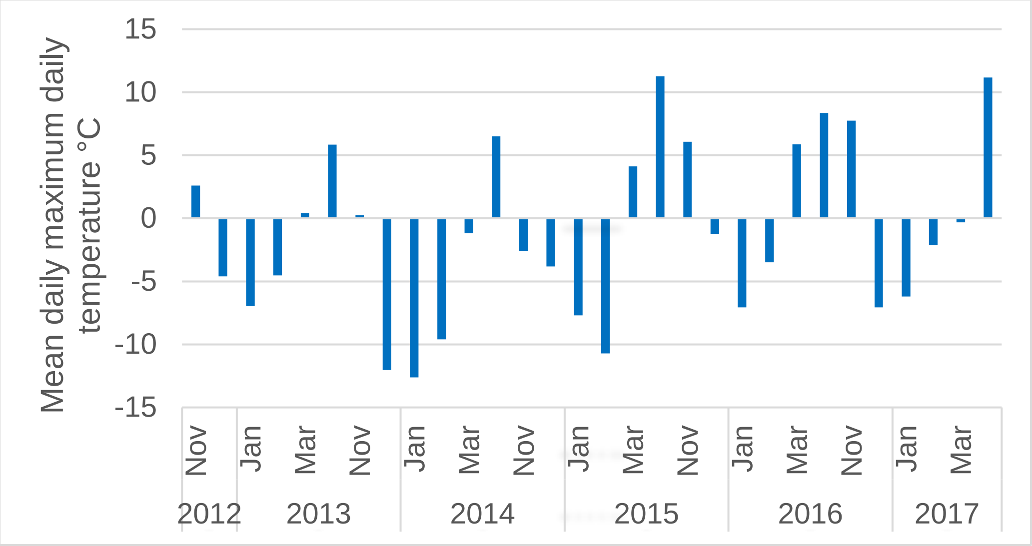


Figure 1. Map depicting the moose study area in northeastern Minnesota and the routes (i.e., roads and snowmobile trails in purple) used to distribute the sampling of fresh moose urine in snow (snow-urine) for nutritional assessments throughout the area, January–March 2013–2017.



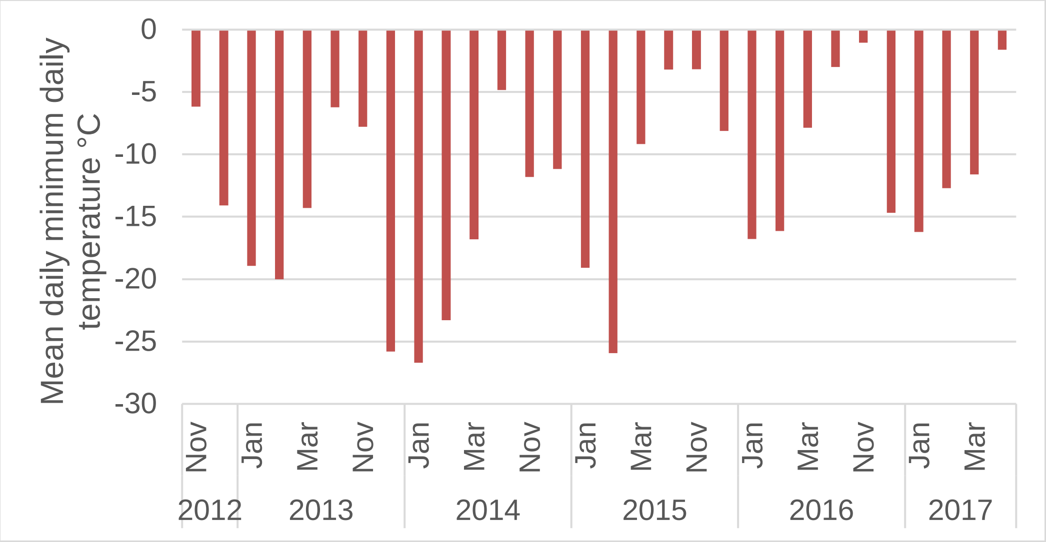


Figure 2. Mean daily maximum (top) and minimum (bottom) ambient temperatures, Ely, Minnesota, November–April 2012–2017 (Midwestern Regional Climate Center 2017).

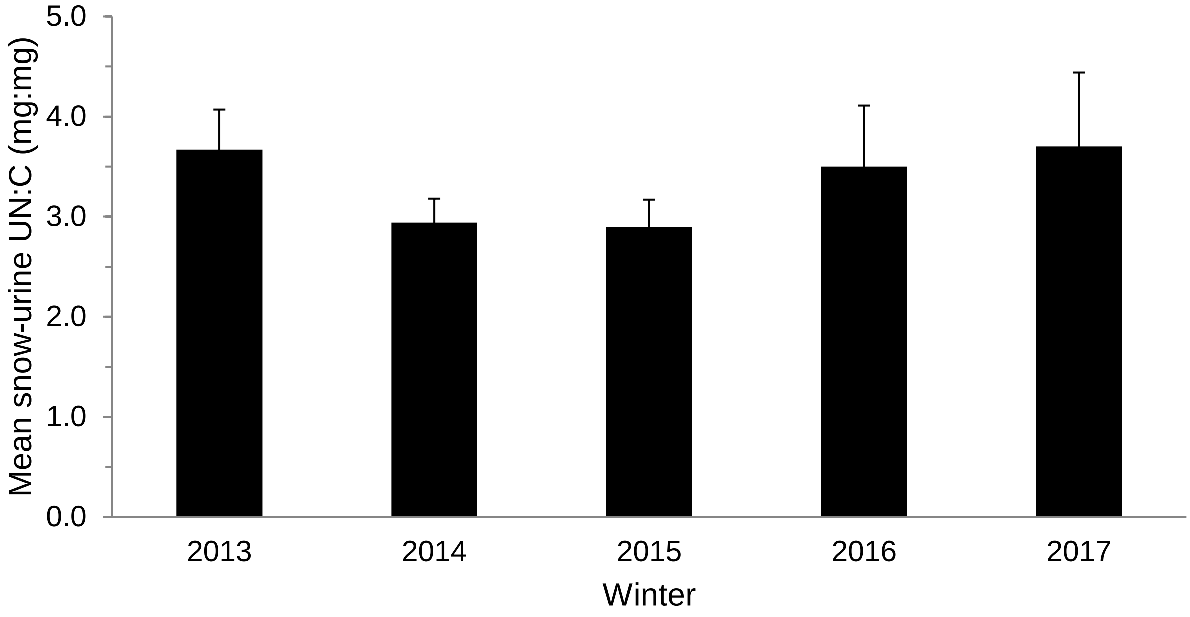


Figure 3. Overall mean (+ SE) urea nitrogen:creatinine (UN:C) ratios of samples of fresh urine voided in snow (snow-urine) by moose and serially collected for assessments of nutritional restriction throughout northeastern Minnesota, January–March 2013–2017.

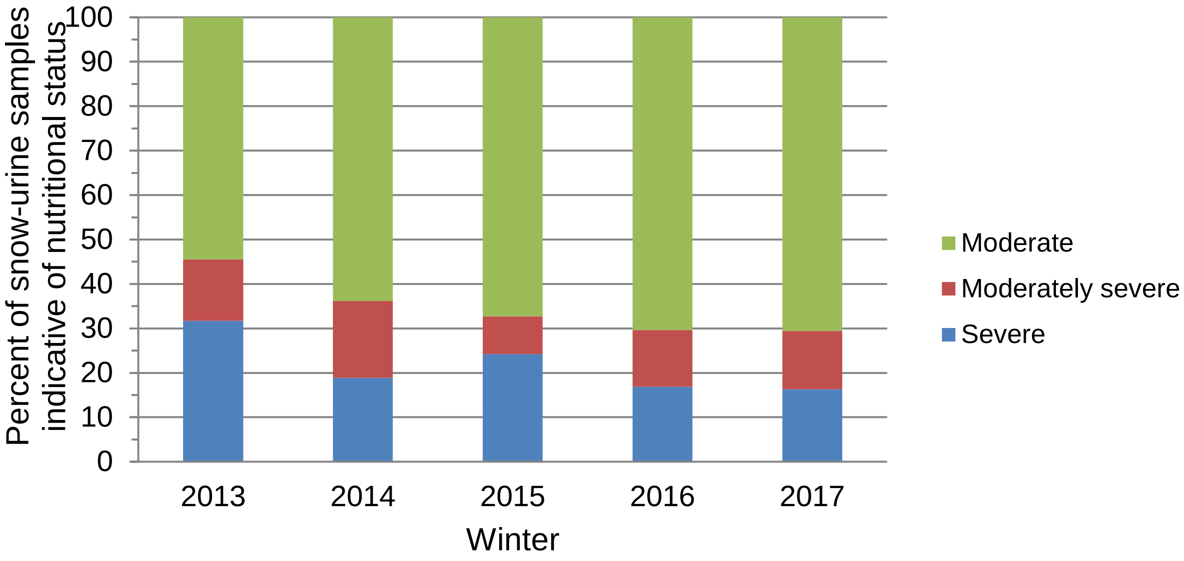


Figure 4. Overall percent of serially collected moose urine samples voided in snow (snow-urine) with urea nitrogen:creatinine (UN:C) ratios indicative of moderate/normal (UN:C <3.0 mg:mg), moderately severe (UN:C = 3.0–3.4 mg:mg), and severe nutritional restriction (UN:C ≥3.5 mg:mg) throughout northeastern Minnesota, January–March 2013–2017.

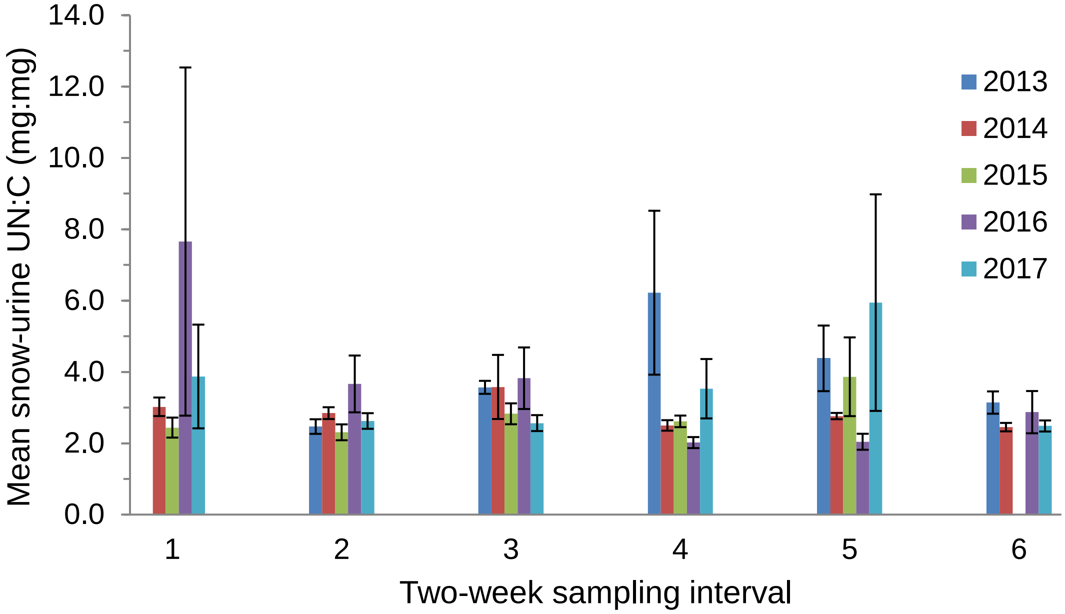


Figure 5. Mean (± SE) urea nitrogen:creatinine (UN:C) ratios of samples of fresh urine voided in snow (snow-urine) by moose and collected during 2-week sampling intervals for assessments of nutritional restriction throughout northeastern Minnesota, January–March 2013–2017.

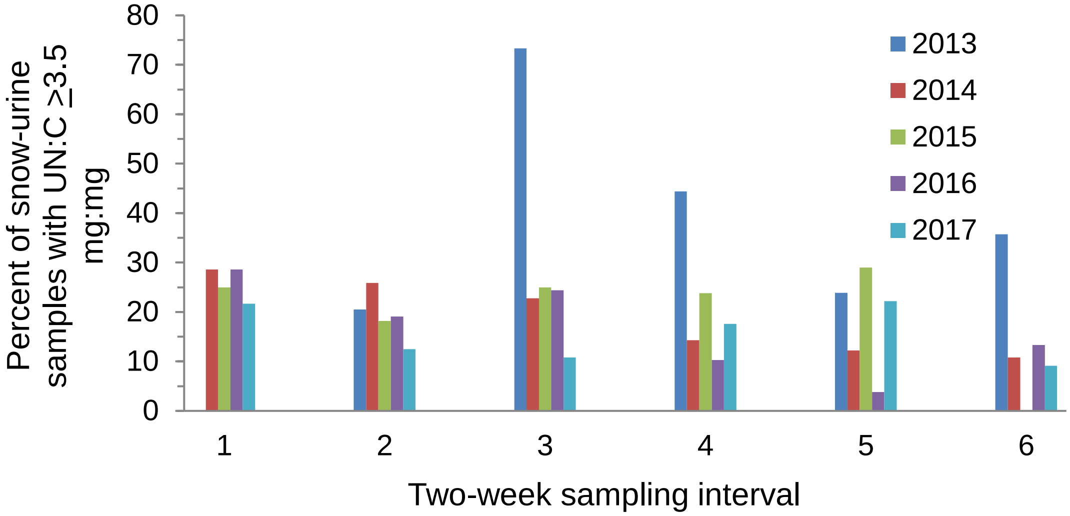


Figure 6. Percent of fresh urine samples voided in snow (snow-urine) by moose and collected during 2-week intervals with urea nitrogen:creatinine (UN:C) ratios indicative of severe nutritional restriction (UN:C ≥3.5 mg:mg) throughout northeastern Minnesota, January–March 2013–2017.

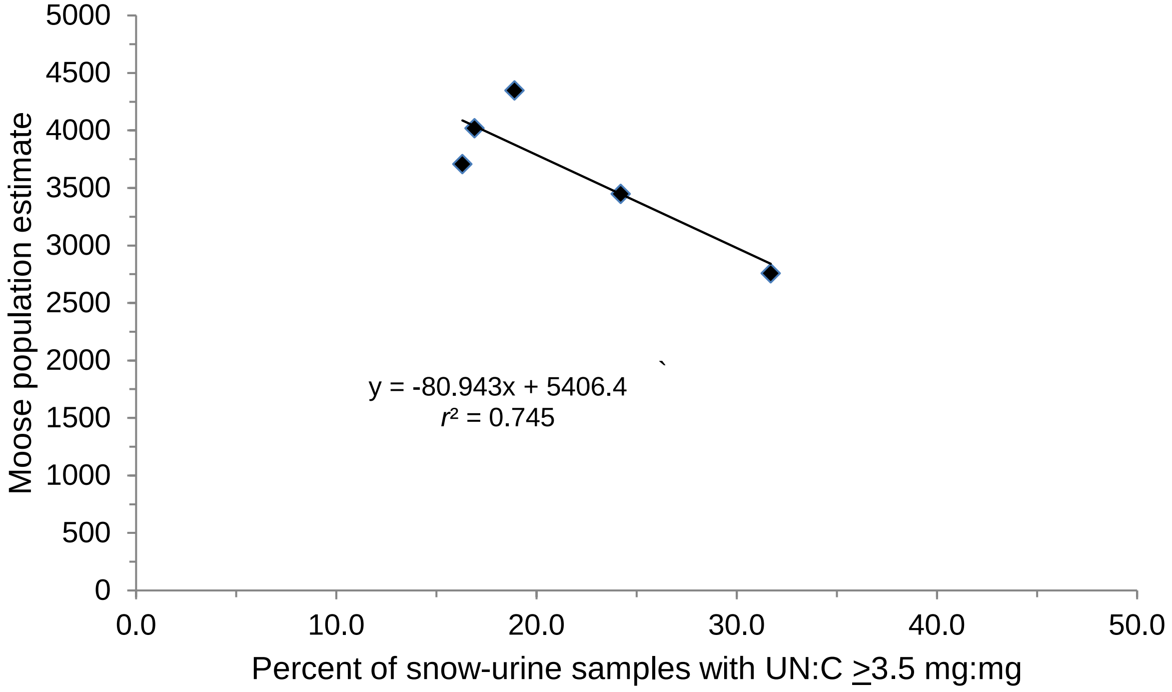


Figure 7. Relationship of the incidence of severe winter nutritional restriction of moose, indicated by the percentage of collected samples of urine in snow (snow-urine) with urea nitrogen:creatinine (UN:C) ratios ≥3.5 mg:mg, to annual population estimates of moose in northeastern Minnesota (estimates from DelGiudice 2017), January–March 2013–2017.

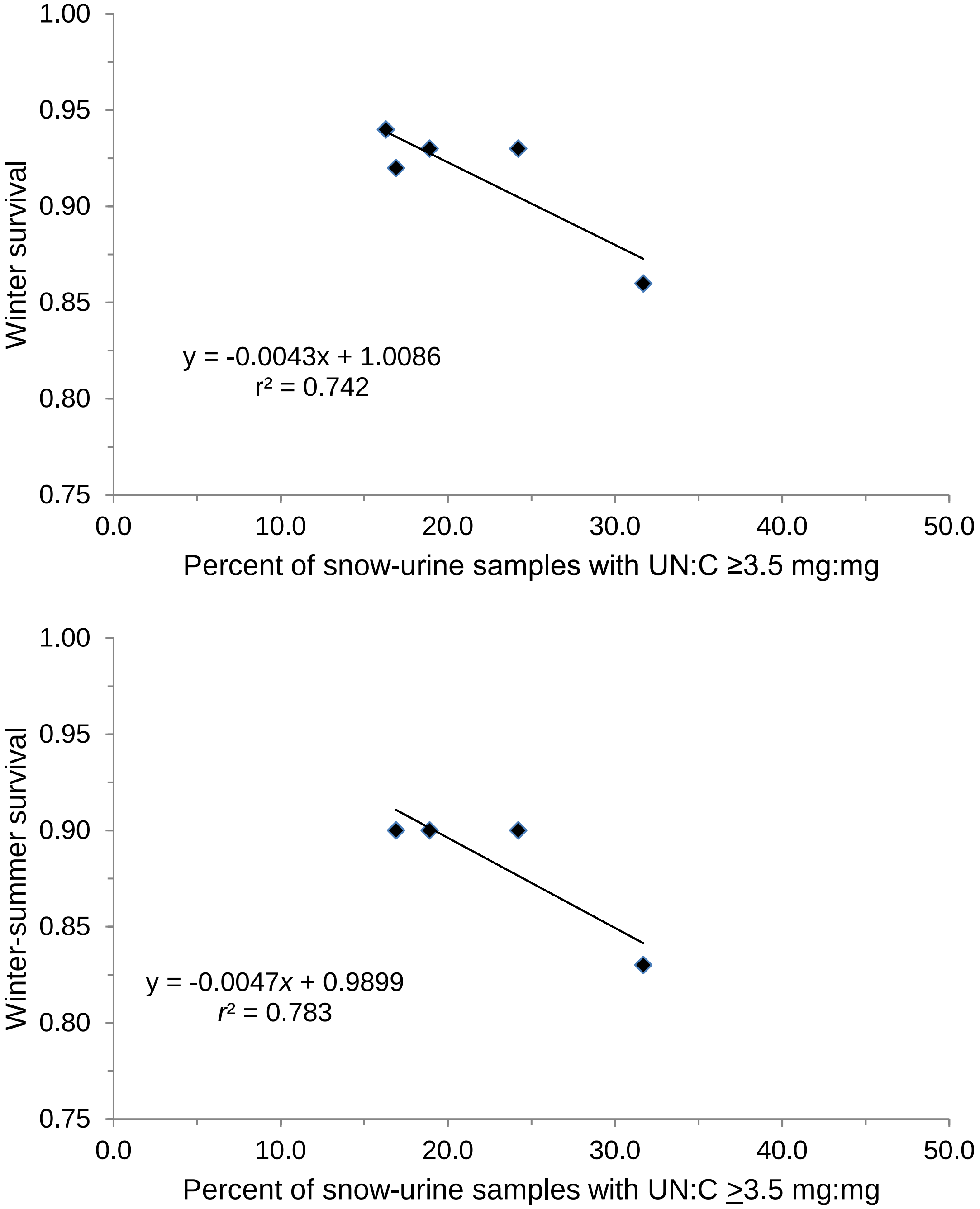


Figure 8. Relationships of the incidence of severe winter nutritional restriction of moose at the population level, indicated by the percentage of collected samples of urine in snow (snow-urine) with urea nitrogen:creatinine (UN:C) ratios ≥3.5 mg:mg, to winter (top, 1 Nov–31 May 2013−2017) and winter-to-summer (bottom, 1 Nov–31 Aug 2013−2016) survival of GPS-collared adult moose in northeastern Minnesota.

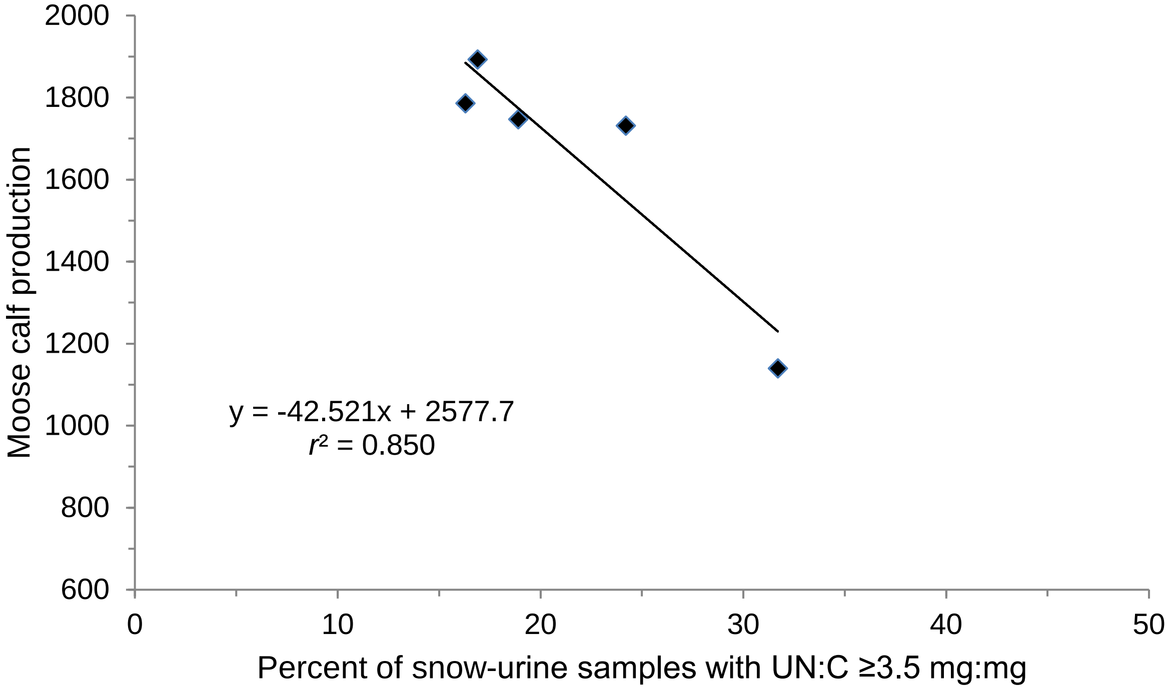


Figure 9. Relationships of the incidence of severe winter nutritional restriction of moose, indicated by the percentage of collected samples of urine in snow (snow-urine) with urea nitrogen:creatinine (UN:C) ratios ≥3.5 mg:mg, to annual calf production, northeastern Minnesota, 2013–2017.

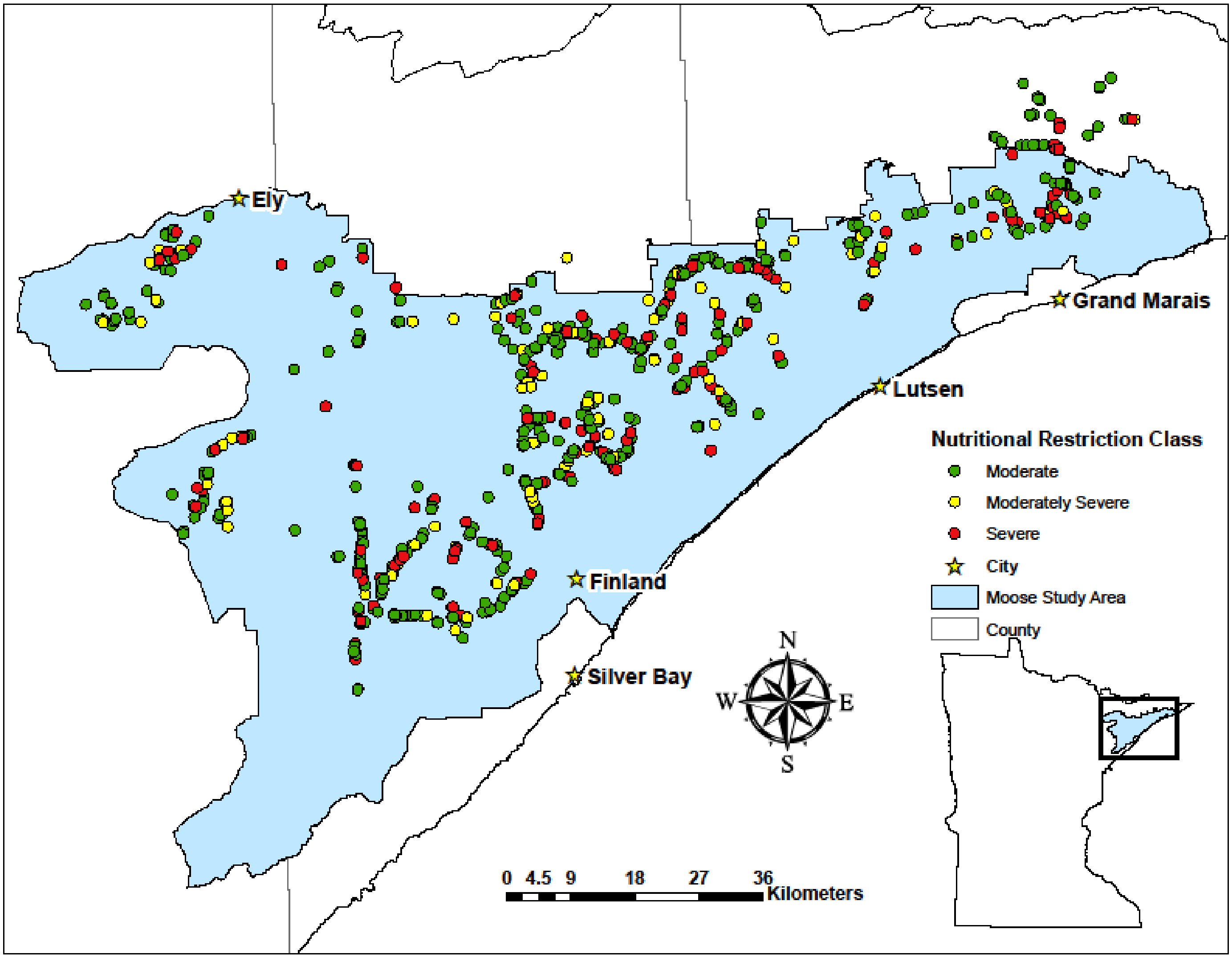


Figure 10. Spatial distribution of fresh urine samples of moose, serially collected for chemical analysis to assess the severity of winter nutritional restriction. Urinary urea nitrogen:creatinine (UN:C) ratios of <3.0, 3.0–3.4, and ≥3.5 mg:mg are indicative of moderate/normal (green), moderately severe (yellow), and severe (red) nutritional restriction, northeastern Minnesota, 6 January–28 March 2013−2017.

NEONICOTINOIDS ON THE LANDSCAPE: EVALUATING AVIAN EXPOSURE TO TREATED SEEDS IN AGRICULTURAL LANDSCAPES

Charlotte Roy, Da Chen[1], Julia Ponder[2], Mark Jankowski[3]

SUMMARY OF FINDINGS

Neonicotinoid pesticides (e.g., imidacloprid, thiamethoxam, thiacloprid, clothianidin) are commonly applied to agricultural seeds (e.g., corn, soybean, wheat, sunflower), and are known to cause lethal and sub-lethal effects in birds. Neonicotinoid-treated seeds could be available to wildlife through spillage or exposure to treated seeds near or at the soil surface after planting (de Leeuw et al. 1995, Pascual et al. 1999, Lopez-Antia et al. 2016). We are examining sub-lethal exposure of wild birds to these pesticides in agricultural landscapes of Minnesota. We are quantifying seed availability at the soil surface in recently planted fields and the rate of seed spills during planting, as well as documenting birds eating treated seeds through field studies with trail cameras and harvested birds. Thus far, we have documented ring-necked pheasants (*Phasianus colchicus*), Canada geese (*Branta canadensis*), American crows (*Corvus brachyrhynchos*), various species of sparrows (Emberizidae) and blackbirds (Icteridae), as well as white-tailed deer (*Odocoileus virginianus*), rodents, leporids, and raccoons (*Procyon lotor*) consuming seeds. In 2016, we documented 212 seed spills in 38 townships during planting but missed the peak of planting in many of the townships we surveyed. We documented exposed seeds at the soil surface in plots at 25% of 48 fields sampled after planting in 2016. Field work is ongoing for 2017. We are still conducting analyses to determine the length of time that neonicotinoids persist on seeds exposed at the soil surface, and whether the seeds are consumed before the chemicals have degraded.

We also conducted laboratory experiments to try to identify non-lethal sampling methods that could lead to methods for measurement of individual and population-level exposure, including residues in excreta and blood. Residues were highest (geometric mean) in the brain, followed by liver, spleen, muscle, blood, kidney, then feces in birds dosed in the lab. Residues were detected in 90.9% of domestic chicken fecal samples collected in the lab, the highest detection frequency of all tissues tested. Forty-one of 46 (89%) liver samples collected from hunter-harvested sharp-tailed grouse (*Tympanuchus phasianellus*) and 18 of 27 (67%) hunter-harvested greater prairie-chickens (*Tympanuchus cupido*) have been analyzed and contained detectable concentrations of at least 1 neonicotinoid. Similarly, 22 of 34 (65%) fresh prairie-chicken fecal pellets and 47 of 56 (84%) sharp-tailed grouse pellets collected from leks have been analyzed and had detectable concentrations of at least 1 neonicotinoid. Data collection will continue through fall 2017.

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INTRODUCTION

Neonicotinoids are the most widely used pesticides worldwide (Mineau and Palmer 2013), comprising 25% of the global agricultural chemical market. Their action is highly specific to invertebrates, with comparatively low toxicities for vertebrates compared to pesticide options predating the early 1990s (Tomizawa and Casida 2005, Jeschke et al. 2011). This high specificity contributed to their widespread and rapid adoption, beginning in 1994 with the registration of imidacloprid in the United States.

Recently, neonicotinoids have received a lot of attention because of their potential toxicity to bees and other pollinators, and their possible role in colony collapse disorder. Several neonicotinoid treatments were banned or placed under a moratorium in Europe in 2013, and neonicotinoids are currently under registration review by the Environmental Protection Agency (EPA) in the United States. The Minnesota Department of Agriculture (MDA) is currently reporting a process and criteria for review of neonicotinoid use with an emphasis on pollinators (MDA 2014). However, recent concern has not been limited to pollinators; the American Bird Conservancy called for research on the effects of neonicotinoids on birds and a ban on neonicotinoid seed treatments (Mineau and Palmer 2013). Evidence is accumulating that vertebrates are also adversely affected by these pesticides (see reviews in Mineau and Palmer 2013, Gibbons et al. 2014). MDA (2014) acknowledged that, “Although neonicotinoids are less toxic to vertebrates than to arthropods, direct consumption of neonicotinoid treated seeds may expose birds and other taxa to acute or chronic doses.”

The most likely route of exposure to large doses of neonicotinoids for birds is ingestion of treated seeds (Goulson 2013, Gibbons et al. 2014), although numerous other mechanisms exist (e.g., soil, trophic transfer; SERA 2005, Douglas et al. 2015). Ingestion of a small number of neonicotinoid-treated seeds is lethal to birds; for example, a single treated corn kernel can kill a blue-jay sized bird (see reviews in Mineau and Palmer 2013, Gibbons et al. 2014). However, toxicity generally varies by chemical and species, given differences in physiological make-up such as size and digestive processes. Lethal impacts are rapid and difficult to detect in the wild although a few pesticide poisoning incidents have been detected (Greig-Smith 1987, Fletcher et al. 1995, Berny et al. 1999, de Snoo et al. 1999). Sub-lethal exposure might be easier to detect in the wild. Sub-lethal effects in birds in the lab include hyporeactivity, lack of coordination, wing drop, immobility, eggshell thinning, reduced egg hatching rate, impaired testicular function, immune suppression, and low weight in chicks (Cox 2001, Lopez-Antia et al. 2013 and 2015, Tokumoto et al. 2013, Mineau and Palmer 2013). Reproduction can be affected by consumption of just 1/10th of a treated corn seed per day during egg-laying (Mineau and Palmer 2013).

Thirty bird species were observed picking up treated seeds from cereal fields in Spain and 3.1% of partridge gut contents collected by hunters tested positive for imidacloprid after planting of winter cereal crops (Lopez-Antia et al. 2016). Dead and poisoned partridges have been found in agricultural fields in France following use of imidacloprid-treated seed (Berny et al. 1999). The EPA estimated that ~1% of seeds remain accessible to granivores after planting (as reported by Goulson 2013, Lopez-Antia et al. 2015). Unfortunately, neonicotinoid use of “treated articles,” such as seed, is not currently tracked by the government due to the exemption in 40CFR §152.25(a). Yet, almost all corn planted in the Midwest has been treated with these pesticides (Stokstad 2013), as well as most soybean, wheat, and sunflower seeds, and they are widely used with other application methods for other crop types.

Studies of neonicotinoid effects on vertebrates are overwhelmingly laboratory-based (91% of studies), which limits our ability to interpret the significance of findings in more natural settings (Gibbons et al. 2014). Higher densities of exposed seeds result in greater attraction of birds to fields (Murton et al. 1963, Feare et al. 1974). Bednarska et al. (2013) identified a need for feeding rate information in the field to allow extrapolation of lab data to the field. Lopez-Antia et al. (2013) pointed to a “need for evaluation of real exposure to coated seed ingestion by wild birds, including feeding behavior analyses and estimation of food intake rates.” We are therefore conducting a study to develop tools with which we are ascertaining whether birds are at risk for exposure to neonicotinoid-treated seeds in agricultural landscapes.

OBJECTIVES

The overarching objective is to ascertain whether birds are at risk for exposure to neonicotinoid-treated seeds in agricultural landscapes. Specifically, we will:

1. Identify birds consuming neonicotinoid-treated seeds and quantify consumption per foraging bout.
2. Quantify the rate of seed spillage and surface seed exposure after planting within fields.
3. Quantitatively link exposure and tissue/blood/excreta to neonicotinoid concentrations in chickens (lab study).
4. Determine whether neonicotinoid exposure in wild prairie grouse can be detected from non-lethal sampling methods or from hunter harvested birds (pilot field study).

METHODS

Documenting Consumption of Treated Seeds

In 2016, we selected 12 Wildlife Management Areas (WMAs) to place trail cameras from the 1,707 WMAs in Minnesota, of which a subset have food plots or Cooperative Farming Agreements (CFAs). The available data on CFAs on DNR-managed land indicated 7,420 acres (3,003 ha) of row crops in 341 CFAs in Region 4 (southern region) and 2,431 acres (984 ha) of row crops in 66 CFAs in Region 1 (northwest region; M. Benage and J. Williams, respectively, pers. comm.). We selected WMAs with a land cover composition similar to that of the surrounding landscape using the 2014 National Cropland Data Layer (USDA-NASS 2015) in ArcGIS 10.2 (ESRI 2015), but required them to have food plots or Cooperative Farming Agreements (CFAs) after they met the first criterion. Working on WMAs minimized bias in farming activities that might result from prior knowledge of the study. Furthermore, neonicotinoid-treated seed has been commonly used by private farmers on WMAs and many of the managers reported difficulty finding seeds that had not been treated. We prioritized this portion of the study in 2016 because farmers were prohibited from planting neonicotinoid-treated seeds on WMAs beginning in 2017.

Cameras were placed to minimize risk of theft and to view a recently planted field to document foraging at a simulated seed spill and exposed or submerged seeds or seedlings. Spills were simulated with 1000 corn or soybean seeds to allow determination of the time for birds to discover spills and the number of seeds consumed in each foraging bout/bird. Additionally, we placed cameras at 2 privately owned fields. Cameras were deployed in each location for 3–6 weeks after planting. At each field, 2 cameras were deployed; one that captured 1 image/sec in still photos and a second that captured 60 sec of video when triggered by motion. The camera set for still photos also took field scans at 5 min intervals between 0600–0800 hr and 1830–2030 hr to document birds foraging in fields during sunrise and sunset periods during the planting season. Images are currently being examined to identify species, number of birds consuming seeds, and number of seeds consumed per foraging bout, or in broader views, to document birds using crop fields after planting.

In 2017, we included more privately owned fields, which were generally larger than fields planted on WMAs. We placed 1 camera at each of 24 privately-owned fields in addition to placing cameras at 16 WMAs. Instead of capturing still images at simulated spills, which often produced ambiguous information about whether seeds were ingested, we instead set the cameras to record video only. In 2017 we also simulated spills with wheat, in addition to corn and soybean. We checked cameras once weekly to replace batteries and data cards and deployed cameras in each location for 2 weeks. Data collection was still underway at the time this report was written.

Quantifying Spills and Seed Surface Exposure

All chemically treated seeds (e.g., neonicotinoids, fungicides, other pesticides) are unnaturally colored, as mandated by the Federal Seed Act. These seeds are highly visible and easily identified by their unusual color (e.g., pink, blue, green, purple), which is used to prevent accidental feeding to livestock. We quantified the frequency of seed spills on the landscape by inspecting fields with visual access from roads, field access points, and roadsides in agricultural areas. We hoped to avoid bias in spill rates that might result from obtaining permission to access privately owned fields on foot, but this method makes the implicit assumption that spill rates associated with refilling hoppers and overfilling is similar for fields adjacent to roads and fields that are not adjacent to roads.

We identified 211 townships in the western third and southeastern part of the state with at least 50 miles of roads and 50% of the area in corn, soybeans, and/or wheat production using the 2014 Cropland Data Layer (USDA-NASS 2015) and the Department of Transportation (DOT) Roads Layer (DOT 2008) in ArcGIS. These criteria were used to select townships with visual access to fields from roads, while also not becoming so restrictive that the spatial distribution of the sample was constrained. We drew a spatially balanced sample of 50 townships and surveyed the 38 most western townships selected due to a later start to planting during the spring of 2016. In 2017, we selected 50 different townships and again surveyed the 38 westernmost townships due to a late start to planting, for a total of 76 townships surveyed during the 2 years of the study. We began in the southern counties and worked north beginning in late April as crops were planted.

We recorded locations and approximate number of seeds in spills near *recently planted* fields with the DNRSurvey mobile computer application. Documenting only *recently planted* fields allowed for control in temporal variation in the timing of planting. For example, a field that has not been planted yet will not have a spill at the time of sampling, which is different from a spill not occurring during planting. Thus, by only including recently planted fields in our estimates, we measured spills during planting. We defined a “field” as a quarter quarter-section (i.e., 40 acres). We recorded each quarter quarter-section in agricultural production, whether any part of it was recently planted (i.e., ≤early seedling stage), documented the amount (number of seeds) of spilled seed on the road, field edge, or visible in the field, and crop type (when possible). To determine the proportion of seed spills that contain neonicotinoid-treated seed, we collected seeds from accessible spills and will quantify 7 neonicotinoids (Chen et al. 2014).

To estimate the amount of seed at the soil surface after planting, we used a 1-m^2 frame to define plots in recently planted fields and counted all treated seeds visible within the frame after planting (Lopez-Antia et al. 2016). We sampled 5 plots in a field corner and 5 plots in the field center as estimated visually from field boundaries while standing in the field. For corner locations, we randomly selected 1 field corner per field by flipping a coin twice, and paced 15 m and 30 m along each edge in an L-shape that had the field corner for a vertex for a total of 5 measurements. This approach incorporated sampling parallel and perpendicular to planting rows, and we suspected that seed exposure would be greater at the end of rows at turning points than within rows. For field centers, we paced 15 m in each cardinal direction to sample for a total of 5 measurements including the center.

Linking Exposure to Concentrations in the Lab

We are quantitatively linking field sample concentrations to lab exposure concentrations through work with University of Minnesota - College of Veterinary Medicine (UMN-CVM) and Southern Illinois University Carbondale (SIUC). We are determining how many days post-exposure imidacloprid (i.e., the most common seed treatment in Minnesota, J. Zachmann, MDA, pers. comm.) is detectable in both non-lethally and lethally collected samples. A non-lethal method to determine sub-lethal exposure would facilitate data collection during spring planting when spills would be expected to be most numerous.

At UMN, domestic chickens (*Gallus gallus domesticus*) were orally exposed to imidacloprid (IMI) for 7 days and serially sampled during and after the course of exposure to simulate repeated sub-lethal exposures. Chickens served as our model species given their suitability to captivity and close taxonomic relationship with wild grouse (Family Phasianidae). Small sample sizes are commonly used in dosing studies because the differences among treatment groups are expected to be very large and variability within groups low (e.g., Berny et al. 1999, Bednarska et al. 2013). We exposed animals (n = 5) to 1, 5, and 20% of the LD_{50} (104.1 mg/kg IMI, Kammon et al. 2010) daily for 7 days by giving ~1.5 kg birds a daily IMI bolus of 1.04 mg/kg/day (“low”), 5.20 mg/kg/day (“medium”), and 20.80 mg/kg/day (“high”). The LD_{50} is the single dose that is expected to be lethal to 50% of test subjects. The LD_{50} could be obtained if chickens ingested ~260–946 corn seeds (depending on application rate to seeds, which varies among seed companies), or stated differently, 3–10 seeds is comparable to the 1% LD_{50} dose. Thus, these were realistic doses. Prairie grouse are smaller (0.6–1.2 kg) and thus a smaller dose (104–780 seeds depending on bird weight) would be expected to produce similar results. Other neonicotinoids have a lower LD_{50} than IMI so lethality would be expected at much lower seed ingestion levels than for IMI.

The full experiment was completed only for birds in the low and medium treatment groups, as birds in the high group were humanely euthanized on day 1 due to severe neurological and respiratory depression. Prior to exposure, baseline blood and excreta samples were collected. Sequential blood and excreta samples were collected on experiment days 1–21. Blood samples were collected at 0, 8, and 24 hours post-exposure, and then on days 8, 14, and 21 post-exposure. Birds that were considered at endpoint and euthanized had blood samples taken immediately before euthanasia. The low group was sampled for feces 1 day earlier than the medium group due to logistical challenges. Internal organ (i.e., brain, kidney, liver, spleen) and muscle samples were taken from birds that died during the treatment period or on day 21, whichever came first. Birds were weighed on all days of sampling. Samples were sent to SIUC for residue analysis (Chen et al. 2014).

Descriptive statistics and graphing of the available data from these lab studies was performed to understand in a preliminary sense how IMI concentrations changed over time, and in response to dose, on a tissue-specific basis. According to best practices, we have used geometric rather than arithmetic mean for chemical concentration data, which are typically lognormally distributed. Arithmetic mean is often biased high. Further statistical analyses will be conducted once the full dataset, including metabolites (i.e., neonicotinoids modified through metabolic processes), is obtained.

Detecting Neonicotinoids in Free-Ranging Birds

We also collected samples from wild birds through both invasive and non-invasive methods to try to identify ways to assess exposure to neonicotinoids in the field. Fresh fecal pellets and blood samples from trapped prairie grouse were collected during lek visits for a genetic study in spring 2015. Samples were stored frozen until shipped to the lab at SIUC. Hunters also voluntarily submitted harvested prairie grouse in fall 2015. Tissues and fecal pellets are being tested for thiacloprid (THIA), acetamiprid (ACE), thiamethoxam (TMX), imidacloprid (IMI), clothianidin (CLO), dinotefuran (DIN), and nitenpyram (NTP).

MNDNR staff also assisted with collections of birds observed foraging on treated seeds in the spring of 2016 under federal permit MB682323-0 issued to MNDNR. We are examining exposure from ingesta and tissue residue levels according to Chen et al. (2014) at SIUC.

RESULTS

Documenting Consumption of Treated Seeds

We are still viewing images collected by trail cameras at simulated spills during spring 2016 (*n* = 188,399 photos and 12,602 videos). In the images viewed to date, we have documented ring-necked pheasants, Canada geese, American crows, various species of sparrows and blackbirds, white-tailed deer, rodents, lagomorphs, and raccoons consuming treated seeds. We will continue viewing images during fall 2017 and winter 2017–18 and summarize results in future research summaries.

Quantifying Spills and Seed Surface Exposure

We observed 212 large seed spills that were visible from the road during surveys of 38 townships during 2016. However, we missed the peak of planting in many of the townships surveyed because the spring of 2016 was very wet and crops were planted later than usual. Nevertheless, at the time of our road-based surveys, 79,386 acres of corn, 82,341 acres of soybeans, 69,293 acres of wheat, and 7,753 acres of other crops were planted in the areas surveyed. Spill rates in the areas surveyed were calculated as 4 spills/10,000 ac corn, 14 spills/10,000 ac soybeans, 7 spills/10,000 ac wheat, and 15 spills/10,000 ac other crop types. Extrapolating statewide requires the assumption that spill rates visible in fields adjacent to roads are representative of spill rates in fields located elsewhere. If spills near roads are more likely to be cleaned up than those less visible to passersby, then this assumption may not be tenable. Yet, we did not observe spills being cleaned up during our surveys. Furthermore, most spills occur during hopper refilling, and this often occurs near field access points along roads. Thus we think our assumptions are reasonable. Applying our spill rates across the acres farmed statewide (8,450,000 acres of corn, 7,550,000 acres of soybeans, and 1,321,000 acres of wheat were planted in Minnesota during 2016 (National Agricultural Statistics Service; last accessed 5 June 2017 National Agricultural Statistics Service), we estimate nearly 15,000 large seed spills statewide and expect that if there is a bias, our estimates are biased low.

We documented exposed seeds at the soil surface in 25% of the 48 fields where we sampled 1 m^2 plots in 2016. Seeds were exposed in $\geq$1 centrally located plot in 14.6% of fields measured. Exposed seeds were detected in $\geq$1 corner plot of 18.8% of fields measured. Most (79%) of the fields we measured were planted to corn, 17% were planted to soybeans, and 4% were planted to wheat. Most (96%) sampled fields were on public land but 79% of the sampled fields on public land were planted by private cooperating farmers with their own equipment. We suspect that spill rates are influenced by the type of equipment used for sowing (Lopez-Antia et al. 2016) and possibly the seed type. These numbers are considered preliminary and subject to future revision. Data for the 2017 field season will be included in future reports.

Linking Exposure to Concentrations in the Lab

We collected 72 blood samples, 100 fecal samples, 15 muscle, brain, liver, and kidney samples, and 103 eggs during experiments for neonicotinoid analysis. Imidacloprid (IMI) was detected more frequently and for a longer duration post-exposure in fecal samples (90.9%, $\leq$21 days post exposure) than blood (32.9%, $\leq$7 days post exposure). Blood concentrations increased from the first samples taken at the start of the experiment (hr 0), increased at hr 8 and declined again at hr 24 (Figure 1); after this time, samples did not contain detectable IMI except for 1 sample taken on day 8. Fecal IMI concentrations followed a 3rd order polynomial pattern, increasing from the start of the experiment (day 0) until approximately day 6, decreasing until day 18 and holding steady or slightly increasing by day 21 (Figure 2). The low dose group tended to exhibit lower IMI fecal concentrations than birds in the medium dose group, as expected (Table 1). IMI was rapidly removed from blood, but the change in concentrations varied 17,234-fold (c.f., 279-fold in feces; fold change is maximum detected concentration/minimum detected concentration across all groups and times), and thus blood may provide a more sensitive indicator of an acute exposure than feces. By contrast, fecal samples provided a more integrated, longer, and more consistent detection in exposed birds (Figure 2) and thus may be more applicable to field applications where time from chemical exposure will be more variable.

IMI was measured in internal organs (Figure 3) collected on the final day of the experiment, depending on when birds were euthanized. Low- and medium-dosed birds were euthanized on day 21, whereas high-dosed birds were euthanized after showing clinical signs of distress on day 1. Detection frequency of IMI was highest in kidney, liver, and spleen (73.3%), although muscle and brain also exhibited similar detection frequencies (66.7%). Geometric mean tissue concentrations were highest in brain and lowest in the kidney (Table 2).

Detecting Neonicotinoids in Free-Ranging Birds

Field-collected prairie grouse samples sent for neonicotinoid analysis included 61 sharp-tailed grouse fecal pellet groups and 34 greater prairie-chicken fecal pellet groups collected in 2015, and 46 and 27 pellet groups, respectively, in 2017. We also collected 5 blood samples from trapped sharp-tailed grouse, as well as 2 brains and 3 breast muscles from sharp-tailed grouse for which we had whole carcasses and sent them for neonicotinoid analysis. Hunters submitted livers from 11 prairie-chickens, 22 sharp-tailed grouse, and 3 prairie-chicken/sharptail hybrids during fall 2015, and 16 prairie-chicken, 26 sharp-tailed grouse, and 2 pheasant livers during fall 2016.

A subset of field samples from wild prairie grouse has been analyzed for neonicotinoids thus far. Forty-one of 46 (89%) livers collected from hunter-harvested sharp-tailed grouse, 18 of 27 (67%) greater prairie-chicken livers, and 3 of 3 sharptail-chicken hybrids from hunter-submitted samples had detectable concentrations of at least 1 neonicotinoid. Three of 3 blood samples analyzed thus far have tested negative for neonicotinoids. Dinotefuran and NTP were not detected in any samples. The most commonly detected neonicotinoids in prairie-chicken livers were IMI (63%), CLO (11%), THIA (4%), ACE (4%), and TMX (4%). The most commonly detected neonicotinoids in sharp-tailed grouse livers were IMI (83%), CLO (13%), THIA (13%), ACE (9%), and TMX (2%). Maximum concentrations of neonicotinoids in prairie-chicken livers were 8.3 ng/g IMI, 4.2 ng/g CLO, 1.1 ng/g THIA, 0.21 ng/g, ACE, and 0.43 ng/g TMX, respectively. Maximum concentrations detected in livers of harvested sharp-tailed grouse were 84.5 ng/g IMI, 3.58 ng/g CLO, 1.18 ng/g THIA, 0.71 ng/g ACE, and 0.5 ng/g TMX. Similarly, 22 of 34 (65%) fresh prairie-chicken fecal pellets and 47 of 56 (84%) sharp-tailed grouse pellets collected from leks during spring 2015 contained detectable concentrations of at least 1 neonicotinoid. The most commonly detected neonicotinoid in the greater prairie-chicken fecal pellets was IMI (71%), followed by CLO (9%), and THIA (9%). Acetamiprid and TMX were not detected in feces, perhaps due to differences in the way they are metabolized or excreted. Maximum concentrations of IMI, CLO, and THIA in feces were 6.12 ng/g, 0.90 ng/g, and 1.05 ng/g, respectively. In sharp-tailed grouse pellets, the most commonly detected neonicotinoids were IMI (80%), CLO (21%), THIA (11%), ACE (2%), and TMX (2%). Maximum concentrations were 39.7 ng/g IMI, 7.57 ng/g CLO, 0.9 ng/g THIA, 0.2

ng/g ACE, and 0.5 ng/g TMX. Samples which contained multiple neonicotinoids (n = 16 livers and 14 pellets) generally contained IMI, except for 4 livers and 2 pellets.

Birds collected while foraging on treated seeds included 1 ring-necked pheasant, 5 red-winged blackbirds (*Agelaius phoeniceus*), 2 yellow-headed blackbirds (*Xanthocephalus xanthocephalus*), 4 brown-headed cowbirds (*Molothrus ater*), and 5 common grackles (*Quiscalus quiscula*). Two brown-headed cowbird livers tested positive for exposure to IMI and CLO. One yellow-headed blackbird liver tested positive for IMI. Livers of all other birds collected while foraging on treated seeds tested negative for recent neonicotinoid exposure.

DISCUSSION

Fecal samples appear to provide a possible non-invasive means to detect exposure in birds based on our findings and the potential to refine analytical methods. Previous studies have demonstrated that neonicotinoids (e.g., thiamethoxam) are excreted primarily through the kidneys in mammals (Bednarska et al. 2013, Tomizawa and Casida 2005). Ongoing analytical work to measure metabolites of imidacloprid in feces is expected to provide a more sensitive (i.e., higher fold concentration change) assay than current parent compound (i.e., imidacloprid unmodified by metabolic processes) data. Further work will be required to quantify how the potential environmental imidiacloprid exposure scenarios (concentration, duration, and frequency) influence the detection of parent compound and metabolites in feces and the uric acid wash. However, fecal samples could be collected from the GI tract of hunter-killed birds, from live birds, or non-invasively from the environment. Further work is necessary to refine non-invasive collection because UV light can and microbial degradation may degrade neonicotinoids (Lu et al. 2015; Lu et al. 2016; Ma et al. 2014), so pellet freshness would be an important consideration.

Our data provide evidence that internal organs can serve as an indicator of imidacloprid exposure in lethal collections including hunter-killed birds. However, based on detection frequencies in organs and feces, fecal samples may provide a more reliable index of exposure than organs. Berny et al. (1999) reported that liver and kidney had the most consistent imidacloprid concentrations in fatally exposed wild birds, whereas crop and gizzard provided inconsistent concentrations. However, Lopez-Antia et al. (2015) reported that imidacloprid could be consistently detected in crops and livers of dosed partridges (*Alectoris rufa*).

The highest concentration of imidacloprid detected in livers of harvested prairie grouse was higher than that of chickens in the low and medium dose group at the end of the experiment. However, it was lower than the high LD_{50} group after early euthanization. Similarly, the highest concentration of imidacloprid detected in field collected feces was lower than both the 1% and 5% dose groups shortly after exposure, and was more similar to both of these groups a few weeks post-exposure. We cannot know if this indicates a lower initial exposure, the passage of time since exposure, or both; but, given that 1% LD_{50} (1.04 mg/kg) is comparable to the dose received after consuming 3–10 corn seeds and that imidacloprid can be detected in tissues at least 21 days post-exposure, we consider it likely that this finding reflects an exposure to imidacloprid that occurred a few weeks prior to sample collection. Winter wheat is planted in September and October in Minnesota, so grouse might be newly exposed to treated seeds in the fall, although it is not clear how long spring exposure would be detectable in organs. At a minimum, detection of imidacloprid in tissues of wild birds provides us with a qualitative index of exposure, which is one step closer to understanding the effects of imidacloprid in wild birds in Minnesota.

The high detection frequencies of imidacloprid in internal organs on experimental day 21 after 7 consecutive days of exposure indicates a persistence of imidacloprid that is notable but not easily comparable to other acute studies. Most studies have suggested a rapid metabolism and elimination (~48 hours) of parent (i.e., unchanged) compound in the urine after *single* oral doses (Bednarska et al. 2013; Tomlin 2004). Our findings demonstrated a relatively high persistence of parent compound in feces and organs and may therefore indicate an appreciable toxicological risk for birds.

The locations of the compounds in the tissues provide insight into which systemic effects warrant examination. Based on the high splenic concentrations, we hypothesize imidacloprid will cause immune system changes in birds. The detection of imidacloprid in neurological tissues (brain) indicates a potential for behavioral changes as well. If immune system or behavioral effects impact survival and reproduction, then population–level impacts are plausible. Our laboratory data will be useful in understanding the absorption, distribution, excretion, and effects of imidacloprid, as well as in the design of future laboratory and field studies in birds. We will also contribute some of the first information on exposure of wild birds in the United States to neonicotinoids.

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Table 1. Summary of imidacloprid detections in domestic chicken blood and feces in each of 3 dose groups at University of Minnesota – College of Veterinary Medicine in 2015. Note that birds in the high dose group were euthanized early, which may have limited the ability to eliminate imidacloprid in feces.

	Dose (mg/kg/day)	N	Percent detects	Fold change	Median	Geometric mean	Minimum	Maximum
Blood (ng/ml)	1.04	6	20.0	4.2	1.7	1.4	0.5	2.1
	5.02	10	33.3	9.8	2.6	2.2	0.7	6.9
	20.80	8	61.5	2051.7	3270	805.6	4.2	8617
Feces (ng/g wet weight)	1.04	26	81.3	91.8	14.6	10.1	0.8	73.4
	5.02	39	97.5	278.9	19.1	14.1	0.7	195.2
	20.80	5	100.0	2.8	3.2	3.7	2.3	6.5

Table 2. Summary of tissue concentrations of imidacloprid in all laboratory-exposed chickens for all dose groups combined at University of Minnesota – College of Veterinary Medicine in 2015.

Tissue	First detection (day)	Last detection (day)	Fold change	N	Percent detects	Min conc[a]	Max conc[a]	Median conc[a]	Geometric mean conc[a]	SD
Feces	1	21	279	70	90.9	0.7	195	14.6	11.3	35.9
Kidney	NA[b]	NA	1681	11	73.3	0.5	823	1.7	13.4	276.5
Liver	NA	NA	19882	11	73.3	0.3	5766	6.7	64.6	2473.6
Spleen	NA	NA	30413	11	73.3	0.2	6387	16.8	63.6	2320.8
Brain	NA	NA	10410	10	66.7	0.6	5725	1212.7	76.7	2295.8
Muscle	NA	NA	3469	10	66.7	0.8	2775	382.3	62.8	1128.5
Blood	1	8	17234	24	32.9	0.5	8617	4.1	14.1	2389.5

[a] Conc = concentration (ng/g wet weight in tissues and ng/ml for blood).

[b] NA = Not applicable because tissues were collected when chickens were killed the last day.

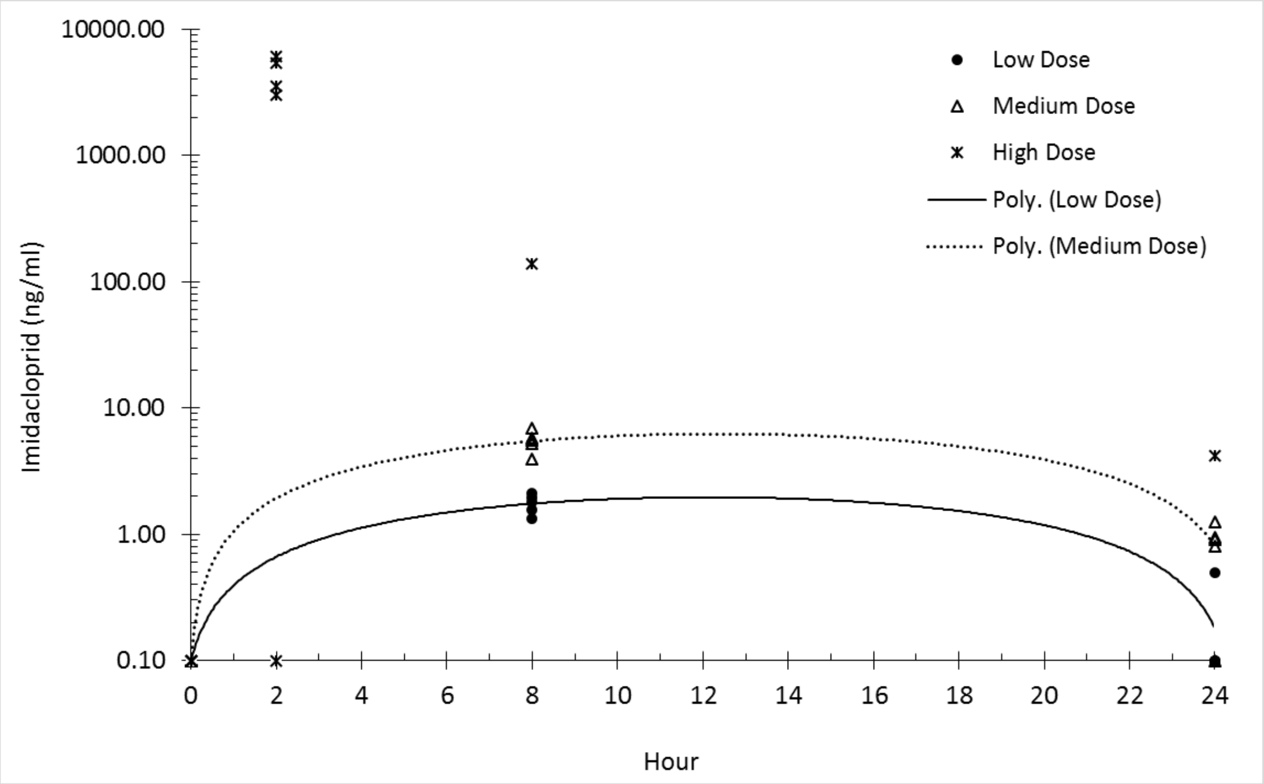

Figure 1. Changes in imidacloprid (IMI) concentrations in blood of dosed domestic chickens over time after one dose at University of Minnesota – College of Veterinary Medicine in 2015. IMI doses were 1%, 5%, and 20% of a reported IMI LD_{50} for chickens (i.e., low, medium, and high dose groups, respectively). IMI detection limit is 0.10 or -1.0 $\log_{10}$ ng/ml in blood. Data points overlap when plotted on x-axis minimum value. A polynomial (Poly) trend line was fit for the low- and medium-dosed birds, but could not be fit to the data from high-dosed birds because chickens in this dose group were euthanized within 24 hours due to animal welfare concerns. Thus, the high dose group is not directly comparable to the other dose groups.

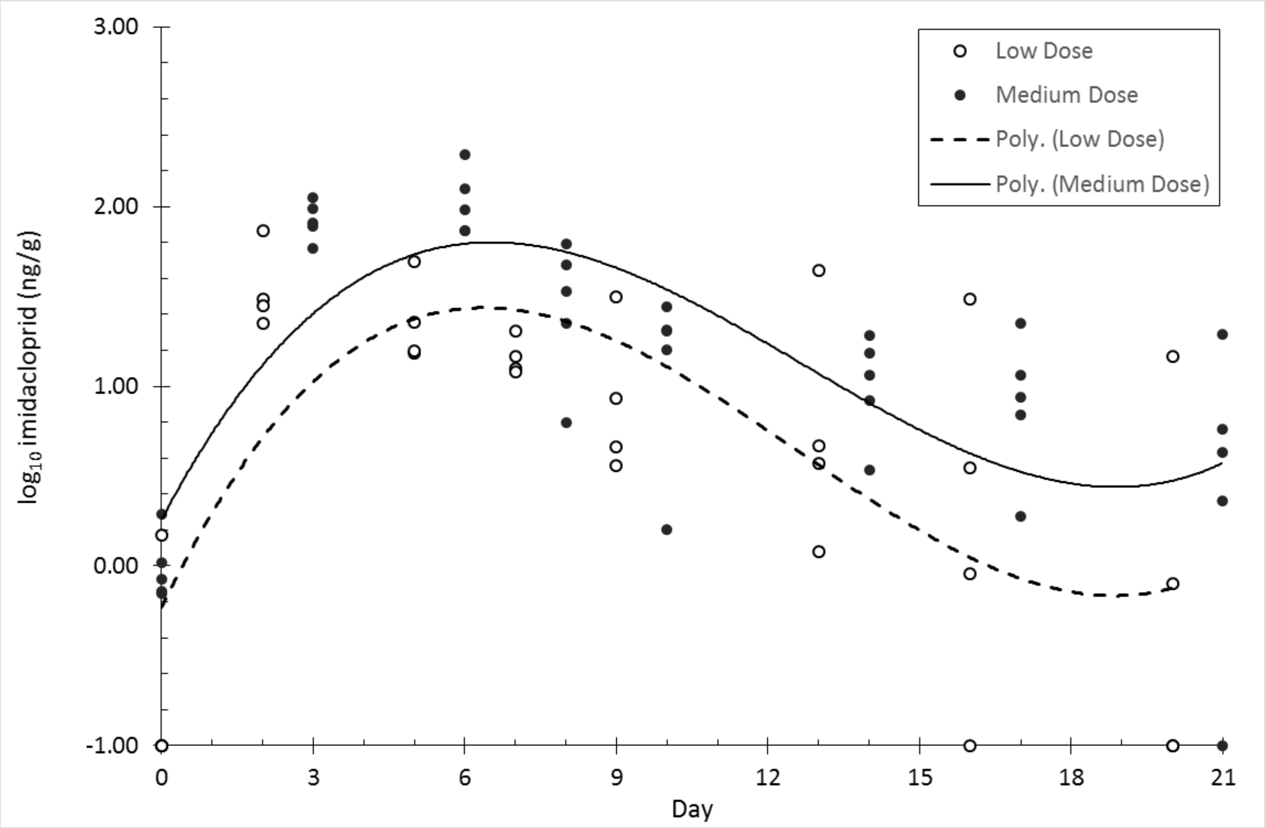

Figure 2. Changes in imidacloprid (IMI) concentrations in feces of dosed domestic chickens over time at University of Minnesota – College of Veterinary Medicine in 2015. Samples collected on day 0 were baseline samples, prior to exposure. Birds received a daily IMI dose for 7 days of 1% (low dose) and 5% (medium dose) of a reported IMI LD_{50} for chickens. The last day of dosing occurred on day 7 of the 21 day experiment. IMI detection limit was 0.10 or -1.0 $\log_{10}$ ng/g in feces. The high dose group is not included because samples were collected only on day 0 so no temporal trends could be determined. Chickens in the high dose group were euthanized within 24 hrs after dosing due to animal welfare concerns. Thus, the high dose group is not directly comparable to the other dose groups. Polynomial (Poly) trend lines were fit to the data for the low and medium dose groups.

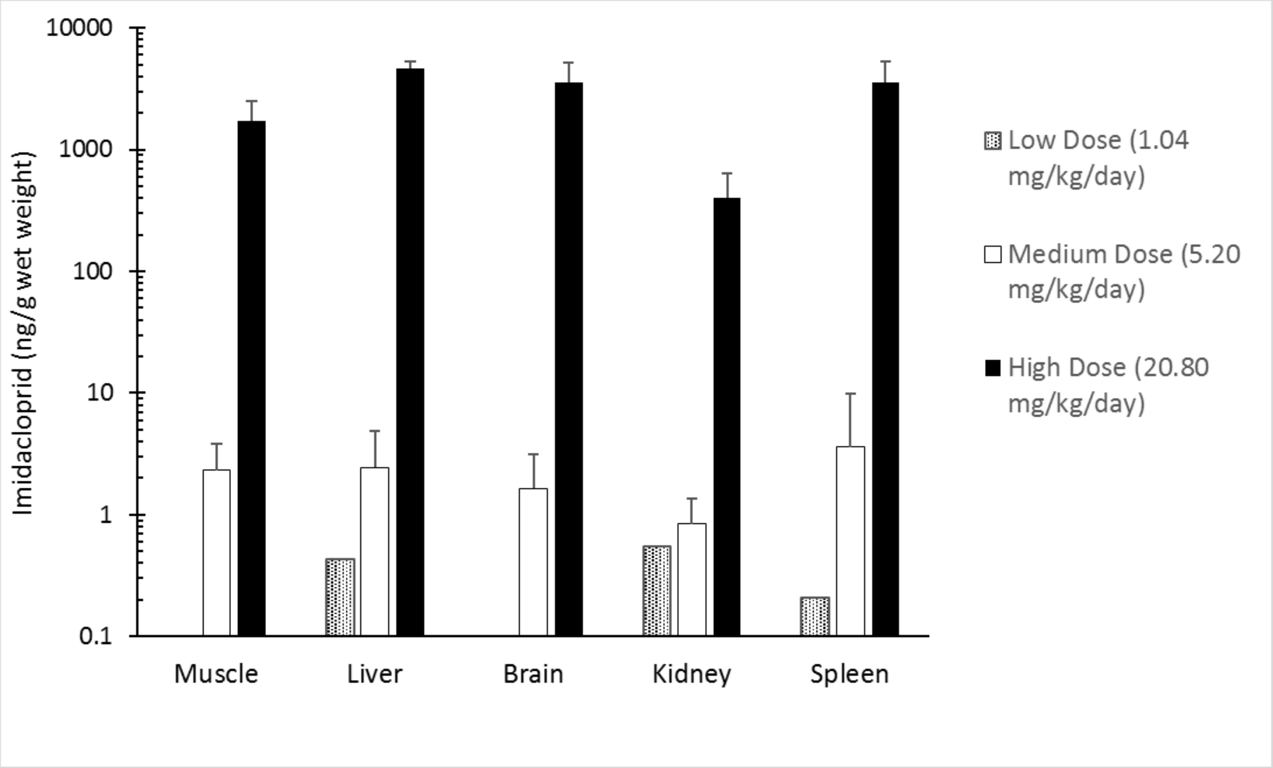

Figure 3. Concentrations of imidacloprid (geometric mean + SD ng/g wet tissue weight) in tissues of laboratory-exposed domestic chickens on experimental day 1 (high dose) or 21 (low and medium dose) at University of Minnesota – College of Veterinary Medicine in 2015. Data at the detection limit of 0.10 ng/g are not visible. Error bars represent the standard deviation of observations for a given group.

DEPARTMENT OF
NATURAL RESOURCES

MONITORING SPRUCE GROUSE IN MINNESOTA: SURVEY DEVELOPMENT (2014–2017)

Charlotte Roy, John Giudice, and Chris Scharenbroich

SUMMARY OF FINDINGS

Data collection began in 2014 to develop survey methodology for a large-scale survey of spruce grouse (*Falcipennis canadensis*) in Minnesota. During 2014 and 2015, we examined 2 primary methods of spruce grouse detection: a cantus-call survey and a fecal pellet survey. Based on field work conducted in 2014 and 2015, we determined that pellet surveys had 3-5 times the apparent detection rate of call surveys (20% and 4%, respectively). During 2015, pellet and call surveys at paired points on and off roads allowed examination of the effects of roads on survey counts. These paired surveys indicated that detections at road-based points were lower than at points located off roads at 1 of 2 study areas, but this effect was minimal in forest types preferred by spruce grouse. In 2016, we piloted a road-based pellet survey throughout the probable spruce grouse range in Minnesota. Results were consistent with anecdotal accounts of spruce grouse observations from wildlife managers and indicated that spruce grouse are relatively rare in the Northern Minnesota Drift and Lake Plains Ecological Classification System (ECS) section and more abundant in the Northern Superior Uplands and Northern Minnesota and Ontario Peatlands sections, with relative abundance increasing along a southwest to northeast gradient. We conducted simulations to examine our ability to detect meaningful changes in the population (>15% decline over 10 years) and concluded that the pellet survey could accomplish this goal. In 2017, we visited 1,426 potential survey points (85% of the potential sampling frame) to determine which points were suitable for the operational survey. We were able to eliminate 266 points due to inaccessibility (e.g., the presence of wetlands, ditches, steep topography, private land ownership) or unsuitable habitat (e.g., recent harvest, blowdown, or fires). Our findings will inform the selection of the set of points to be included in the annual operational survey, which we plan to initiate in 2018. Upcoming work will identify survey routes (clusters of points) and training cooperators so that surveys are conducted similarly across the survey region.

INTRODUCTION

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). Minnesota is unique among the Lake States in having a sizeable spruce grouse population that still permits spruce grouse hunting. Yet, the only data the Minnesota Department of Natural Resources (MNDNR) collects on spruce grouse is estimated total harvest as part of the annual MNDNR small game mail survey (Dexter 2016). Estimated total harvest has been 10,000–27,000 birds/year over the last 10 years (Dexter 2016). However, spruce grouse harvest may be more reflective of ruffed grouse hunter numbers than spruce grouse numbers; thus these data cannot be used as a population index (Gregg et al. 2004). The MNDNR mail survey also provides some information on geographic distribution via a “county hunted most” question, but it is probably insufficient for monitoring anything less than large-scale range changes. Hence, the

MNDNR has limited data on spruce grouse distribution, abundance, and population trends in Minnesota despite a responsibility to manage spruce grouse during a period of expected habitat loss due to climate change (see Roy et al. 2013a). Thus, there is a need for better population-monitoring data for spruce grouse in Minnesota.

Developing large-scale monitoring programs that are both reliable and cost effective is challenging, especially when the species is relatively rare and occupies habitats that are not easily accessible. New York (Fritz 1979) and Wisconsin (Worland et al. 2009) have conducted statewide surveys of spruce grouse. Wisconsin used a spatially balanced stratified sampling design with 4 stand size classes (range: 8.1–1,242 ha), in which they surveyed multiple points in 81 forested wetlands during 3 visits. In New York, 67 habitat patches were surveyed during 220 visits. However, these surveys were only 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, financial, and resource constraints often limit survey-design options for large-scale monitoring efforts. 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 addressed.

As part of a pilot study, we evaluated survey methods that might be useful for monitoring spruce grouse populations in Minnesota. We evaluated an auditory survey using playback of female cantus calls, which is the most common approach to survey spruce grouse (Fritz 1979, Boag and McKinnon 1982, Whitcomb et al. 1996, Lycke et al. 2011, among others). We also conducted pellet surveys and used pointing dogs to locate birds on survey plots following completion of a cantus-call survey (Roy et al. 2013b, 2014).

OBJECTIVE

1. Assess the feasibility of using a roadside survey to determine the distribution and population trends of spruce grouse in Minnesota.

2. Design and implement an annual roadside survey in 2018.

STUDY AREAS

In 2014, we focused on the Red Lake Wildlife Management Area (RLWMA) and Beltrami Island State Forest (BISF; Roy et al. 2013b, 2014). This study area is on the southwestern edge of the presumed spruce grouse range, where changes (range contraction or negative trends in abundance, density, or patch occupancy) might occur earlier than in more central portions of the range. In 2015, we focused on portions of RLWMA and BISF where spruce grouse detections occurred in 2014, so survey methods would be evaluated in areas where birds were known to occur (Figure 1). We also added a second study site near Isabella (Figure 2), which is more centrally located within Minnesota spruce grouse range. This study site offered insights into survey methods where populations might be more robust to initial habitat changes. Hereafter, we refer to this study site as the NE study site and the one at RLWMA and BISF as the NW study site.

In 2016 and 2017, we expanded the survey area to include all or most of spruce grouse range in Minnesota (Figure 3 and 4). The current limits of spruce grouse range are unknown, so we focused on forest types used by spruce grouse within 3 ECS sections (Northern Minnesota and Ontario Peatlands, Northern Superior Uplands, and Northern Minnesota Drift and Lake Plains) to delineate an area to be surveyed for spruce grouse. We also referenced harvest data reported in the Small Game Hunter Mail Survey (Dexter 2015) to incorporate county-level harvest information for spruce grouse.

METHODS

Identifying Spruce Grouse Habitat

The literature is conflicting with respect to forest ages of importance for spruce grouse; earlier successional stages have been reported to be important in the western U.S. (Boag and Schroeder 1992), but mature forest was important in Wisconsin (Anich et al. 2013). In 2014, we included forest types reported to be preferred by spruce grouse in our region, including jack pine (*Pinus backsiana*), black spruce (*Picea mariana*), and tamarack (*Larix lariana;* Robinson 1969, Pietz and Tester 1982, Anich et al. 2013). We included all stand ages because of the lack of clarity in the literature but focused on preferred habitat types rather than all used habitat types. We also included white cedar (*Thuja occidentalis*), which was reported to be used but not a preferred habitat type (Anich et al. 2013), because managers were specifically interested in surveying this forest type.

In 2015, we added balsam fir (*Abies balsamea*) and red pine (*Pinus resinosa*) forest types to our survey. This decision was based on 2014 detections in stands with these species components that exceeded our expectation of use based on their representation in the sample. We also added white spruce (*Picea glauca*) because it was reported as used but not preferred in the literature, and inclusion of these other used but not preferred stand types seemed to warrant its inclusion for consistency. We used Forest Stand Inventory (FIM) data on state managed lands administered by the MNDNR at both the NW and NE study sites to identify survey points based on forest stand types and age. Field Sampled Vegetation (FSVeg) spatial data was also used on lands managed by the U.S. Forest Service Superior National Forest at the NE site to identify survey points in the appropriate forest stand types. We excluded stand ages listed as “under development” (i.e., 0–5 years) in the FIM data to exclude areas that might not have established as forest. Timber harvest data (US Forest Service 2015a), Motor Vehicle Use Maps (U.S. Forest Service 2015b), and fire records (National Interagency Fire Center 2013) were also used for the NE study site to exclude stands that were recently harvested or burned and to identify roads suitable for survey routes.

In 2016 and 2017, we continued to use forest types used in 2015 (black spruce, jack pine, balsam fir, red pine, white spruce, tamarack and white cedar $\geq$6 years old) and we expanded our forest-inventory data to include lands administered by the U.S. Forest Service Chippewa National Forest as well as by county land departments, including Aitkin, Beltrami, Carlton, Cass, Clearwater, Crow Wing, Hubbard, Itasca, Koochiching, Lake, and St. Louis Counties. Since harvest and stand replacement disturbance information was not readily available for all forest-inventory sources, a satellite-interpreted forest loss data layer (Hansen et al. 2013) was used to identify areas of forest stands $\geq$6 years old. Forest stands meeting the cover type and age requirements were further dissolved into patches to determine sites that had a sufficient amount of habitat to support spruce grouse. Habitat patches $\geq$8 ha (Fritz 1979, Whitcomb et al. 1996) that overlapped accessible roads were used to identify potential stands to survey.

Survey Points and Routes

In 2014, we used Geographic Information Systems (GIS) road layers (Minnesota Department of Transportation and MNDNR) to identify roadways that were within 40 m of potential habitat polygons (jack pine, black spruce, tamarack and white cedar; see above). We then classified roadways as primary or secondary based on their accessibility during the April–May survey period (e.g., plowed vs. not plowed). We established survey points on road segments that bisected or were within 40 m of habitat polygons. Points were spaced $\geq$300 m apart to ensure independence among points based on estimates that playback calls can be heard 100–150 m from the speaker (Schroeder and Boag 1989; Lycke et al. 2011; Anich unpubl. data). Road segments and associated survey points were then grouped into survey routes based on logistical considerations.

In 2015, we used the same GIS layers to select survey points, but also used current data for U.S. Forest Service roads, forest harvest, and fire data for the NE study site (U.S. Forest Service 2013, National Interagency Fire Center 2013, U.S. Forest Service 2015a,b). However, our focus in the second season was a comparison of off-road and on-road survey points to examine the impact of roads on survey detections. We selected paired points that had at least 30% spruce grouse habitat (based on selected forest types) within 150 m of each point, but limited our selection to areas where habitat occurred on both sides of the road. Off- and on-road points were separated by 300 m, and we alternated the side of the road where off-road points were selected, except when creeks limited access on foot.

In 2016 and 2017, we used a GIS to identify 1,862 and 1,686 potential survey points, respectively, which were in probable spruce grouse range, located on accessible roads, permitted access off road without limitations by water barriers, had ≥30% spruce grouse habitat within 150 m of each point on both sides of the road, and were associated with spruce grouse habitat patches ≥8 ha. In 2017, we eliminated nearly 200 points after review of air photos indicated that they obviously lacked the habitat and access requirements that were otherwise selected through analysis of the available forest inventory, roads, and hydrography data. We spaced points ≥400 m to obtain the greatest spatial coverage of focal stands throughout the probable spruce grouse range. In 2017, we manually added points that met survey criteria on tribal lands owned by the White Earth Nation, Red Lake Band of Chippewa, Leech Lake Reservation, Fond Du Lac Band and Grand Portage Band of Lake Superior Chippewa, and 1854 Treaty Authority. In 2016, we used a Generalized Random Tessellation Stratified Sampling approach (Stevens and Olsen 2004) to select seed points for approximately 80 routes and attempted to identify groups of 8–10 points that were in spatial proximity to construct survey routes. We also considered proximity to potential lodging centers (travel time), local expertise on accessibility, and the distribution of routes by ECS subsections and sections when selecting the final sample of routes and points. Our final sample in 2016 consisted of 65 routes with 2–13 survey points/route (median = 10). Forty-three routes (400 survey points) were located in the N. Superior Uplands (core of probable spruce grouse range in MN), 11 routes (120 points) were in the N. Minnesota & Ontario Peatlands (containing the RLWMA and BISF), and 11 routes (93 points) were in the N. Minnesota Drift & Lake Plains (southern edge of probable spruce grouse range in MN). In 2017, we did not cluster points into routes, but rather attempted to scout all possible points that would be suitable for inclusion in an annual survey.

Cantus Call Surveys

We used a playback of female cantus calls to conduct point-count surveys of spruce grouse (Fritz 1979, Boag and McKinnon 1982, Schroeder and Boag 1989, Whitcomb et al. 1996, Lycke et al. 2011). In 2014, we surveyed as many points as possible to provide information on survey duration (1–15 min), time needed to complete multiple surveys, habitat associations, and the responsiveness of spruce grouse to cantus calls (i.e., time of day and season). Surveys were conducted during April–May, beginning at sunrise, when winds were <10 mph and precipitation was absent or light. Each point count lasted 15 min (Lycke et al. 2011, Anich et al. unpubl. data) and was divided into 5 consecutive 3-min listening intervals. The 8-sec cantus call was broadcast once per min throughout the 15-min listening period. Observers recorded initial and subsequent detections of each spruce grouse by listening interval, which allowed us to construct individual detection histories for a time-of-detection analysis (TOD, Alldredge et al. 2007). We also recorded type of initial detection (flutter flight, approach, etc.), survey date, arrival time, wind speed, temperature, dominant tree species (as classified from the roadway: jack pine, black spruce,tamarack, white cedar, red and white pine, balsam fir, deciduous, other), and background noise (none, low, medium, high).

In 2015, we modified call-survey methods to incorporate findings from 2014. Specifically, we reduced the survey length from 15 to 9 min, began surveys 30 min earlier, and ended call surveys by 0930 hr. For analysis, we used a dynamic occupancy modeling approach (MacKenzie et al. 2006:183–224) to look at TOD and revisits in the same analysis. We used the ‘colext’ function in the R package ‘unmarked’ (Fiske and Chandler 2011, R Core Team 2016) to fit models. We used visits as the primary sampling unit and TOD as secondary sampling occasions, and we allowed the true occupancy status to change between visits (i.e., via transition probabilities). We examined 4 site-level covariates (study area, year, location [road vs. off-road], forest type), 5 visit-level covariates (observer, survey date, start time, start temperature, and wind speed), and 2 observation-level covariates (TOD interval and previous detection).

Call surveys were discontinued in 2016. Data collected in 2014 and 2015 indicated that pellet surveys had substantially higher detection probabilities and would be easier to implement in a large-scale survey of spruce grouse in northern Minnesota

Pellet Surveys

We counted grouse pellets and roost piles <1 m on either side of transects. We distinguished ruffed grouse pellets from spruce grouse pellets on the basis of length, thickness, uric acid wash, and color (N. Anich, A. Ross, M. Schroeder, pers. comm.). Ruffed grouse pellets tend to be shorter, thicker, and usually have a uric acid wash, whereas spruce grouse pellets are longer, thinner, and infrequently have a uric acid wash. Spruce grouse pellets are also darker green in color when spruce grouse are consuming conifer needles (during winter), but color changes depending on diet (personal observation); spruce grouse pellets can have a similar color to ruffed grouse pellets later in the spring. Finally, we recorded dominant and subdominant tree species along each circular path to compare forest-type classification based on GIS, roadside observations, and pellet surveys.

In 2014, we surveyed circular transects of 75-m and 100-m radii centered on call survey points on roads. In 2015, we surveyed circular transects of 100-m radius centered on paired points on and off roads, because the larger radius improved detection. Surveys were repeated up to 3 times to allow for modeling of detection using function ‘occu’ in R package ‘unmarked’ (Fiske and Chandler 2011). In 2016, pellet surveys were conducted at each transect once to maximize spatial coverage, and all pellet transects were centered on roads.

In 2016, we fit generalized linear mixed-effect models to the data to compute 2 monitoring metrics: an occupancy index (using presence-absence of pellets and a binomial link function) and a pellet-count index (using a Poisson link function). In each case we treated ‘route’ as a random effect to account for the clustered sampling design, and evaluated potential covariates affecting the response metric (e.g., amount of spruce grouse habitat, spatial location) as well as covariates that might serve as a surrogate for probability of detection (i.e., snow coverage [none, partial, complete] and days since last snow; based on results from 2014 and 2015). We used the function ‘glmer’ in the R package ‘lme4’ (Bates et al. 2015, R Core Team 2016) to fit the models, and we used AIC to select among competing models. Because our sample of points was not proportionally allocated (we sampled more heavily in core areas), we used our best approximating models, with the surrogate variable for detection held constant, to predict mean naïve occupancy and pellet abundance for each point in the sampling frame (1,862 points). We then computed a simple arithmetic mean prediction by ECS section and rangewide to generate 2 monitoring metrics. We used a bootstrap of routes (200–300 replicates, with replacement) to compute percentile confidence intervals that included uncertainty in model-fitting and prediction. We also used the R package ‘akima’ (Akima and Gebhardt 2015) to implement a bivariate interpolation of our irregularly spaced prediction surface, which we used to qualitatively assess how model predictions varied over Minnesota’s probable spruce grouse range as a function of spatial location and the relative abundance of jack pine and black spruce cover type.

Lastly, we used a Monte Carlo simulation with 200 replicates to evaluate whether the proposed sampling effort was adequate to detect a true decrease in a simulated population over a 10-yr monitoring interval, where the decrease was manifested by a decreasing trend in probability of site use. We allowed the decrease in mean probability of use to vary by ecoregion ($\lambda = 0.935$ annual decline in the southern part of the range and smaller decreases elsewhere, $\lambda = 0.984$ and 0.972, corresponding to 10-year declines of 49%, 15%, and 25%, respectively) and included small-to-moderate amounts of random variation that reflected annual variability (process variation), geographic variability, and binomial variation in the state (probability of use) and observation (probability of detection) processes. We used pilot-study results to inform starting parameter values and sampling and process variation in a Monte Carlo simulation. We used a generalized mixed-effects model to estimate a trend (in our index of use) for each simulated population and then evaluated the distribution of the estimated trends and the proportion of estimated trends that were negative (a qualitative power analysis).

In 2017, we attempted to refine our detection covariates by conducting pellet surveys at 76 points, of which 74 had been surveyed in 2016. We documented the time since last snow fall and rated the survey conditions on a scale of 0-10, with 0 being the poorest and 10 indicating optimal survey conditions. We used 0 to indicate that recent snowfall covered all but the freshest pellets, and 10 to indicate a strong contrast against pellets created by a dissipating snow pack, with 100% ground coverage by snow received >10 days prior. We also noted whether pellets were found on snow or on bare ground and documented how much of each transect (to the nearest 5%) was in a forest type identified as being used by spruce grouse.

We also scouted 1,426 potential survey points (85% of the potential sampling frame) to help determine the appropriateness of a point for inclusion in an annual pellet survey based on a coarse assessment of site-level vegetation structural density (i.e., open/closed), forest maturity (i.e., early/mature), and the absence of wetlands, ditches, steep topography, private property, or recent timber disturbance. The remainder of points could not be scouted due to impassable roads during the scouting period.

RESULTS

2014 Abridged

We detected spruce grouse at 26 (4%) of 530 call-survey points. Birds were detected in all 5 listening intervals, although 78% of birds were detected in the first 3 intervals. Our best approximating hierarchical occupancy model included detection covariates for survey date, arrival time, whether the bird was detected in a previous listening interval, and an occupancy covariate describing the relative amount of spruce grouse habitat surrounding the listening point (habitat sides = 0, 1, 2). Mean probability of detection was negatively associated with survey date and arrival time (Figures 5 and 6). Not surprisingly, probability of detection increased dramatically if a bird was detected in a previous listening interval. The mean probability of occupancy for a listening stop with spruce grouse habitat on both sides of the road was 0.23 (95% CI = 0.02–0.78; Figure 7), and the overall probability of detection for the entire 15-min survey, given mean covariate values for survey date and arrival time, was 0.25 (95% CI = 0.02–0.93).

We conducted pellet surveys at 230 listening points and detected pellets at 45 (20%) points. Pellet surveys and cantus-call surveys had 82% concordance for presence-absence of spruce grouse.

However, we detected pellets at 36 points (16%) where we failed to detect a bird during cantus-call surveys. This contrasts with failure to detect pellets at 5 (2%) points where we detected spruce grouse during cantus-call surveys. The 100-m radius survey path resulted in 28 detections (39%) compared to 18 (11%) detections with a 75-m radius path. Nineteen additional spruce grouse were located while walking transects around survey points with dogs.

2015 Abridged

We surveyed 200 paired points in the NW study area and 190 points in the NE study area 1–3 times. Our findings for the cantus call survey in 2015 were qualitatively similar to those in 2014, with higher detection probabilities earlier in the day, earlier in the season, earlier in the listening period or in a former listening period (Figures 5–6). The call detection rate was 3-fold higher (compared to 2014), but still 3–4 times lower than that for pellet surveys. Comparisons between study areas indicated similar detection rates with the call survey and pellet survey on road-based points, but slightly higher detection rates at off-road points in the NE study area (Table 1). However, this effect was much smaller in stands that are preferred by spruce grouse based on the literature (Figure 8). Based on these findings, a pellet survey was deemed the better approach for a large-scale survey.

2016

Our final sample consisted of 567 survey points organized into 65 survey routes. However, 77 (14%) of the 567 survey points would probably need to be removed or replaced in an operational survey due to significant access challenges (e.g., water crossings, long walks, difficult terrain, etc.). Eighty-two percent of the points were located on dirt roads and traffic was light to none during most (86%) surveys. Based on GIS data, spruce grouse habitat at the 567 survey points comprised, on average, 80% of the cover, with jack pine and black spruce cover types accounting for 38% (range = 0–100%). Upon inspection, 8 plots (1%) were dominated by deciduous or open cover types, but they contained at least some marginal spruce grouse habitat on 1 side of the road (habitat = 1). Fifty-two percent of the points contained $\geq$30% jack pine or black spruce cover types. Thus, the GIS data performed reasonably well in identifying potential survey points.

Spruce grouse pellets were detected at 24% of the survey points, but it varied by ECS section (Table 2). On points where spruce grouse pellets were detected, we counted a mean of 5.3 pellet groups (SD = 6.2); 87% of these points contained roost piles and 24% contained fresh pellets (Table 3). Ruffed grouse pellets were detected at 56% of the survey points where spruce grouse pellets were detected. Unknown pellets (could not be confidently assigned to a species) were detected on 8% of “occupied” points but only 2% of “unoccupied” points.

The probability of detecting spruce grouse pellets was positively correlated with percent cover of jack pine and black spruce habitat (based on GIS data), negatively correlated with complete snow cover (a surrogate for detection probability), and positively correlated with a southwest to northeast spatial gradient (Figure 9). The same model structure best explained variation in pellet-group counts, but uncertainty associated with the mean functions was much greater (e.g., Figure 10). Consistent with anecdotal information, both monitoring metrics suggested spruce grouse were relatively rare in the N. Minnesota Drift & Lake Plains eco-section and more abundant in the Northern Superior Uplands and N. Minnesota & Ontario Peatlands eco-sections (Figure 11). Likewise, when viewed over a smoothed prediction surface, both metrics suggested the relative abundance of spruce grouse increased on a southwest to northeast gradient (Figure 12).

2017

Based on site visits in 2017, we eliminated 266 (19%) survey points from the sampling frame due to wetlands (n = 76), ditches (n = 57), steep topography (n = 58), recent or impending timber harvest (n = 64), private landownership (n = 7), and other reasons such as recent fires or blowdowns (n = 64). We could not scout 260 points because of impassable roads during our visits or time limitations resulting from unsafe travel conditions. Thus, the final sampling frame consisted of 1,160 potential roadside-survey points.

We were able to detect a true decrease in probability of use in $\geq$93% of simulations under the proposed sampling effort across all ECS sections examined. At the section level, we could detect a true decrease in 79% of simulations in the Northern Minnesota and Ontario Peatlands, 91% of simulations in the Northern Minnesota Drift and Lake Plains, and 93% of simulations in the Northern Superior Uplands (Table 4).

We detected spruce grouse pellets at 13 (17%) of 76 points, with more detections on bare ground (11, 14%) than snow covered ground (5, 7%). However, unlike previous years with near complete snow cover, in 2017 many points lacked snow cover (16 points; 21%), only 7 points (9%) had complete snow cover, and the remainder had partial snow cover.

DISCUSSION

We would like to launch an annual, range-wide spruce grouse survey beginning in spring 2018. We propose that this survey, like the ruffed grouse, sharp-tailed grouse, and greater prairie-chicken surveys, will be conducted by cooperating biologists in MNDNR, U.S. Forest Service, County governments, tribal entities, non-governmental agencies, universities, and community colleges. Our intent is for the survey to be completed annually by cooperators and staff to allow for detection of population trends and changes in distribution. During winter 2017-2018, we will visit with cooperators and staff at locations throughout spruce grouse range to train biologists and volunteers in spruce grouse pellet identification and survey methodology.

We should be able to detect large changes in the population of spruce grouse with this survey. Current predictions of climate change suggest that the impending impact on spruce grouse will be large. Johnson (2008) suggested that as long as variation in detectability is small compared to variation in population size, then indices can be useful for monitoring. The intention of the statewide survey is to provide an index of population size that can be used to estimate the trend over time. We also hope to be able to monitor changes in spruce grouse distribution with the survey data.

Ideally, this survey would be conducted as the snow pack is melting in late winter to increase contrast between pellets and snow and to facilitate detection of pellets. Snow cover may become less typical as a result of the warming winters expected with climate change. Surveys can be conducted earlier in the calendar year, if snow cover is lost earlier. However, snow cover is not required to complete the survey; pellets are also visible against the forest floor. We will track snow conditions so that we can incorporate snow cover in the detection function. Climate change will likely affect the optimal timing of many different wildlife surveys.

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LITERATURE CITED

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Table 1. Naïve detection rates for spruce grouse using 2 survey methods in northern Minnesota during springs 2014 and 2015.

Year	Study area	Location[1]	Method[2]	n (total)	n (used)	Prop. with detection[3]	95% LCL[4]	95% UCL[5]
2014	RLWMA	R	Call	530	26	0.05	0.03	0.07
2014	RLWMA	R	Pellet	230	45	0.20	0.15	0.25
2015	RLWMA	R	Call	100	13	0.13	0.06	0.20
2015	RLWMA	OR	Call	100	19	0.19	0.11	0.27
2015	Ely	R	Call	95	13	0.14	0.07	0.21
2015	Ely	OR	Call	95	24	0.25	0.16	0.34
2015	RLWMA	R	Pellet	100	64	0.64	0.55	0.73
2015	RLWMA	OR	Pellet	100	63	0.63	0.53	0.73
2015	Ely	R	Pellet	95	59	0.62	0.52	0.72
2015	Ely	OR	Pellet	95	76	0.80	0.72	0.88
2015	RLWMA	R	P+Add	100	68	0.68	0.59	0.77
2015	RLWMA	OR	P+Add	100	67	0.67	0.58	0.76
2015	Ely	R	P+Add	95	60	0.63	0.53	0.73
2015	Ely	OR	P+Add	95	76	0.80	0.72	0.88
2015	RLWMA	R	C+P+Add	100	70	0.70	0.61	0.79
2015	RLWMA	OR	C+P+Add	100	69	0.69	0.60	0.78
2015	Ely	R	C+P+Add	95	62	0.65	0.55	0.75
2015	Ely	OR	C+P+Add	95	77	0.81	0.73	0.89

[1] Location of survey points: R = road, OR = off-road.
[2] Survey method: C or call = call survey, P or pellet = pellet survey,
Add = additional sightings of spruce grouse at survey points.
[3] Proportion of survey points where spruce grouse or spruce grouse sign were detected.
[4] 95% lower confidence limit of proportion.
[5] 95% upper confidence limit of proportion.

Table 2. Sample statistics and occurrence indices for a survey of spruce grouse pellets at points (pts) in northern Minnesota during spring 2016.

ECSS[1]	No. possible sample pts	Prop. sample	No. pts surveyed	Sample fraction	No. survey routes	Prop. pts pellets detected	Prob. pellets post-adjust[2]	85% LCL[3]	85% UCL[4]
NSU	865	0.46	364	0.42	43	0.297	0.243	0.205	0.294
NMOP	407	0.22	115	0.28	11	0.209	0.173	0.080	0.308
NMDLP	590	0.32	88	0.15	11	0.034	0.036	0.019	0.055
All	1,862	1.00	567	0.31	65	0.238	0.166	0.129	0.207

[1] ECSS = Ecological Classification System Section (NSU = Northern Superior Uplands; NMOP = Northern Minnesota & Ontario Peatlands; NMDLP = Northern Minnesota Drift & Lake Plains).

[2] Mean predicted probability of observing $\geq$1 pellet after adjusting for snow coverage (surrogate for detection), % jack pine and black spruce cover, a spatial gradient (X+Y), and the non-proportional allocation of sample points among ECSS.

[3] Lower 85% percentile confidence limit on mean predicted probability of observing $\geq$1 pellet.

[4] Upper 85% percentile confidence limit on mean predicted probability of observing $\geq$1 pellet.

Table 3. Sample statistics and count indices for a spruce grouse pellet survey at points (pts) in northern Minnesota during spring 2016.

ECSS[1]	No. possible sample pts	Prop. sample	No. survey pts	Sample fraction	No. routes	No. pts pellets detected	No. pts fresh pellets detected	No. pts roost piles detected	Mean pellet grp count[2]	SD[3]	Mean predict pellet-grp count [4]	85% LCL [5]	85% UCL [6]
NSU	865	0.46	364	0.42	43	108	27	98	5.2	6.20	0.64	0.41	1.04
NMOP	407	0.22	115	0.28	11	24	5	19	5.9	6.42	0.90	0.30	3.36
NMDLP	590	0.32	88	0.15	11	3	0	1	3.7	4.62	0.07	0.03	0.17
All	1862	1.00	567	0.3	65	135	32	118	5.3	6.18	0.70	0.32	1.12

[1] ECSS = Ecological Classification System Section (NSU = Northern Superior Uplands; NMOP = Northern Minnesota & Ontario Peatlands; NMDLP = Northern Minnesota Drift & Lake Plains).

[2] Mean pellet-group count (excluding zero counts).

[3] Standard deviation of the mean pellet-group count.

[4] Mean predicted pellet-group count after adjusting for snow coverage (surrogate for detection), % jack pine and black spruce cover, a spatial gradient (X+Y), and the non-proportional allocation of sample points among ECSS.

[5] Lower 85% percentile confidence limit on mean predicted pellet-group count after adjusting for snow coverage.

[6] Upper 85% percentile confidence limit on mean predicted pellet-group count after adjusting for snow coverage.

Table 4. The proportion of simulated 10-year periods of population decline (i.e., the state process; *n* = 200) for which the estimated trend in the index of use from simulated pellet-count surveys (i.e., the observation process) was negative, indicating sufficient sampling intensity. The simulations and estimated trends were based upon the real survey effort and pellet-count index data collected during 2016, including the observed spatial, temporal, and sampling variance.

ECSS[1]	Expected occupancy of survey points in year 1	Index of use in year 1	Mean growth rate in occupancy[2]	Growth rate of simulated populations			Proportion with negative estimated trend
				Min	Median	Max	
NSU	0.587	0.297	0.984	0.951	0.983	1.013	0.93
NMOP	0.676	0.209	0.972	0.936	0.987	1.031	0.79
NMDLP	0.110	0.034	0.935	0.811	0.937	1.081	0.91
All	0.531	0.238	NA	0.950	0.983	1.011	0.94

[1] ECSS = Ecological Classification System Section (NSU = Northern Superior Uplands; NMOP = Northern Minnesota & Ontario Peatlands; NMDLP = Northern Minnesota Drift & Lake Plains).

[2] The mean occupancy values used to simulate the populations corresponded to declines over 10 years of 15%, 25%, and 49% in the NSU, NMOP, and NMDLP sections, respectively.

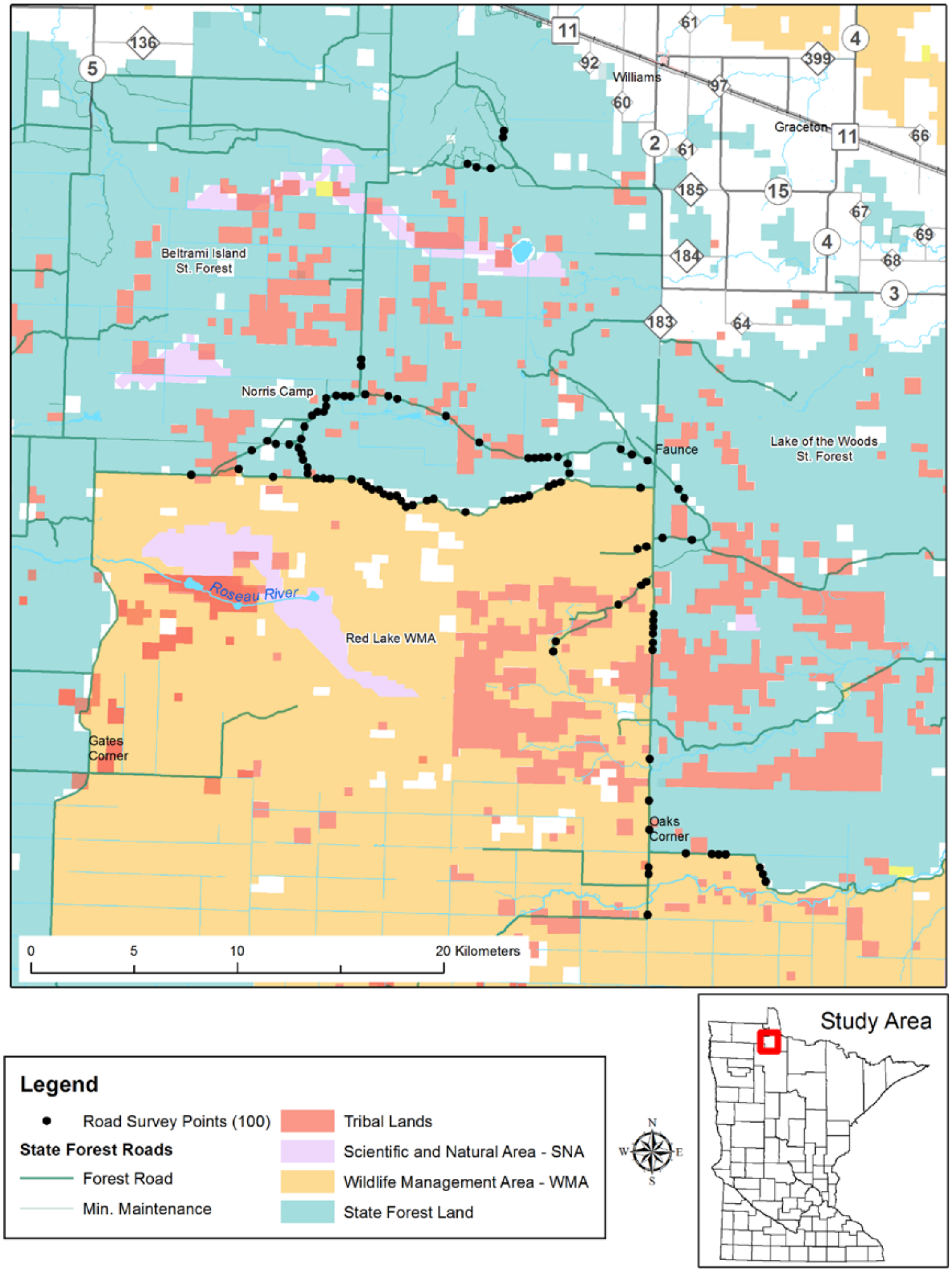


Figure 1. Study area at Red Lake Wildlife Management Area and Beltrami Island State Forest in Minnesota during 2015. The study area was reduced to focus on areas where spruce grouse were detected in 2014. Off-road points were 300 m from road points and alternated sides except when access was prohibited.

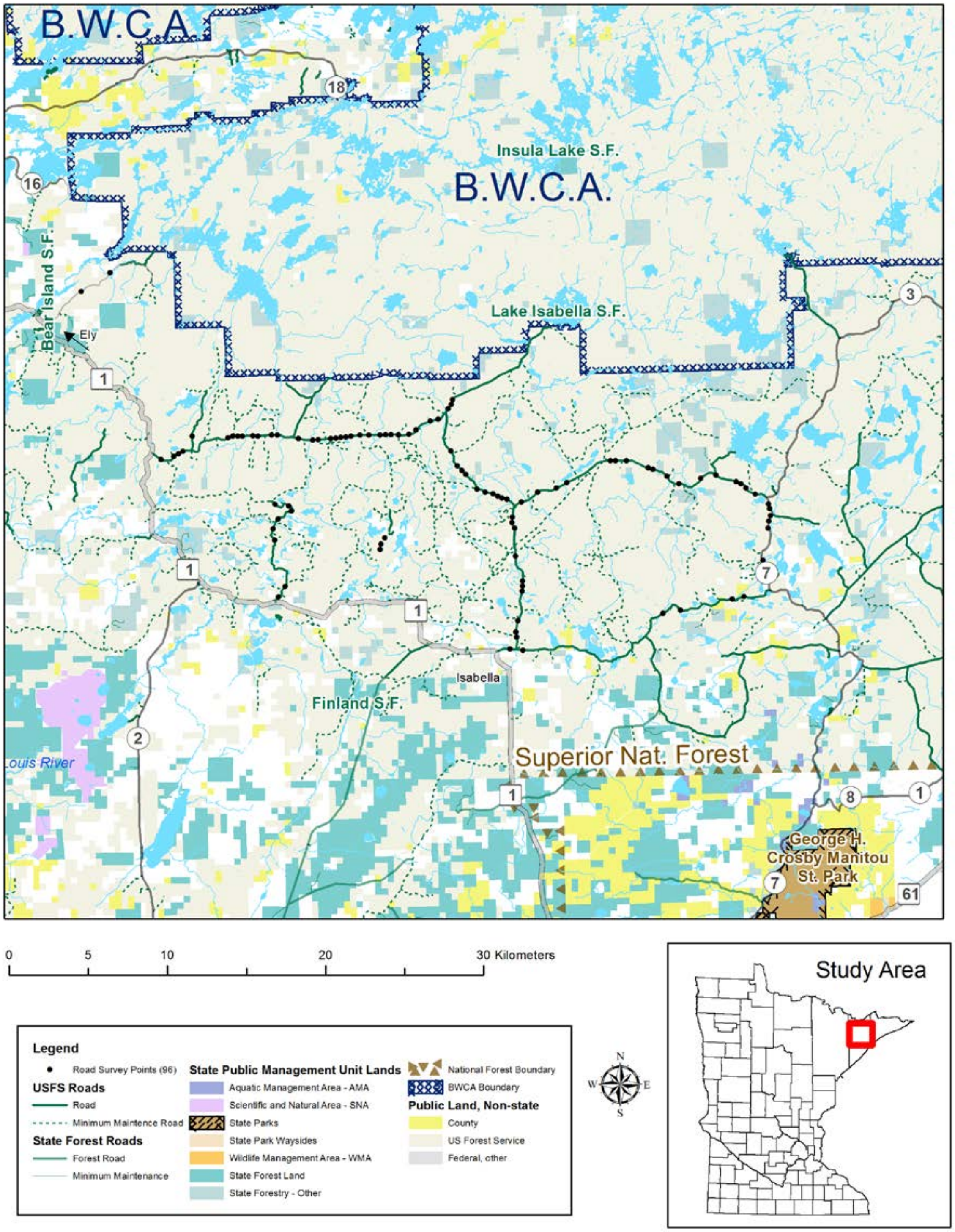


Figure 2. Study area near Isabella, Minnesota (NE) in 2015. Points indicate survey locations along roads. Off-road points were within 300 m of road points and alternated sides except when access was prohibited.

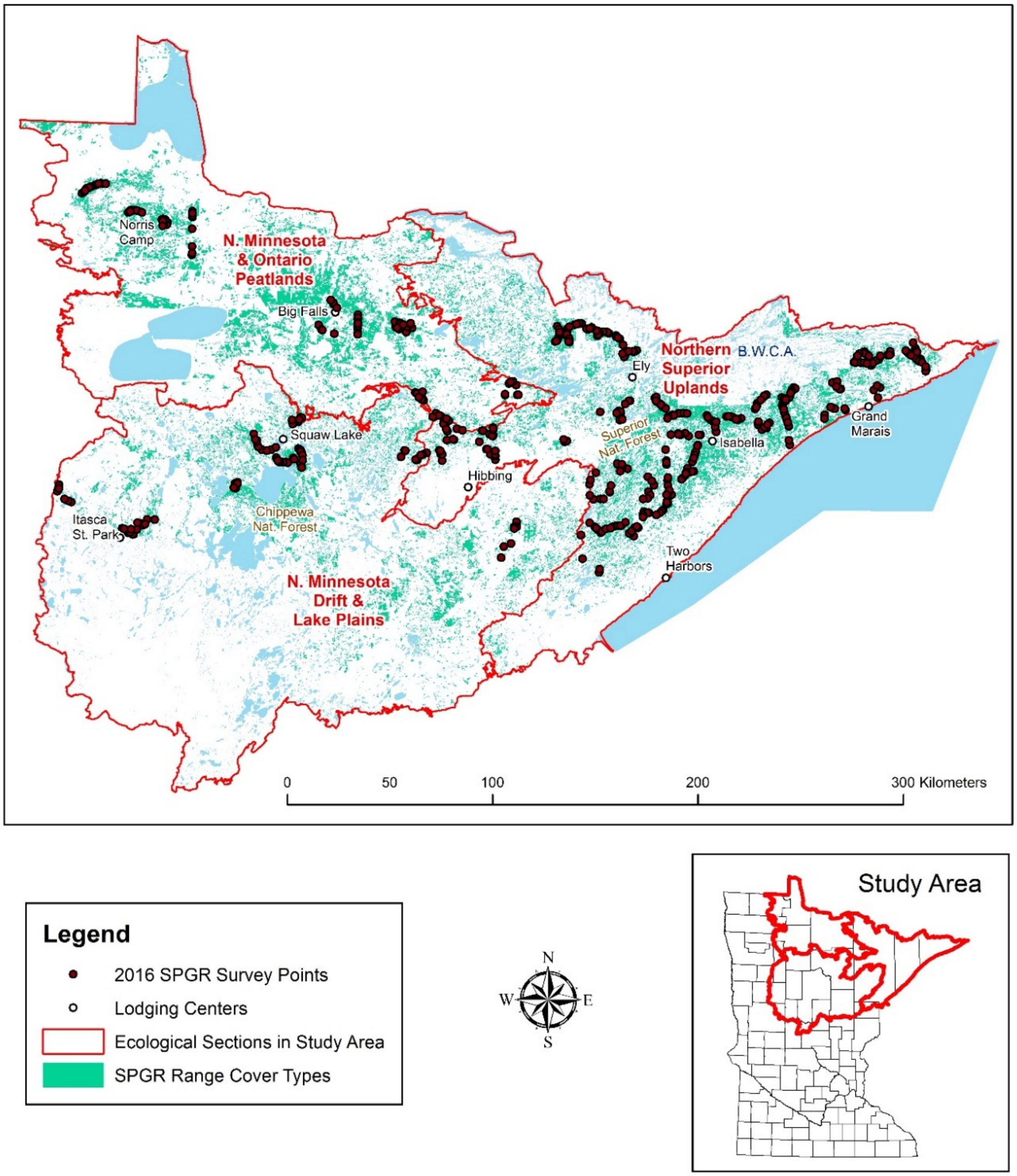


Figure 3. Spruce grouse study area in Minnesota during 2016. Survey points are depicted within the 3 Ecological Classification System sections.

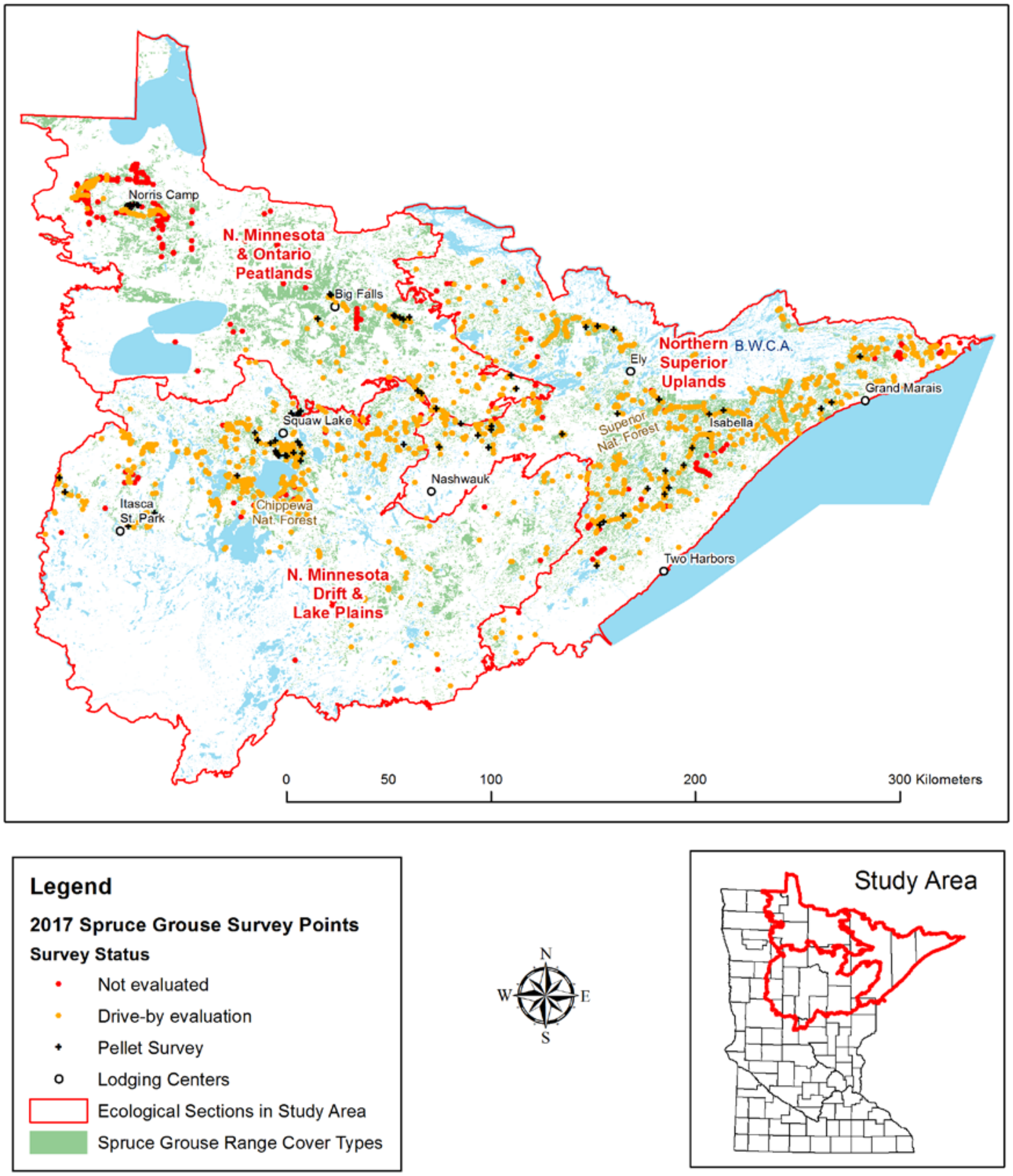


Figure 4. Spruce grouse study area in Minnesota during 2017. Survey points are depicted within the 3 Ecological Classification System sections.

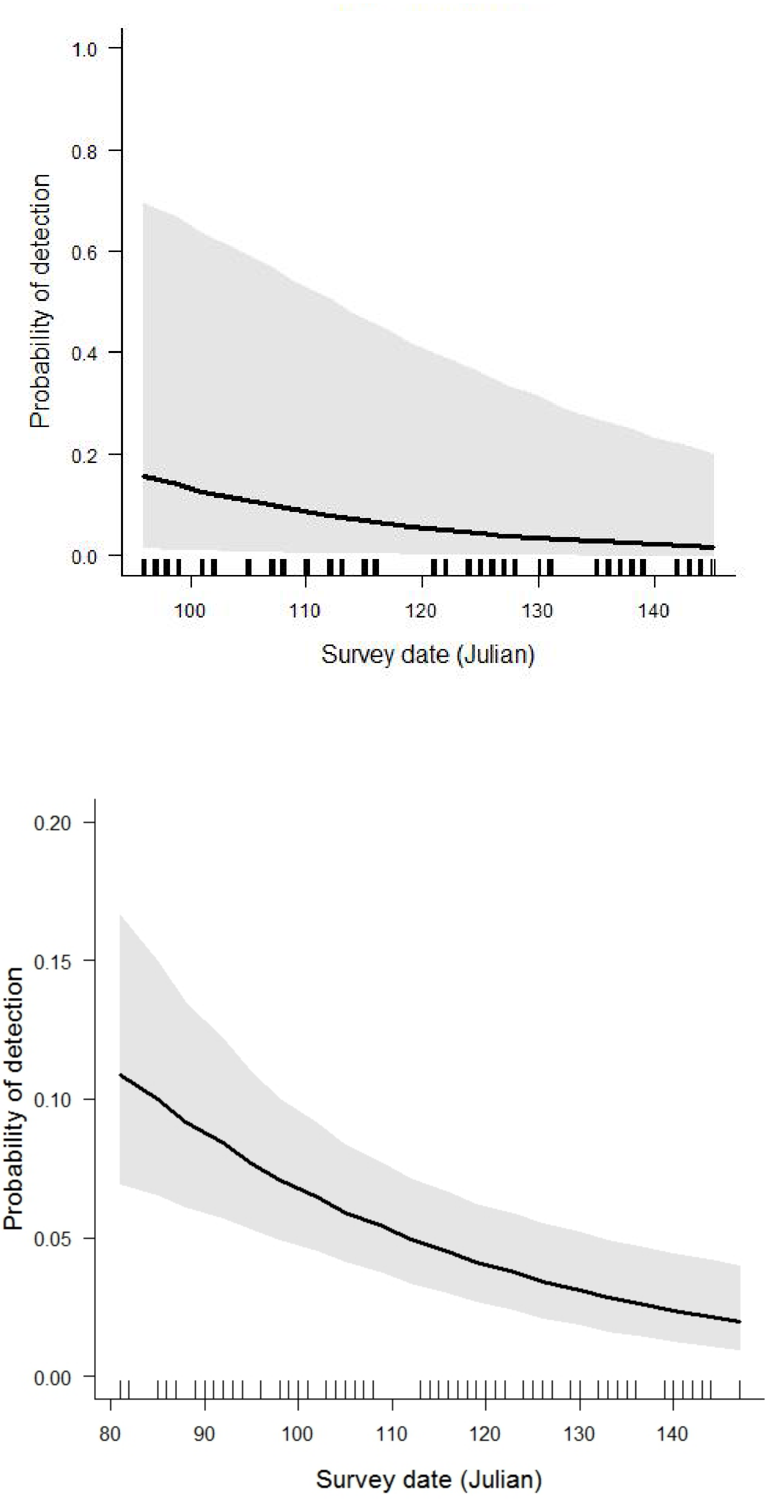
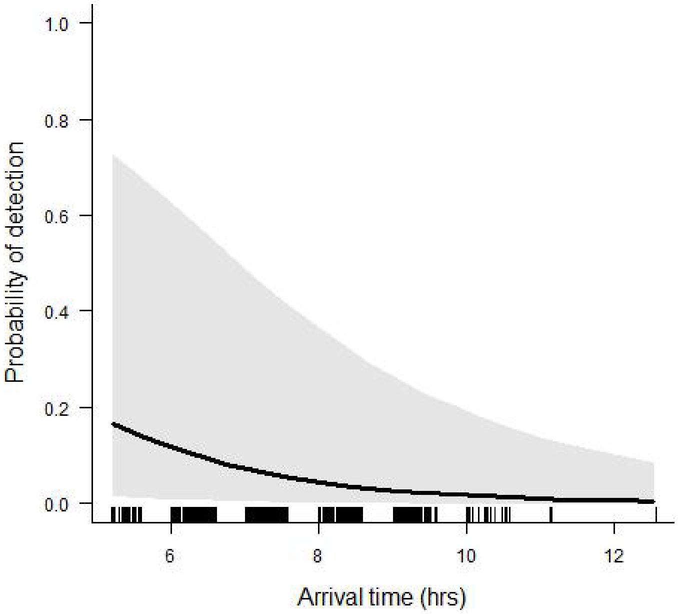


Figure 5. Mean conditional probability of detection (solid line; conditional on spruce grouse being present and available for detection) in each listening interval as a function of survey date at Red Lake Wildlife Management Area and Beltrami Island State Forest in 2014 (top) and both study areas in Minnesota during 2015 (bottom). Gray polygon denotes 95% confidence interval. The "rug" on the x-axis denotes the sample distribution.



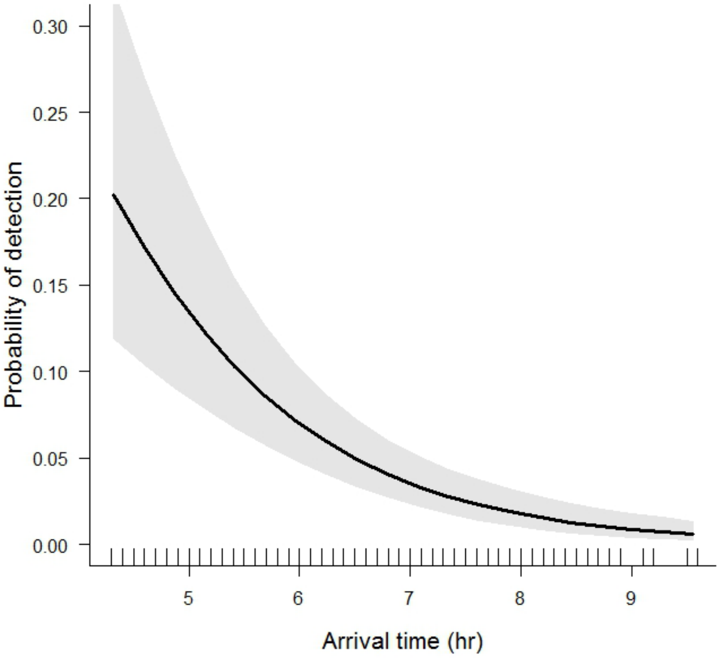


Figure 6. Relationship between spruce grouse call detections and cantus call survey arrival time (i.e., 6 = 0600 hours) at Red Lake Wildlife Management Area and Beltrami Island State Forest in 2014 (top) and in both study areas in Minnesota during 2015 (bottom). Gray polygon denotes 95% confidence interval.

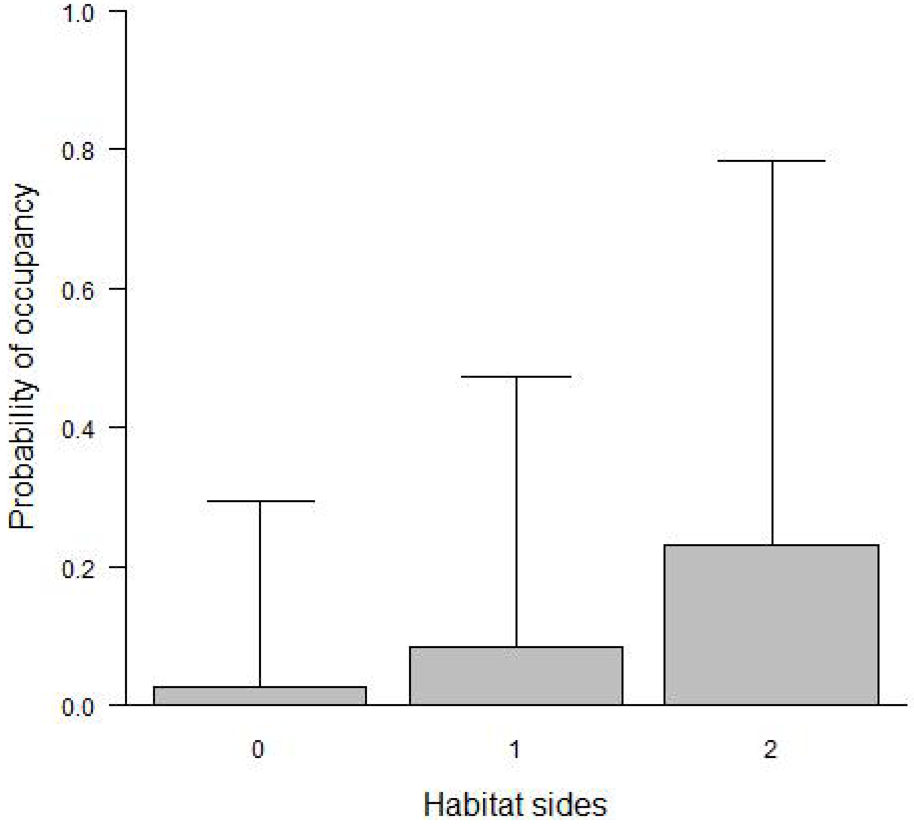


Figure 7. Relationship between the probability of spruce grouse occupancy and the presence of habitat on 0, 1, or 2 sides of the road during cantus call surveys at Red Lake Wildlife Management Area and Beltrami Island State Forest in Minnesota during 2014.

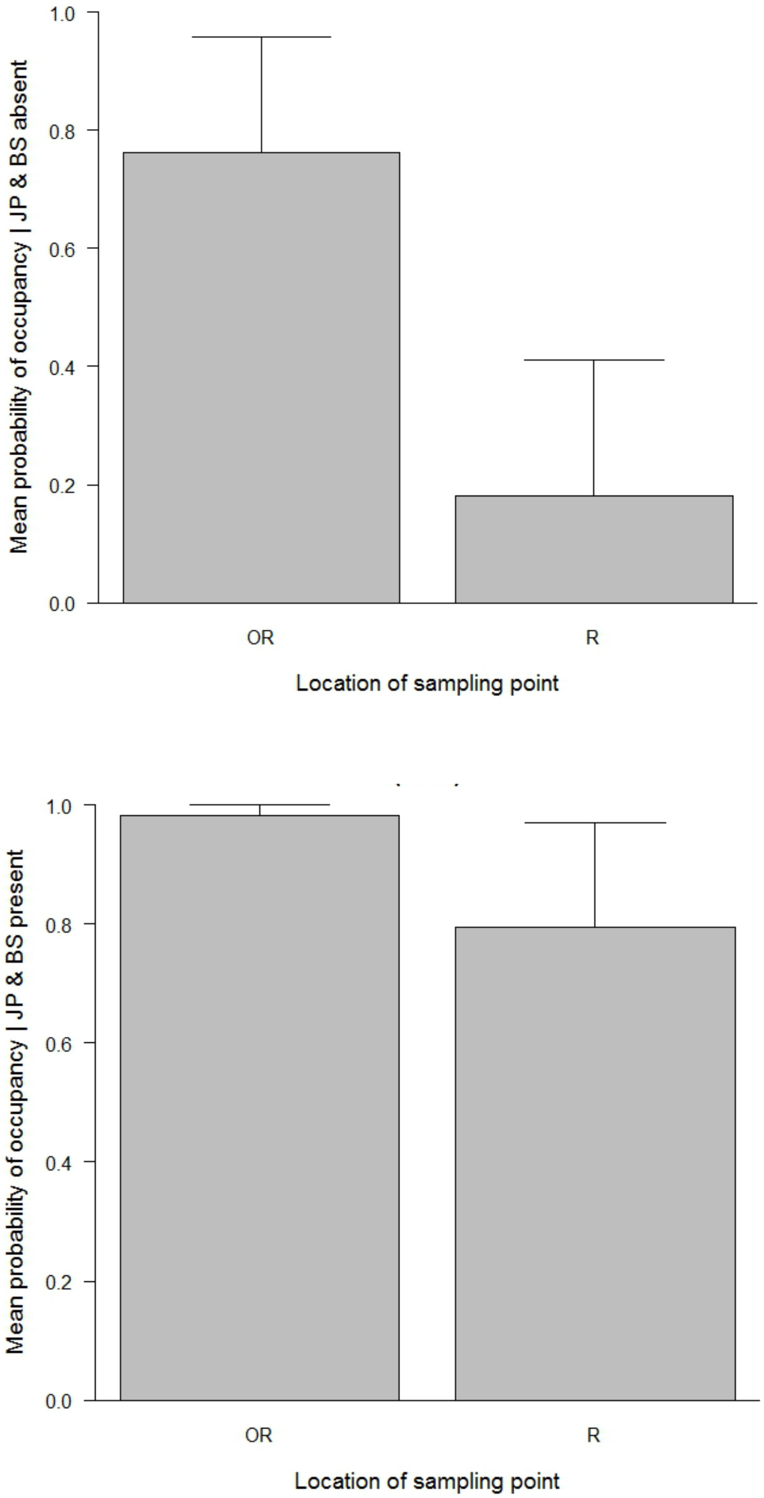


Figure 8. The mean probability of occupancy of spruce grouse at survey points located on roads (R) and off roads (OR) during cantus call surveys at points where jack pine or black spruce were not (top) and were (bottom) present in the Isabella, Minnesota (NE) study area in 2015.

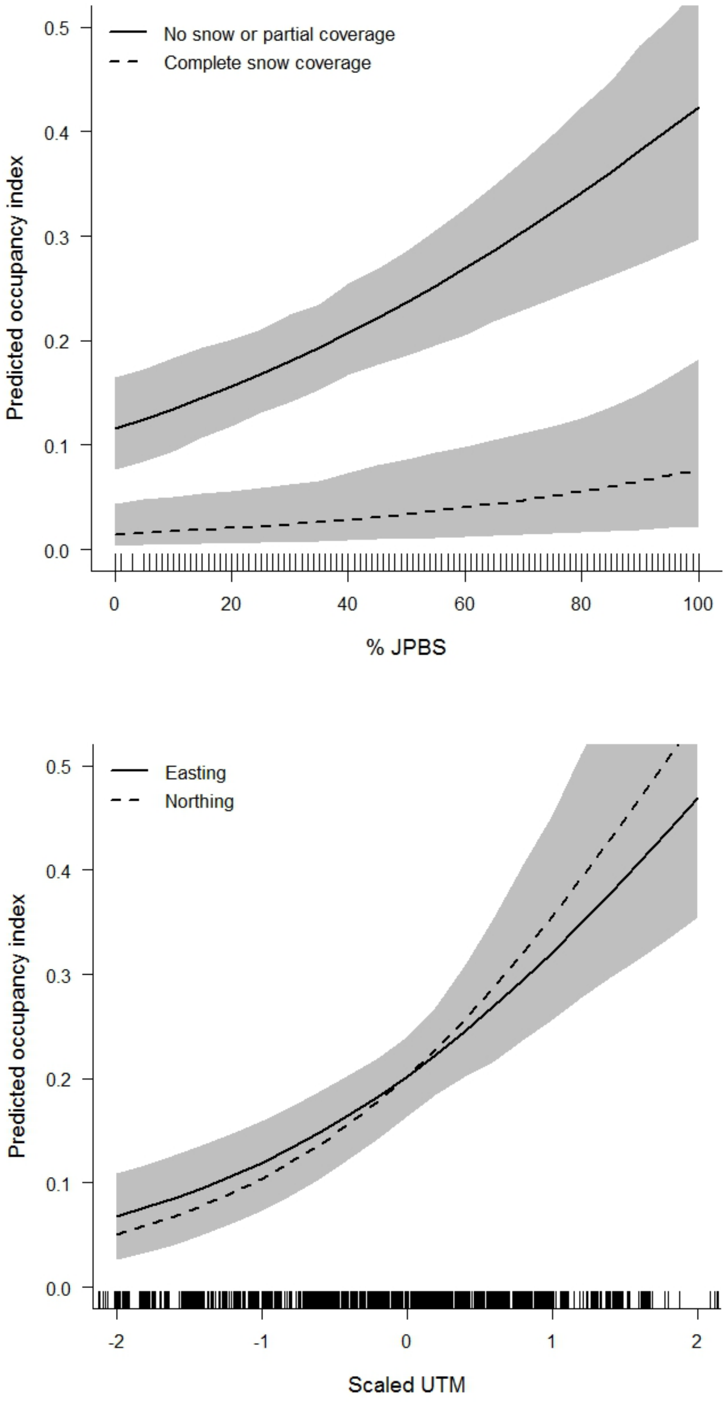


Figure 9. Mean probability of detecting spruce grouse pellets as a function of (A) percent cover of jack pine and black spruce habitats (%JPBS) and snow cover (surrogate for detection probability), and (B) the spatial location of survey points (with other covariates fixed at mean or base values). Figure is based on a generalized linear mixed-effects model fit to pellet-survey data in northern Minnesota during spring 2016.

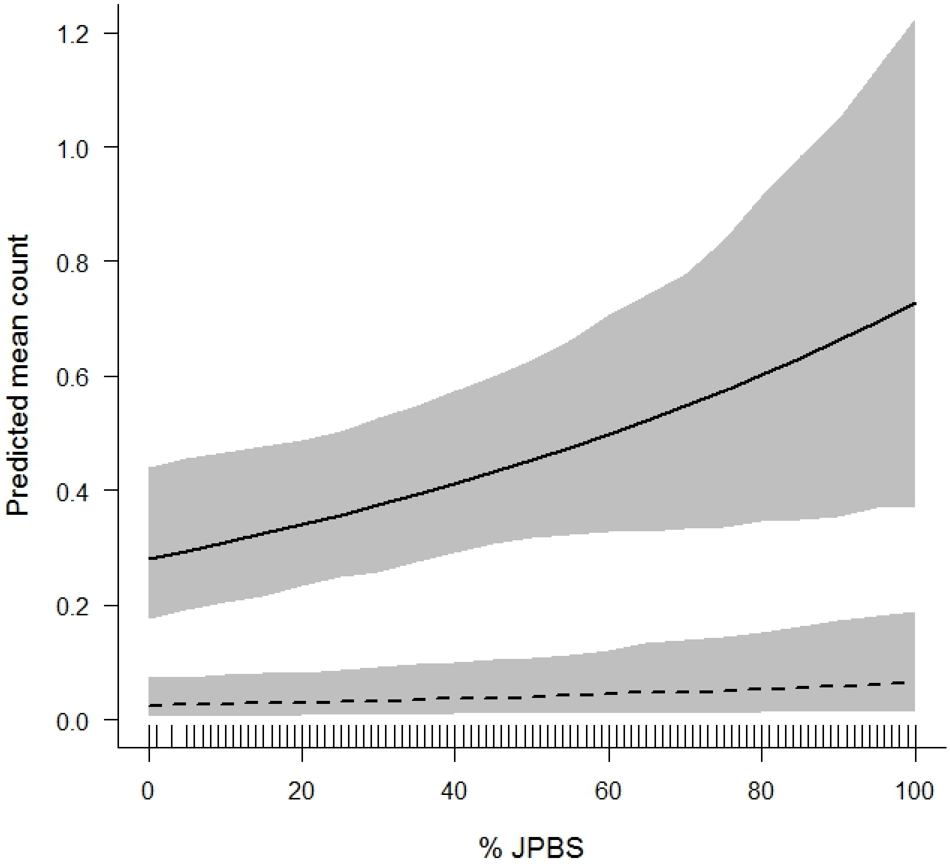


Figure 10. Mean count of spruce grouse pellet groups as a function of percent cover of jack pine and black spruce (%JPBS) habitats and snow cover (surrogate for detection probability). No snow or partial snow coverage is indicated by the solid line and the dashed line represents complete snow coverage. Figure is based on a generalized linear mixed-effects model fit to pellet-survey data in Minnesota during spring 2016.

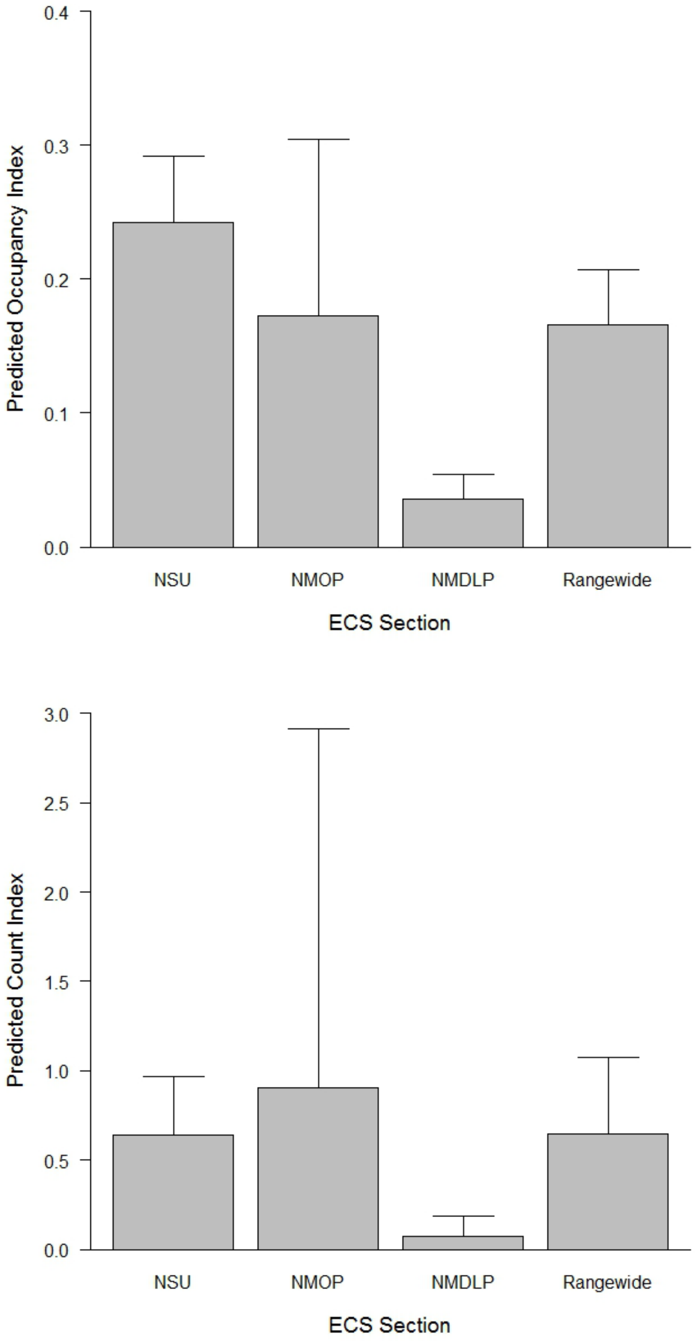


Figure 11. Potential monitoring metrics for spruce grouse in northern Minnesota during spring 2016. Figures are based on the arithmetic mean of model predictions applied to all potential roadside sampling points while holding the categorical predictor snow cover (surrogate for detection probability) to “None or partial.” Ecological Classification System (ECS) sections included Northern Superior Uplands (NSU), Northern Minnesota & Ontario Peatlands (NMOP), and Northern Minnesota Drift & Lake Plains (NMDLP).

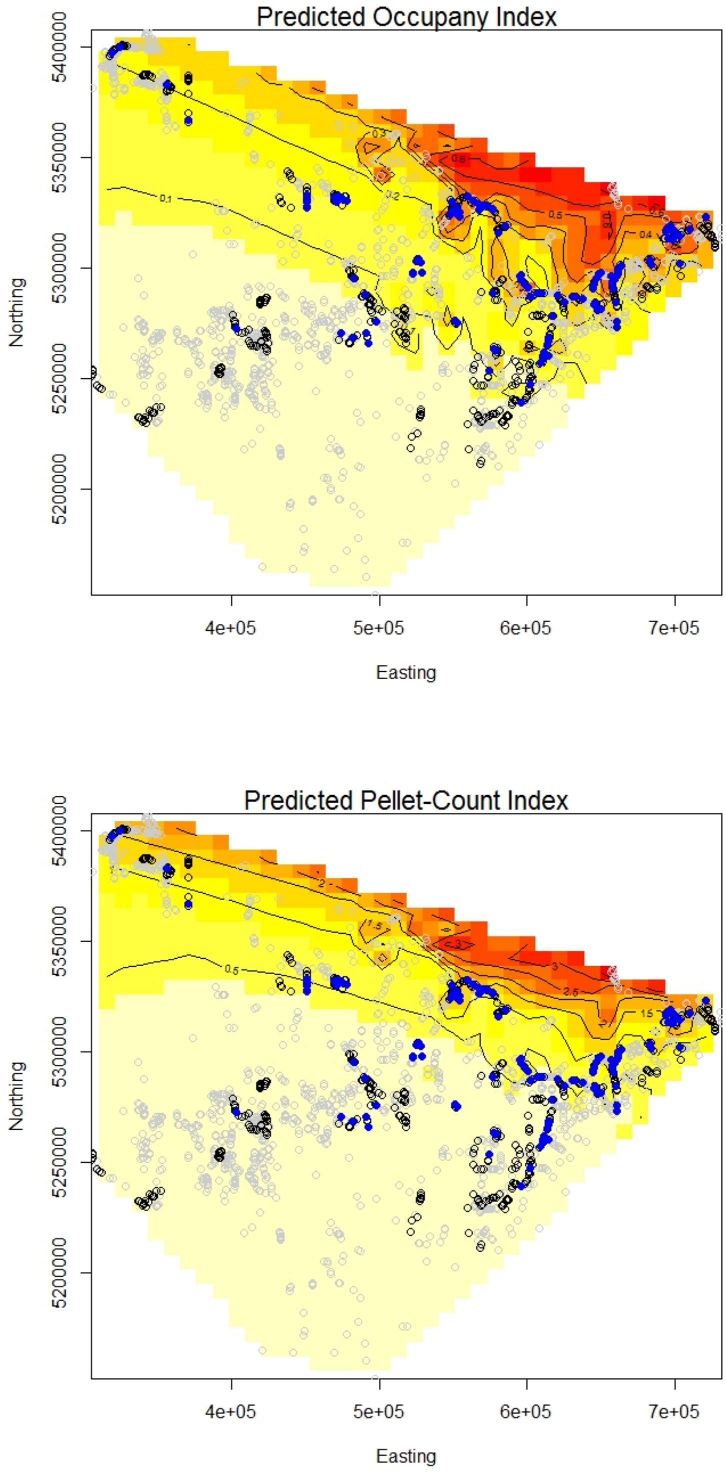


Figure 12. Smoothed prediction surface for spruce grouse monitoring metrics (red = highest; light yellow = lowest predicted index values) in northern Minnesota during spring 2016 based on a bivariate interpolation of model predictions. Contour lines with the highest predicted index values are also depicted.

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

Charlotte Roy, Andrew Gregory[1], Eric Nelson[2]

SUMMARY OF FINDINGS

Sharp-tailed grouse (*Tympanuchus phasianellus*) and greater prairie-chickens (*Tympanuchus cupido*) are area-sensitive species that rely on open landscapes of early successional habitats. Although once abundant and widespread, grassland and brushland habitats today are highly fragmented by agriculture and other human land uses. We used a landscape genetics approach to identify landscape features that impede movement and to identify gaps in connectivity for prairie grouse in Minnesota. With the help of numerous cooperators, we collected 509 prairie-chicken and 831 sharp-tailed grouse samples, which included hunter-submitted wings from 82 sharp-tailed grouse and 52 prairie-chickens. After we eliminated juveniles not sampled again as adults, duplicate samples, and samples with genotyping errors, we were left with a unique genetic sample of 294 prairie-chickens and 451 sharp-tailed grouse, including 367 individuals from the northwest (NW) and 84 individuals from the east-central (EC) regions. Results for prairie-chickens indicated good connectivity in the existing range but further improvements along the Prairie Plan corridor in Norman and Clay Counties would be beneficial. For sharp-tailed grouse, both the NW and EC management regions are genetically diverse and distinct, with high connectivity indicated between them. We cannot be sure whether the gene flow indicated between these regions is best explained by contemporary connectivity or a historical connection that has recently been lost. The population in the EC region shows signs of a recent demographic compression, consistent with surveys that indicate recent declines in population size. Inbreeding is not currently a problem in the areas sampled, but if the population size in the EC region continues to decline or stay small, genetic diversity would be expected to be lost gradually and the population may eventually face inbreeding depression. We recommend increasing the quantity and quality of habitat in the EC region to increase population size and maintain genetic diversity. We provide recommendations about where land management can achieve the greatest impact on genetic connectivity.

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

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greater prairie-chicken as an indicator species for upland prairie and grassland habitat (Minnesota Prairie Plan Working Group 2011). Similarly, for sharp-tailed grouse to move among suitable habitat areas in isolated grassland, brushland, savanna, and peatland habitat patches (Berg 1997), they must traverse 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 geographic place-based information with information about genetic variation within or among populations (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 as measured by genetic connectivity
3. To improve corridor planning and provide guidance to keep connected populations connected

METHODS

Wildlife managers, cooperators, and seasonal technicians surveyed prairie-chickens and sharp-tailed grouse at leks throughout Minnesota in the springs of 2014 and 2015. Feathers lost during male contests, copulations, and as a result of other activities were collected from leks. To maximize the probability of sampling many different individuals, staff and technicians were instructed to spread out the sampling at each lek, sample feathers from discrete locations on the lek, and only collect one sample per location or cluster of feathers encountered. Each sample of feathers, or single feather when necessary to ensure that only one individual was represented, was placed in an envelope and labeled with the lek location (coordinates or Township, Range, Section, and quarter-section information), date, collector name, contents, and species. Information from each envelope was recorded in a database and assigned a unique sample number. Areas underrepresented in 2014 were given greater effort in the spring of 2015. Feather samples from leks were supplemented with samples from hunter-harvested birds in both 2014 and 2015. Wings from harvested birds were aged based on plumage characteristics (Bihrle 1993).

All samples were analyzed at the Wildlife Genetics International Lab in British Columbia. At the lab, DNA was extracted and amplified at 15 microsatellite loci. Microsatellites are highly variable, neutral (non-coding) genetic loci. Recent studies of prairie-chickens and sharp-tailed grouse identified polymorphic microsatellite loci in these species and populations (see citations in Gregory 2011 and Malone 2012). The sex of birds was determined molecularly using techniques such as those in Fridolfsson and Ellegren (1999).

STATISTICAL ANALYSIS

We tested and accounted for the presence of null alleles using Program MicroChecker (Oosterhout et al. 2004). We used Program GenAlEx 6.5 to calculate estimates of genetic diversity (Peakal and Smouse 2012) and to test the Probability of Identity (PI) and the Probability of Identity among siblings (PI-Sibs). PI and PI-Sibs are estimates of the power of the genetic markers to differentiate unique individuals from a population of unrelated individuals (PI) or from a population of siblings (PI-Sibs). GenePop was used for Hardy Weinberg exact tests and to estimate gene flow (F_{ST} and Number of Migrants *Nm*) between the EC and NW sharp-tailed grouse management regions (Raymond and Rousset 1995).

We used Program Structure to implement a Bayesian clustering algorithm to test for genetic isolation first among management zones and then within management zones (Pritchard et al. 2000).[3] More spatially explicit formulations of the clustering algorithm were applied using Package Geneland in Program R (Guillot et al. 2008). Whereas Structure uses a spatially implicit clustering algorithm (you can assign individuals to populations based on locations but not to specific coordinates), Geneland uses map locations of samples to create a Poisson weighting matrix based on Tobler's First Law of Geography (Miller 2004), which essentially assigns individuals to specific locations relative to each other and gives greater weight to samples that are physically closer together (Guillot et al. 2008). The outputs of both Structure and Geneland are a negative log likelihood for a particular number of population clusters and the probability of assignment for each individual to each putative cluster.[4] We used the number of populations and the individuals assigned to each population from Geneland as the putative number of populations for subsequent analyses. Lastly, we used a Standardized Difference Test and a Wilcoxon Test to test for excess heterozygosity, which is a signal of a relatively recent (10-15 generations, or ~15-30 years) population bottleneck (Cornuet and Luikart 1996), and estimated effective population size (N_e) using linkage disequilibrium methods implemented in Program NeEstimator (Waples 2007, Waples et al. 2014).

Spatial Analysis

All analyses were carried out at the spatial extent of the MNDNR greater prairie-chicken or sharp-tailed grouse management regions with the boundaries buffered by 50 km. A 50-km buffer was used because it exceeds the average dispersal distance of most prairie grouse (Johnson et al. 2011; Connelly et al. 1998), completely encompassed the extent of our data, and minimizes possible boundary effects that might occur as a result of an artificially imposed boundary on a dynamic ecological system (Franklin 2009). Restricting the analysis to our region of interest is necessary to reduce the potential for spurious correlation that can sometimes occur with spatial data analysis (Loiselle et al. 2003). For greater prairie-chickens, we conducted a separate, secondary analysis within the extent of the Prairie Plan corridor. For sharp-tailed grouse we

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[3] Program Structure uses a Bayesian clustering algorithm to identify the most likely number of distinct genetic groups, given the observed allele frequencies and levels of linkage disequilibrium within the genetic data. You can choose to include information on the putative population of origin or sampling unit when you run Structure and thereby test the degree to which *a priori* defined populations or management units are distinct genetic populations or subpopulations. Moreover, because Structure uses a Bayesian algorithm it can be implemented in a hierarchical fashion to test for genetic clustering within a local area below the level that would normally be associated with a population or subpopulation level of genetic isolation. This is a useful feature of Structure if you are trying to identify landscape features that are affecting dispersal and gene flow (Pritchard et al. 2000).

[4] You can use the probability of assignment values to identify migrants. A genotype assigned to 1 cluster that was sampled in another cluster is most likely a migrant, genotypes with ~50% assignment to 2 different clusters are most likely offspring of a migrant mating with a local; 25% assignment, with the grandchildren; and so on.

conducted analyses in a hierarchical fashion to assess movement between regions as well as movement and structure within each region.

We used the Multiple Resolution Land Cover Data (MRLCD, usgsmrlcd.org) set clipped to the extent of our study system(s), which included 16 distinct land use classifications (Table 2). We reclassified the land cover and land use data into a resistance surface following the methods outlined in Spears (2010) and Gregory (2011, Table 2). We also acquired data sets for linear features (i.e., highways, railroad lines, and power lines) and anthropogenic disturbance intensity [Wildlife Conservation Society (WCS) 2005]. An index of anthropogenic influence or disturbance intensity was determined by WCS (2005) using information on population density, land use and infrastructure, and human accessibility to create the Global Human Footprint Dataset. These data sets were combined following the methods used by the Washington Connected Landscape Project (waconnected.org; McRae 2006), to produce 8 putative resistance surfaces (Table 2 GRPC, Table 3 STGR, Figure 1).

We used Circuitscape to calculate a metric of functional isolation, called resistance, between each sample location (McRae 2006). Resistance values create a relative index of hypothesized landscape interactions and influences on species movements, and therefore are a hypothesis of how the landscape influences observed genetic structure (Storfer et al. 2007). Resistance values are relatively arbitrary and are typically assigned based on expert opinion or via the use of a species distribution model; pros and cons exist for both approaches (Spears 2010). Here we used an expert opinion optimization based on the literature and knowledge of each species biology. Each of the 8 resistance surfaces is a different hypothesis about how landscape attributes influence prairie-grouse movement and viability on the landscape (McRae et al. 2008; Spear et al. 2010). Hypothesis 1 (H1) predicts that connectivity is a function of land cover and land use with primary habitat (i.e., grasslands, wet meadows, or shrublands) being highly suitable land cover and land use types of low resistance (Tables 2, 3). H2 predicts that connectivity is a function of the amount of grassland within an area where areas of higher grassland availability are of lower resistance and high suitability. H3 is similar to H2 but predicts that grassland suitability is reduced by the presence of cultivation. H4 predicts a similar response to H2 and H3 but also includes an interaction of agriculture and grassland land cover. H5 predicts that genetic structure is a function of avoidance of linear features on the landscape. H6 predicts that land cover and the intensity of anthropogenic disturbance interact to drive genetic structure. H7 predicts the same as H6 but also includes avoidance of linear features. H8 predicts that all attributes previously mentioned combine to influence genetic structure. Isolation by distance is a null hypothesis that tests the assumption that gene flow is not related to land use or its configuration, but simply to physical distance among spatially structured subpopulations. The Reverse model tests the counterintuitive hypothesis that highly modified and disturbed landscapes are highly beneficial to prairie grouse (Tables 2, 3). This model is an important control because if it does not perform poorly relative to the other models, we clearly do not understand the system sufficiently to be using an expert opinion optimization. However, if it does perform poorly relative to the other models, we can be moderately confident that our assigned resistance values provide useful insights about how the landscape is impacting these species, and we can therefore use the results of the analysis to help guide management actions. Collectively, these model formulations allow us to test the influence of 8 hypothesized land cover and land use interactions of greater prairie-chickens or sharp-tailed grouse movement and gene flow.

We used a Partial Canonical Correspondence Analysis (PCCA, also controlling for Euclidean distance, Balkenhol et al. 2009) to ordinate genetic differentiation among subpopulations along explanatory variable axes. In this way, the strength and direction of genetic isolation can be examined, as well as identifying which attributes are driving population subdivision and isolation

(Cushman 2006). The PCCA was implemented using the vegan package in Program R (Oksanen

et al. 2007). We also used PCCA to ordinate the population by cluster based on landscape attribute data. The PCCA included 5 landscape predictor values (i.e., MRLCD resistance surface, Human Footprint, Linear Features, %Grassland, %Agriculture, and %Agriculture×%Grassland; Table 4).

Lastly, we mapped F_{IS} values for both species in ArcInfo 10.4. F_{IS} values are an indication of how much inbreeding is occurring at a sample location or within a population. Values range 0-1, and the higher the value the more inbreeding is occurring (Frankham et al. 2002). We performed a HotSpot analysis using the Getis Ord Gi* procedure in ArcInfo Spatial Analyst Tools to test for significant high×high and low×low clustering of F_{IS} values (Getis and Ord 1992). This analysis identifies areas where the genetic data suggest that either sharp-tailed grouse or greater prairie-chickens are potentially genetically isolated or where movement is strongly structured by landscape characteristics (F_{IS} HotSpots). This can be useful in conservation planning because it can identify areas where targeted investment in habitat improvements can have the largest population-wide benefits for the species by enabling connectivity.

RESULTS

Greater Prairie-Chickens (Statewide)

With the 294 individual greater prairie-chicken genotypes used in this analysis, we had adequate discriminatory power to identify individuals (Prob Identity = 1.0×10^{-20}; Prob Identity Sibs = 2.7×10^{-7}) and identify population structure. We found no indication of null alleles among loci, except for locus SGCA6. SGCA6 was also not at Hardy Weinberg Equilibrium or at Linkage Disequilibrium (Table 1), which means that this marker was not appropriate for use in this analysis. Consequently we censored SGCA6 from the analysis.

Genetic diversity within the greater prairie-chicken population was high (H_O= 0.76 ± 0.04; AR = 35 ± 18.75). The effective population size of greater prairie-chickens was large (Linkage Disequilibrium N_e = 243.1-*infinity*) and a population bottleneck was not indicated (Wilcoxon Test for Excess Heterozygosity P = 0.34; Standardized Difference Test P = 0.08). Collectively, these observations suggest that the greater prairie-chicken population is panmictic and possibly expanding.

Analysis with Program Structure indicated greatest support for K = 2 populations [-LN(K) = 17,542.1 $\pm$ 511.9] with correlated allele frequencies and high genetic exchange (F_{ST} = 0.004). Analysis with Geneland also indicated that greater prairie-chickens were likely 2 highly connected but distinct subpopulations [-LN(K) = 15,344.3 ± 90.567; F_{ST} = 0.0044, F_{IS} Cluster 1= 0.042; F_{IS} Cluster 2 = 0.047; Figure 2]. Collectively, these results indicate a weakly structured population with exchange among genetic clusters.

Based on the PCCA, the most supported resistance landscape is depicted by H6 (Table 2), which indicates that greater prairie-chicken movement across the landscape is partially influenced by land use and land cover and also by anthropogenic disturbance (Table 4). Entering this resistance surface and greater prairie-chicken sample locations into Program Circuitscape yields a connectivity landscape that identifies regions important for maintaining greater prairie-chicken connectivity (Figure 3a). When we overlaid the results of this connectivity analysis with the Prairie Plan corridor, we identified critical gaps in corridor coverage to ensure connectivity of greater prairie-chicken genetic clusters (Figure 3b).

Lastly, HotSpot Analysis indicated the areas where significant high×high (HotSpots) and low×low (ColdSpots) F_{IS} value clustering occurred. With 95% confidence we identified a single

HotSpot near the northern extent of the greater prairie-chicken range in Minnesota (Figure 4) and a ColdSpot near the southern extent of the greater prairie-chicken range (Figure 4). In the context of this analysis a HotSpot indicates an area with low dispersal and genetic exchange, an isolated area; whereas a ColdSpot indicates an area of high genetic exchange, or panmixia (Figure 4). These identified regions roughly correspond to the genetic clusters identified by Geneland (Figure 2).

Sharp-Tailed Grouse (Statewide)

We obtained 84 unique sharp-tailed grouse genotypes from the EC management region, and 367 individual genotypes from the NW management region. We had adequate discriminatory power (Prob Identity = 5.2×10^{-20}; Prob Identity Sibs = 3.8×10^{-7}) to determine localized population genetic structure, gene flow among management regions, and sufficient resolution to link genetic isolation to physical attributes of the landscape. Across both regions, genetic diversity was high ($H_O = 0.77 \pm 0.03$; AR = 29.4 ± 11.9). Bayesian clustering algorithm within Program Structure found greatest support for K = 2 populations, with each management region being a distinct genetic cluster [-LN(K)= 27,781.11 ± 581.8]. However, the populations were admixed and exhibited a high degree of genetic exchange among regions ($F_{ST} = 0.003 \pm 0.001$), suggesting gene flow was recently or is occurring across Itasca county between leks sampled in Aitkin and St. Louis Counties and leks sampled in Koochiching County. To confirm this, we also tested for gene flow across Cass County between grouse in Aitkin and Beltrami counties ($F_{ST} = 0.029 \pm 0.004$), which further supports the notion that gene flow was or is occurring across Itasca county. As previously mentioned, because Structure is a Bayesian clustering algorithm it can be used hierarchically to explore possible latent subpopulation-level clustering. By restricting the analysis to just individual genotypes sampled in Aitkin, St Louis, and Koochiching counties and eliminating the population of origin as a starting point, we allowed Structure to converge on the number of populations in just this subset of sampled leks. Structure again found greatest support for K = 2 populations with correlated allele frequencies and admixture [-LN(K=2) = 12,316 ± 894.2]. However, this was only moderately greater than support found for K = 1 population with admixture and correlated allele frequencies [-LN(K=1) = 16,821 ± 2,113.4]. Models not assuming correlated allele frequencies and panmixia were not supported [-LN(K) = 34,865 - 49,456.9]. Collectively, these analyses suggest that the 2 management regions are 2 distinct genetic clusters of sharp-tailed grouse connected by current or recent migration between regions, most likely across western Itasca and/or eastern Cass counties (e.g., red/orange areas; Figure 5).

Analysis with PCCA to determine the degree to which landscape features were driving genetic structure between sharp-tailed grouse management regions included 5 landscape predictor variables (i.e., MRLCD resistance surface, Human Footprint, Linear Features, %Grassland, %Agriculture, and a %Agriculture×%Grassland interaction) sampled across Aitkin, St. Louis, Carlton, Cass, Itasca, and Koochiching counties. PCCA was able to explain 61% of the variance in genetic clustering with the first 2 components (Dominant Eigenvalue = 0.33, Secondary Eigenvalue = 0.28, Table 5). Based on these results, the most appropriate landscape resistance model would be H1 or H4 (Table 3), which predicted that populations of sharp-tailed grouse are structured by land cover or by the amount of agriculture and grasslands or wet meadows on the landscape. We elected to present the predictions arising from use of H4 in our Circuitscape connectivity analysis (Figure 5), but predictions arising from use of H1 in Circuitscape indicate more connectivity between the NW and EC regions and are thus less conservative. The results of the connectivity analysis showed limited connectivity between the management regions, but connectivity exists to the west and outside of the management regions in Cass County (Figure 5). Because there is limited connectivity between regions we elected to analyze population- and landscape-genetic attributes of sharp-tailed grouse within each management region.

Sharp-Tailed Grouse (EC region)

Overall the EC region has high genetic diversity (AR = 18.27 ± 5.07; H_O = 0.771 ± 0.04; H_E = 0.779 ± 0.04), and the population-wide inbreeding coefficient (Weir and Cockerham 1984) was low (F_{IS} = 0.017 ± 0.0008). This suggested that these 84 samples were collected from a large outbred population with little genetic evidence for inbreeding. Under an infinite alleles model, we detected a significant excess in heterozygosity using the Standardized Difference Test (P = 0.005) and the Wilcoxon Test (P = 0.002). Moreover, there was a significant right mode shift in allele frequency (P = 0.03). Linkage disequilibrium methods for assessing effective population size indicated an effective population size of 224-771 (95% confidence interval).

Analysis of the EC region with Program Structure indicated greatest support for 3 admixed populations [-LN(K) = 4,755 ± 543.5] with relatively high genetic exchange among them (Avg. Pairwise F_{ST} = 0.06). However, when we mapped individuals based on population assignment, we observed that the population clusters themselves are admixed and not localized (Figure 6). Taken in conjunction, the observed excess in heterozygosity, low inbreeding coefficient, and strong signal of genetic population structure with high admixture of the assigned clusters suggest that the EC region has undergone a recent demographic compression, or bottleneck (Cornuet and Luikart 1996, Luikart et al. 1998a).[5]

To better understand drivers, or lack thereof, of observed population structure we used the hypothesized landscape resistance and suitability model of H8 in a PCCA. We used H8 because it approximates a "full model," and given the relative panmixia suggested by the Structure Analysis, we wanted to maximize our ability to detect any anthropogenic influences on population structure. We orientated the analysis using a discriminant function maximizing the Mahalanobis Distance between explanatory variables. This technique essentially reduces the number of explanatory variables needed by creating synthetic components that each explain the most variance possible. We were able to explain approximately 99% of the variance in the data with the first 2 eigenvectors (Dominant Eigenvalue = 0.87; Secondary Eigenvalue = 0.12). We found that the amount of primary habitat and an interaction between primary habitat and agriculture best explained the observed population clusters (Table 6). However, the confusion error matrix suggested that this analysis had relatively low discriminatory power because it was able to classify only 42% of the sampling locations into the correct genetic clusters based on habitat attributes or land cover and land use attributes. Lastly, the Getis Ord GI* HotSpot analysis of inbreeding using sample location F_{IS} values was non-significant, and we detected no Hotspots or ColdSpots.

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[5] Populations lose neutral allelic diversity due to random genetic drift at a rate of approximately $1/2N_e$, where N_e is the effective population size of the population in question (Hartl and Clark 2007). Thus, we expect small populations to have low allelic diversity and that a population bottleneck that reduces N_e will eventually result in a genetically depauperate population (Lynch et al. 1995; Madsen et al. 1996; Cristescu et al. 2010). However, for a few generations immediately following a population bottleneck, the newly bottlenecked population will have an excess of rare to moderate frequency alleles relative to what would be expected given its smaller population size (Luikart and Cornuet 1998; Luikart et al. 1998a). This is because drift is a random process that probabilistically will affect extremely low and high frequency alleles most strongly (Luikart et al. 1998b). The result is a characteristic right mode shift in the frequency of alleles, meaning that a population will have a higher frequency of occurrence of low to intermediate frequency alleles than expected. This has become a classic and powerful test for a population bottleneck (Luikart et al. 1998b; Luikart and Cornuet 1998). A related test for a bottleneck is an excess in heterozygosity, or population-wide estimates of heterozygosity being larger than expected by chance (Cristescu et al. 2010). Again this indication of a bottleneck is only observable for a few generations following a population bottleneck during which time the surviving population has allele frequencies and heterozygosity characteristics of its formerly larger N_e (Luikart et al. 1998b). As the population persists at its new smaller size, drift will remove genetic diversity eventually resulting in the expected genetically depauperate population (Madsen et al. 1996; Hartl and Cark 2007). From a management standpoint, this

suggests a short-lived opportunity during which time recovery of the population could preserve the majority of the genetic diversity found in the pre-bottlenecked population (Bijlsma et al. 2000; Amos 2001; Crisecsu et al. 2010).

Sharp-Tailed Grouse (NW region)

Overall the NW has high genetic diversity (AR = 40.6 ± 23.4, H_O = 0.77 ± 0.04, H_E = 0.80 ± 0.03), and the population-wide inbreeding coefficient was low (F_{IS} = 0.062 ± 0.003). Under an infinite alleles model, we detected a significant excess in heterozygosity using the Standardized Difference Test P = 0.007, but not with the Wilcoxon Test P = 0.07). There was not a significant mode shift in allele frequency (P = 0.99). Linkage disequilibrium methods for assessing effective population size indicated an effective population size of N_e = 185.6-*infinity*. Analysis of the NW subpopulation with Program Structure indicated greatest support for 5 admixed populations, or genetic clusters [-LN(K=5) = 18,346.1 ± 879.9] with relatively limited genetic exchange among them (Avg. Pairwise F_{ST} = 0.756 ± 0.04, Figure 7).

The PCCA analysis was not able to accurately classify the data, as indicated by being able to explain only 39% of the variance with the first 2 eigenvectors (Dominant Eigenvalue = 0.28, Secondary Eigenvalue = 0.11, Table 7). The discriminant analysis ordination also failed to accurately distinguish genetic clusters because we were able to assign only 34% of the sample sites to the correct genetic cluster. Due to the failure of the PCCA to adequately discriminate genetic clusters based on landscape variables, we did not proceed with a landscape connectivity analysis via Circuitscape.

The Getis Ord Gi* HotSpot Analysis indicated both significant high×high clustering of F_{IS} values (HotSpots) and significant low×low clustering of F_{IS} values (ColdSpots, Figure 8). The ColdSpot near Koochiching County along the eastern edge of the NW region suggested more movement than expected at random. We also identified a potential ColdSpot along the western edge of the region near Kittson County. There was also a HotSpot in the center of the western half of the NW along the border of Pennington and Marshall counties, indicating relatively isolated sharp-tailed grouse populations with limited gene flow into the system or among sample locations within that region (Figure 8).

DISCUSSION

Greater Prairie-Chickens (Statewide)

Our analyses suggested that greater prairie-chickens are most likely a single, large, panmictic population with some subpopulation-level structure starting to develop, or developing along the fringe of an expanding range. We found no evidence of inbreeding depression, and our estimates of the effective population size include infinity, both of which are encouraging and suggest that the population was stable, and possibly expanding. A stable population has also been indicated in recent survey data, after declines coincident with losses in CRP enrollments after 2007 (Roy 2016a).

By correlating observed gene flow with land cover and land use data we were able to evaluate the degree to which land cover and land use contributed to greater prairie-chicken genetic structure. The little population structure that does exist is likely due to an interaction between decreasing grassland and increasing cultivated agriculture. Mapping these connectivity arcs with the Prairie Plan corridors revealed 2 potential gaps in prairie corridor coverage that may need to be addressed for the long-term management of greater prairie-chickens (Figure 3).

One area of management concern for the greater prairie-chickens is the observed lack of connectivity through Norman County (Figure 4). Our analysis suggested that greater prairie-chickens in Polk County are separated from the birds in Wilkin and Ottertail Counties. This conclusion is further supported by the Geneland Analysis of genetic clustering. A second area of potential management concern is the HotSpot band identified in Clay County. The areas south of this band are a ColdSpot, or an area of high gene flow and movement. There are 2 possible explanations for the existence of this band. First, it could have occurred if our sample contained relatively few inbred or highly related birds from 1 or 2 leks (i.e., an artifact of sampling). Alternatively, the pattern is real and is related to landscape fragmentation and management. Upon further investigation, this band of ~200 km^2 contained samples from 15 unique birds (12 males, 3 females) from 5 different leks, which is sufficient sampling to support the conclusion that the observed pattern is real and *not* a sampling artifact. Based on our landscape connectivity model, cultivation is 6-7 times less permeable to movement than is rangeland land cover, and heavily human-modified landscapes are 22-133 times less permeable to movement than range (Table 2). The 200-km^2 block where we detected restricted movement has a median human footprint value of 41 (which corresponds to a moderately high level of disturbance) and is 56% cultivated agriculture and 34% grassland land cover types. Consequently, the existence of this band may be a concern, as it suggests that high movement south of this band is occurring, likely due to marginal habitat conditions to the south.

Sharp-Tailed Grouse (Statewide)

Our analysis suggested that each of the 2 sharp-tailed grouse management regions is a distinct genetic cluster. However, some gene flow is currently or was recently occurring between the EC region and the eastern end of the NW region, and these regions are not genetically differentiated from each other. Thus, management actions within one region may influence population dynamics of the other region. However, because each management region is a distinct genetic cluster, and contemporary gene flow is uncertain, we make separate management recommendations for each region.

Sharp-Tailed Grouse (EC Region)

The possibility of inbreeding depression is a concern in prairie grouse, because of evidence for inbreeding depression in an isolated population of prairie grouse in Illinois (Westmeier et al. 1998, but see Mussman et al. 2017). Given recent demographic data from the EC region suggesting that the sharp-tailed grouse population within that region is declining (Roy 2016b), we were interested in testing for low genetic diversity and inbreeding depression in this region. Contrary to expectations in a population undergoing inbreeding depression, the EC region exhibited high genetic diversity, a low inbreeding coefficient, significant excess in heterozygosity, a right mode shift in allele frequency (Luikart et al. 1998a), and relatively large N_e size relative to the sample size. Moreover, analysis of genetic structure within this region indicated 3 mixed genetic clusters. These results are inconsistent with genetically clustered groups co-occurring on the landscape. However, given the previously observed excess of heterozygosity and that Structure has a proclivity to weight genetic cluster assignments by the co-occurrence of rare alleles (Pritchard et al. 2000), confusing Structure results might be expected within either a rapidly declining or rapidly expanding population (e.g. after a founding event, following a catastrophic population crash, or in the recipient population after a translocation). In light of the demographic observations, the most probable explanation is that the EC sharp-tailed grouse population is not experiencing inbreeding depression, but is in the process of going through a population genetic bottleneck as a result of a relatively recent and rapid reduction in population size (Luikart et al. 1998a). However, we acknowledge that our conclusions relate only to those areas sampled for the analysis; we did not have samples from Pine and Kanabec counties, where population losses have been most extreme.

Population bottlenecks are a concern for sharp-tailed grouse because they have a lek mating system characterized by repeated female choice of a small subset of available males (but see

Hess et al. 2012). Thus, in lek mating systems, the effective population size is typically much smaller than the actual population size. Because genetic drift removes allelic richness from a population at a rate of $1/(2*N_e)$ per generation (~1 year/generation for sharp-tailed grouse, Hartland Clark 2007), demographic compression can have disproportionately large impacts on inbreeding propensity for lek-breeding species. Continued monitoring of the population genetics of this population is warranted to track the potential loss of genetic diversity that may occur as a result of this population reduction. If the population can be recovered rapidly, inbreeding depression should not be an issue in this population. Management efforts should focus on enhancing population recovery.

Based on the correspondence analysis, the most likely cause for the genetic structure (and associated population decline) is a reduction in the quantity and quality of the primary habitat and an increase in other land uses (Table 6). We did not see a strong effect of fragmentation; however, it is difficult to disentangle the influences of habitat loss from fragmentation. Again, focused habitat restoration, increasing the amount of primary habitat, and improving the quality of existing habitat will provide the greatest benefit to maintain connectivity within the EC region (Figure 5, Table 6).

Sharp-Tailed Grouse (NW Region)

The NW region has high genetic diversity, a low inbreeding coefficient, and a large N_e with a 95% confidence interval that encompasses infinity. However, the population is highly structured with K = 5 distinct clusters with limited genetic exchange among them (F_{ST} = 0.68–0.79). Yet, the populations are not highly structured by landscape (land cover / land use) attributes. A common null hypothesis in landscape genetic analyses is isolation by distance, or migration drift equilibrium (Cushman 2006). Although we did not explicitly test for isolation by distance, we did control for it. Our analysis suggested that much of the NW may actually be at or near migration drift equilibrium conditions, with 2 notable exceptions. First, the eastern edge of the region near Koochiching County was found to be an F_{IS} ColdSpot (Figure 8). As previously noted, this may be indicative of a high gene flow system with higher than anticipated migration into and/or out of the system, perhaps indicating marginal habitat conditions along the edge of the current range. Second, the F_{IS} HotSpot identified near Marshall County (Figure 8) indicates that sharp-tailed grouse in this area may have less than expected migration into and out of the system. However, the genetic assignment data (Figure 7) indicates movement within the NW region, suggesting little immediate concern for the genetics or movement of sharp-tailed grouse in the NW.

In conclusion, genetic data suggest that due to high observed genetic diversity, neither species of prairie grouse is in immediate danger of inbreeding depression. Both species exhibited relatively high observed heterozygosity and high observed allelic richness. In addition, analyses of genetic structure indicated connectivity among local populations of both species of grouse.

We also observed that sharp-tailed grouse in the EC management region are not a distinct genetic population from the sharp-tailed grouse in the eastern portion of the NW region. When we linked the genetic data to patterns of land cover/land use, we observed that limited contemporary or relatively recent historical connectivity has occurred between these 2 regions. Furthermore, genetic data suggest that sharp-tailed grouse in the EC region have experienced a demographic bottleneck recently. The landscape genetic analysis further suggests that this demographic bottleneck is likely due to a reduction in primary habitat throughout this region. These genetic data suggest that current declines within the EC sharp-tailed grouse region that are documented with annual surveys are not due to inbreeding depression but are the result of other factors, such as changes in habitat. This would not have been possible to elucidate without the application of landscape genetic approaches.

Finally, we also observed some genetic structure occurring at the subpopulation level within both greater prairie-chickens and sharp-tailed grouse in Minnesota. This observed structuring resulted in inbreeding HotSpots. Inbreeding HotSpots warrant further investigation and monitoring because they represent areas where further landscape management to improve habitat conditions for landscape connectivity may be needed to [limit further genetic structuring].

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Table 1. Summary of microsatellite marker variability and appropriateness for use in genotypic analysis of greater prairie-chickens (GRPC) and sharp-tailed grouse (STGR) in Minnesota based on data collected during 2014-2015.

Marker name(citation)	NA / H_O / H_E[1] GRPC	NA / H_O / H_E STGR	HWE[2] GRPC	HWE STGR
ADL146(Cheng et al. 1995)	7 / 0.63 / 0.68	9 / 0.66 / 0.68	0.053-0.652	0.077-0.555
ADL230(Cheng et al. 1995)	14 / 0.84 / 0.87	14 / 0.80 / 0.85	0.048-0.251	0.236-0.919
BG16(Piertney and Höglund 2001)	18 / 0.75 / 0.88	18 / 0.81 / 0.89	0.043-0.720	0.029-0.199
BG18(Piertney and Höglund 2001)	13 / 0.83 / 0.81	15 / 0.82 / 0.82	0.013-0.751	0.032-0.912
LLSD3(Piertney and Dallas 1997)	17 / 0.85 / 0.85	21 / 0.77 / 0.84	0.048-0.345	0.00-0.589
LLSD4(Piertney and Dallas 1997)	27 / 0.94 / 0.94	28 / 0.93 / 0.95	0.119-0.324	0.093-1.00
LLSD7(Piertney and Dallas 1997)	24 / 0.88 / 0.88	25 / 0.79 / 0.89	0.476-0.589	0.022-0.722
SGCA6(Taylor et al. 2003)	296 / 1.0 / 0.75	367 / 1.0 / 0.75	NA	NA
SGCA9(Taylor et al. 2003)	27 / 0.94 / 0.94	28 / 0.92 / 0.94	0.249 -0.361	0.320-1.00
TUD3(Caizergues et al. 2001)	26 / 0.91 / 0.91	26 / 0.91 / 0.91	0.051-0.975	0.222-0.975
TUT2(Caizergues et al. 2001)	3 / 0.47 / 0.47	3 / 0.43 / 0.47	0.199-0.751	0.182-0.751
TUT3(Caizergues et al. 2001)	9 / 0.78 / 0.78	10 / 0.79 / 0.79	0.106-0.748	0.033-0.981
TTD1(Caizergues et al. 2001)	8 / 0.77 / 0.77	9 / 0.62 / 0.78	0.291-0918	0.104-0.383
TTD3(Caizergues et al. 2001)	20 / 0.51 / 0.57	19 / 0.60 / 0.59	0.026-0.535	0.054-0.190
TTD6(Caizergues et al. 2001)	16 / 0.70 / 0.81	17 / 0.66 / 0.68	0.062-0.652	0.062-0.739

[1] NA = Number of Alleles, H_O = Observed Heterozygosity; H_E = Expected Heterozygosity

[2] HWE = Results of Hardy Weinberg Exact Tests for Neutrality. If markers are entirely homozygous or heterozygous, or otherwise fail to conform to Hardy Weinberg expectations, they are eliminated from the analysis because they will be uninformative. SGCA6 was removed from the analysis due to failure to conform to Hardy Weinberg expectations.

Table 2. Summary of resistance surface modeling values for greater prairie-chickens[1] in Minnesota 2014-2015.

Land cover and use classifications	H1[2]	H2	H3	H4	H5	H6	H7	H8	REV
Trees	500	-	-	-	-	500	500	500	500
Grassland, Meadows, Shrublands	25	-	-	-	-	25	25	25	10,000
Wet Meadows	75	-	-	-	-	75	75	75	10,000
Fresh Water	800	-	-	-	-	800	800	800	800
Cultivated Agriculture	500	-	-	-	-	500	500	500	1,000
Semi-Natural Vegetation	100	-	-	-	-	100	100	100	100
Recently Disturbed	10,000	-	-	-	-	10,000	10,000	10,000	75
Developed/Urban	10,000	-	-	-	-	10,000	10,000	10,000	25
%Grasslands (10km)	-	1-22	1-22	1-22	-	-	-	1-22	22-1
%Cultivated Agriculture (10km)	-	-	33-0	33-0	-	-	-	33-0	0-33
%Cultivated Agriculture × %Grassland	-	-	-	4-55	-	-	-	55-4	4-55
Human Footprint (1km)	-	-	-	-	-	0-100	0-100	0-100	100-0
Power Lines, Roads & Railroads	-	-	-	-	0-600	-	600	600	0

[1] We calculated resistance values based on a thorough review of the scientific literature (Spears 2010), knowledge of prairie grouse biology, and understanding of how the statistical program works. The absolute values used are essentially meaningless, but the relative differences among values yields insights about how the landscape is influencing greater prairie-chickens or sharp-tailed grouse. Our values (Tables 2 and 3) reflect our hypothesis that primary habitat (grasslands, wet meadows, or shrublands) is beneficial (low resistance), other natural features such as open water or trees are a partial barrier (more so for greater prairie-chickens than sharp-tailed grouse; intermediate resistance), cultivated agriculture is also an intermediate barrier to movements, and urban areas or recently disturbed/modified areas are highly avoided (high resistance). Cultivated agriculture at low-to-intermediate densities on the landscape is beneficial; at high densities (>50%) it is detrimental. We chose an intermediate value of resistance to capture this dynamic interaction threshold between prairie-grouse and cultivated agriculture in our landscape model.

[2] H1 = Hypothesis 1. Same notation for all hypotheses. See Methods section for descriptions of the hypotheses.

Table 3. Summary of resistance surface modeling values for sharp-tailed grouse[1] in both the east-central and northwest management regions in Minnesota 2014-2015.

Land cover and use classifications	H1[2]	H2	H3	H4	H5	H6	H7	H8	REV
Trees	200	-	-	-	-	200	200	200	800
Grassland, Meadows, Shrublands	25	-	-	-	-	25	25	25	10,000
Wet Meadows	75	-	-	-	-	75	75	75	10,000
Fresh Water	800	-	-	-	-	800	800	800	800
Cultivated Agriculture	500	-	-	-	-	500	500	500	1,000
Semi-Natural Vegetation	100	-	-	-	-	100	100	100	100
Recently Disturbed	10,000	-	-	-	-	10,000	10,000	10,000	75
Developed/Urban	10,000	-	-	-	-	10,000	10,000	10,000	25
%Grasslands & Wet Meadows (10km)	-	1-22	1-22	1-22	-	-	-	1-22	22-1
%Cultivated Agriculture (10km)	-	-	33-0	33-0	-	-	-	33-0	0-33
%Cultivated Agriculture × %Grassland & Wet Meadows	-	-	-	55-4	-	-	-	55-4	4-55
Human Footprint (1km)	-	-	-	-	-	0-100	0-100	0-100	100-0
Power Lines, Roads & Railroads	-	-	-	-	0-600	-	600	600	0

[1] We calculated resistance values based on a thorough review of the scientific literature (Spears 2010), knowledge of prairie grouse biology, and understanding of how the statistical program works. The absolute values used are essentially meaningless, but the relative differences among values yields insights about how the landscape is influencing greater prairie-chickens or sharp-tailed grouse. Our values (Tables 2 and 3) reflect our hypothesis that primary habitat (grasslands, wet meadows, or shrublands) is beneficial (low resistance), other natural features such as open water or trees are a partial barrier (more so for greater prairie-chickens than sharp-tailed grouse; intermediate resistance), cultivated agriculture is also an intermediate barrier to movements, and urban areas or recently disturbed/modified areas are highly avoided (high resistance). Cultivated agriculture at low-to-intermediate densities on the landscape is beneficial; at high densities (>50%) it is detrimental. We chose an intermediate value of resistance to capture this dynamic interaction threshold between prairie-grouse and cultivated agriculture in our landscape model.

[2] H1 = Hypothesis 1. Same notation for all hypotheses. See Methods section for descriptions of the hypotheses.

Table 4. Factor loadings from Partial Canonical Correspondence Analysis (PCCA) of greater prairie-chicken genetic structure in Minnesota 2014-2015. We did not standardize variances prior to analysis with PCCA; therefore, effect sizes (loadings) are not directly comparable. However, direction and relative strength of influence are comparable. Larger absolute values indicate greater importance, whereas the sign (+ or -) indicates the direction of the interaction.

Factor	Loading axis 1	Loading axis 2
Powerlines	0.05	-0.02
Human Footprint	1.49	2.83
Multiple Resolution Land Cover Data Resistance	39.59	75.37
%Agriculture	-3.44	-6.55
%Grassland	1.35	2.57
%Grassland × %Agriculture	-4.79	-9.12

Table 5. Factor loadings from Partial Canonical Correspondence Analysis (PCCA) of sharp-tailed grouse genetic structure between regions in Minnesota 2014-2015. We did not standardize variances prior to analysis with PCCA; therefore, effect sizes (loadings) are not directly comparable. However, direction and relative strength of influence are comparable. Larger absolute values indicate greater importance, whereas the sign + or -, indicates the direction of the interaction.

Factor	Loading axis 1	Loading axis 2
Powerlines	0.61	-0.76
Human Footprint	0.02	0.25
Multiple Resolution Land Cover Data Resistance	78.57	59.30
%Agriculture	-4.88	0.85
%Primary Habitat	3.17	-1.07
%Primary Habitat × %Agriculture	-4.55	8.79

Table 6. Factor loadings from Partial Canonical Correspondence Analysis (PCCA) of sharp-tailed grouse genetic structure within the east-central management region in Minnesota 2014-2015. We did not standardize variances prior to analysis with PCCA; therefore, effect sizes (loadings) are not directly comparable. However, direction and relative strength of influence are comparable. Larger absolute values indicate greater importance, whereas the sign + or -, indicates the direction of the interaction.

Factor	Loading axis 1	Loading axis 2
Powerlines	-0.54	-1.73
Human Footprint	0.14	2.43
Multiple Resolution Land Cover Data Resistance	31.66	64.93
%Agriculture	-6.40	-30.10
%Primary Habitat	31.39	84.45
%Primary Habitat × %Agriculture	42.56	-18.42

Table 7. Factor loadings from Partial Canonical Correspondence Analysis (PCCA) of sharp-tailed grouse genetic structure within the northwest management region in Minnesota 2014-2015. We did not standardize variances prior to analysis with PCCA; therefore, effect sizes (loadings) are not directly comparable. However, direction and relative strength of influence are comparable. Larger absolute values indicate greater importance, whereas the sign + or -, indicates the direction of the interaction.

Factor	Loading axis 1	Loading axis 2
Powerlines	-11.88	-12.99
Human Footprint	-4.01	-3.43
Multiple Resolution Land Cover Data Resistance	56.95	-19.14
%Agriculture	-1.89	-12.99
%Primary Habitat	26.87	50.04
%Primary Habitat × %Agriculture	3.90	-3.38

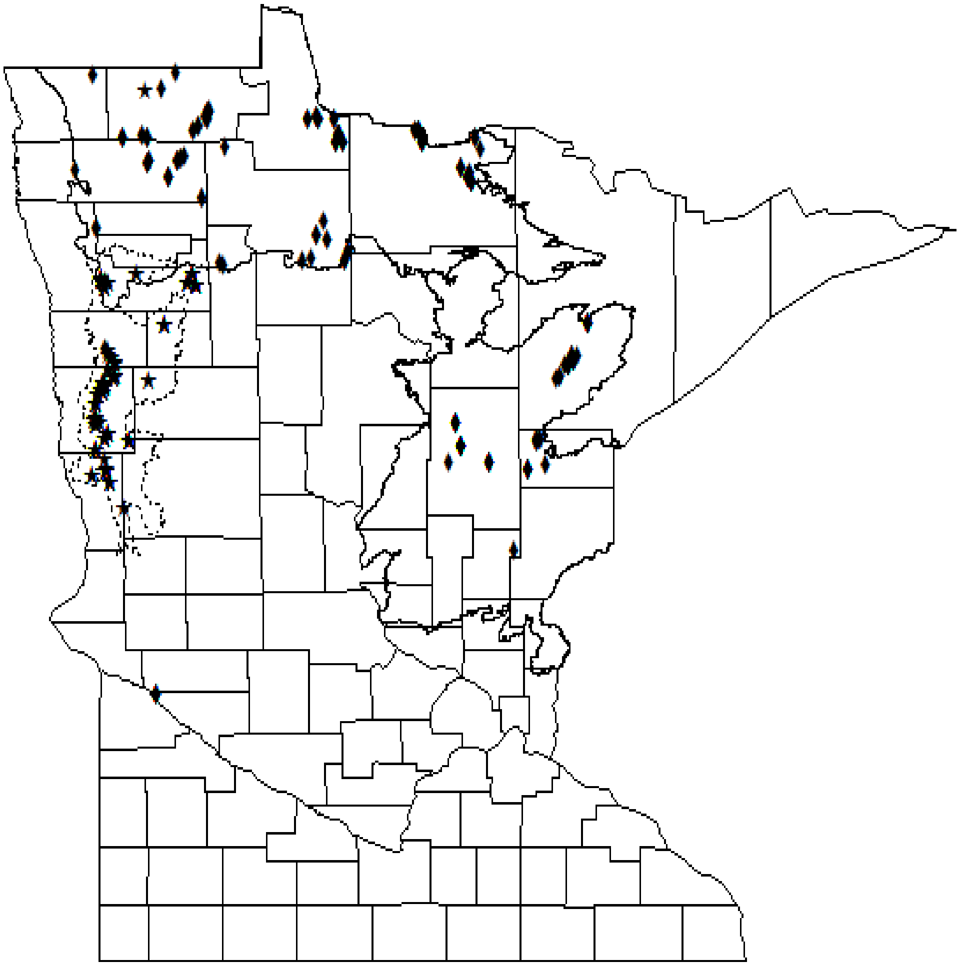

Figure 1. Minnesota sharp-tailed grouse and greater prairie-chicken sample locations (black diamonds and black stars, respectively) during 2014 and 2015 with boundaries of openland focus areas for sharp-tailed grouse (solid line) and approximate current primary range of prairie-chickens (dotted line). The background shows county boundaries.

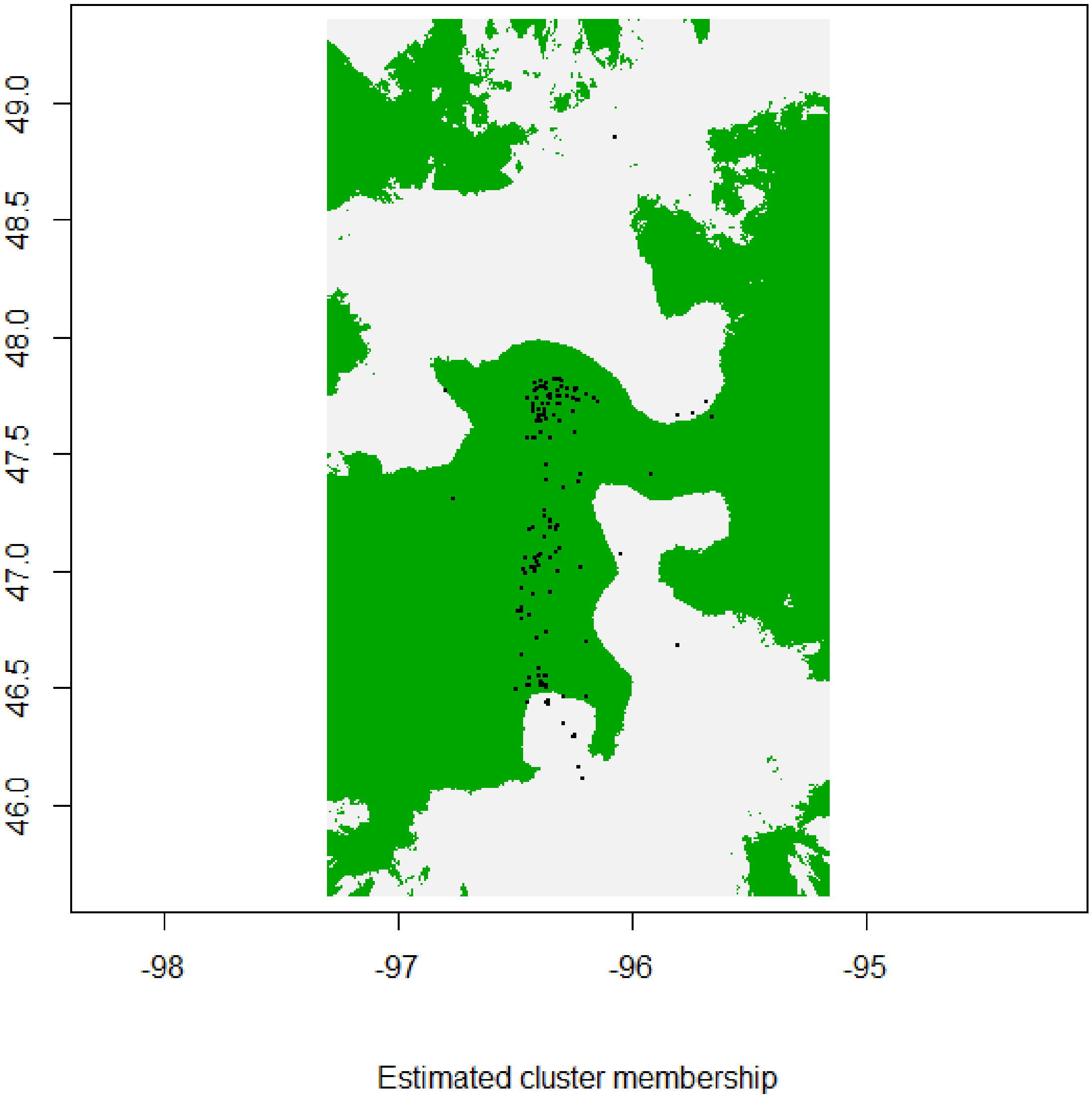

Figure 2. Geneland map of greater prairie-chicken genetic clusters for samples collected in Minnesota during 2014 and 2015. In the figure above the x-axis is degrees of longitude and the y-axis is latitude. Geneland identified 2 distinct genetic clusters—a primary cluster of locations (black dots) in the north and west (green background) and another cluster in the south and east (grey background).

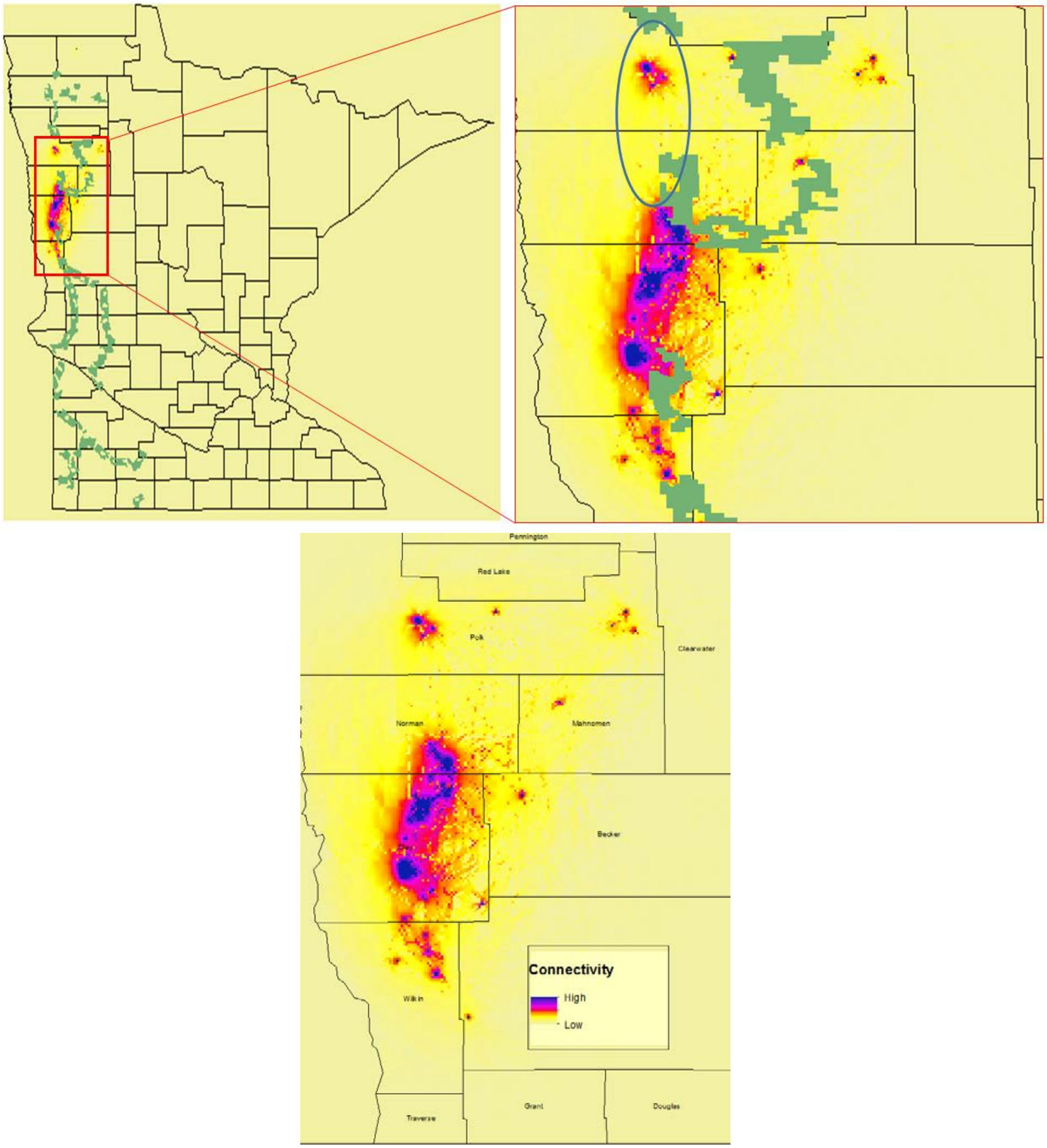

Figure 3a,b. Connectivity model highlighting gaps in corridor coverage for greater prairie-chickens based on the results of the Canonical Correspondence Analysis and Resistance Modeling with Circuitscape for samples collected in Minnesota during 2014 and 2015. When the connectivity map is overlaid with the Prairie Plan corridor area, an area lacking connectivity within the planned corridor is indicated in Norman and Polk counties (blue oval). The top figure depicts the connectivity of the sampled primary prairie-chicken range with the corridors depicted at the statewide scale and the current primary prairie-chicken range scale; the bottom figure excludes the planned corridor areas.

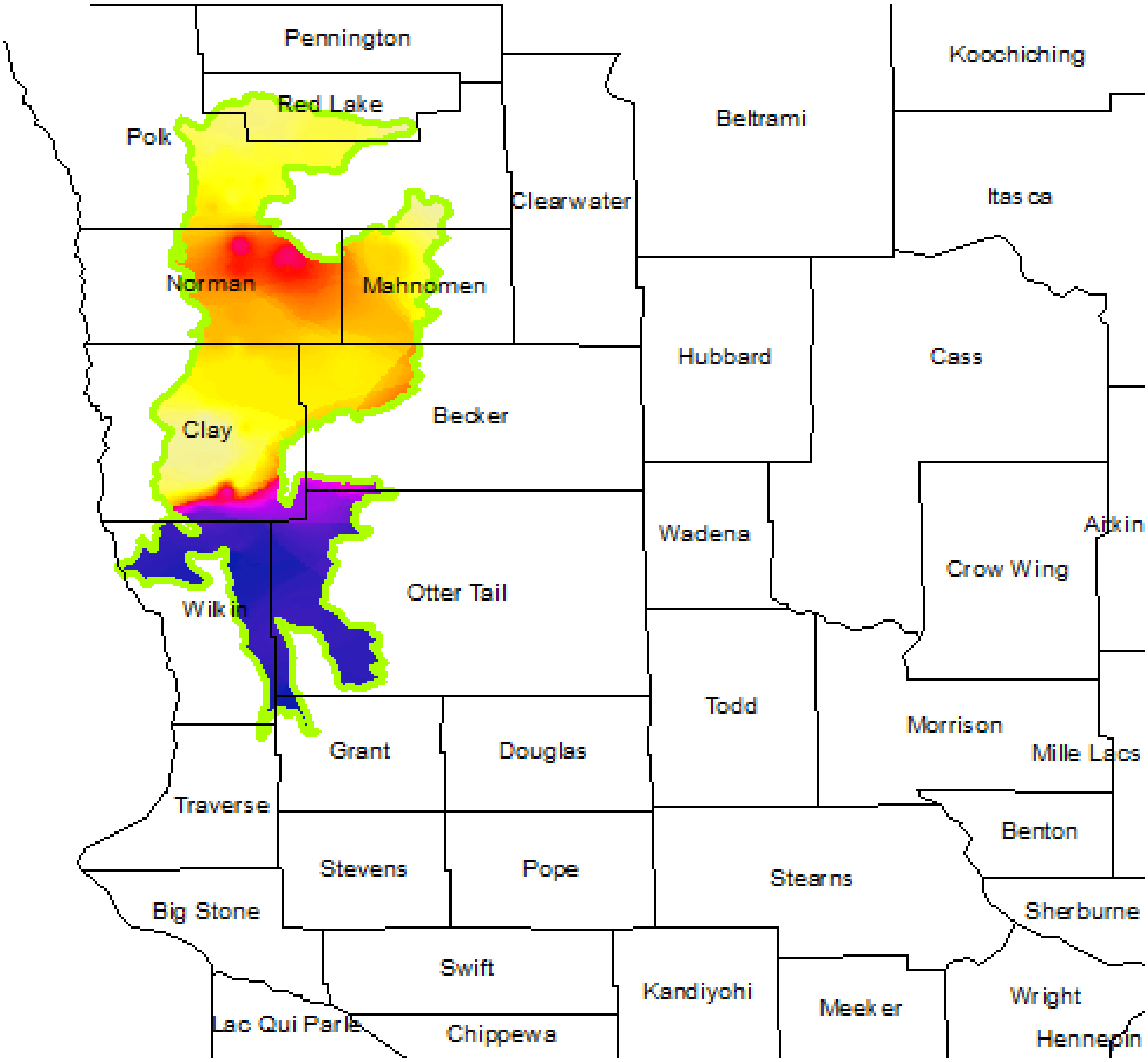

Figure 4. Greater prairie-chicken subpopulation isolation analysis based on HotSpot Analysis of F_{IS} values for samples collected in Minnesota during 2014 and 2015. The map indicates areas with limited migration into or out of the local region. Red areas in the north near Norman and Mahnomen Counties indicate areas where significant high × high clustering of F_{IS} values is occurring (HotSpots). This is indicative of populations that are isolated or becoming isolated due to limited dispersal. Yellow areas in Polk, Clay, and Becker Counties are areas where no clustering was observed; movement is likely unhindered or near migration drift equilibrium. Blue areas in the south near Wilkin and Otter Tail Counties are ColdSpots (low×low F_{IS} value clustering) and are areas where the genetic data suggest that movement is greater than anticipated due to random processes. This might be indicative of high movement due to marginal habitat quality. The red band in Clay County immediately north of the source area suggests a disruption in gene flow that may be indicative of a barrier to dispersal.

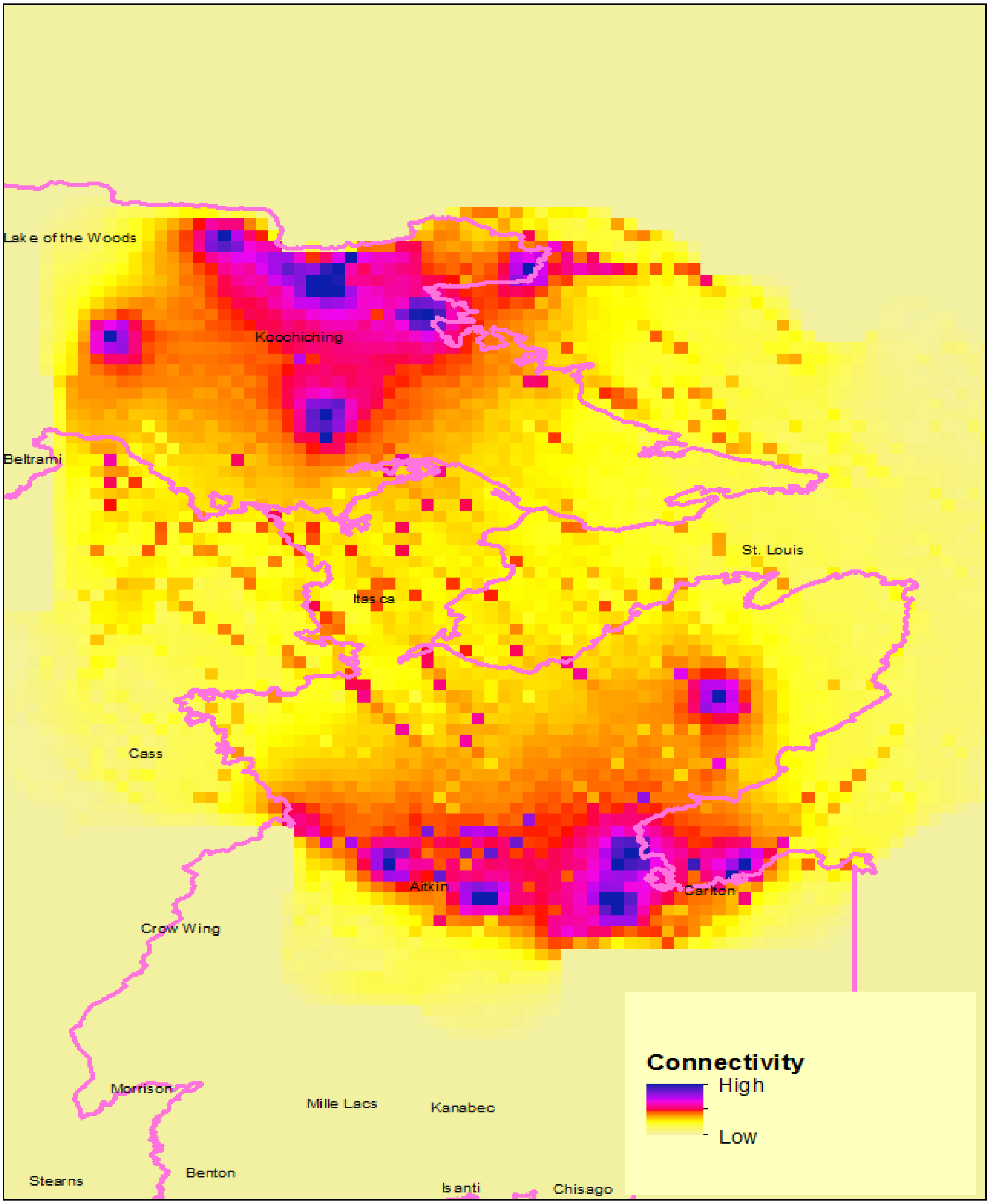

Figure 5. Connectivity model predicting connectivity between sharp-tailed grouse management regions that are delineated along boundaries of Ecological Classification Section subsections (sinuous [color] lines) in Minnesota. The map depicts high connectivity within portions of each management region, but limited connectivity between the northwest and east-central management regions based on samples collected during 2014 and 2015.

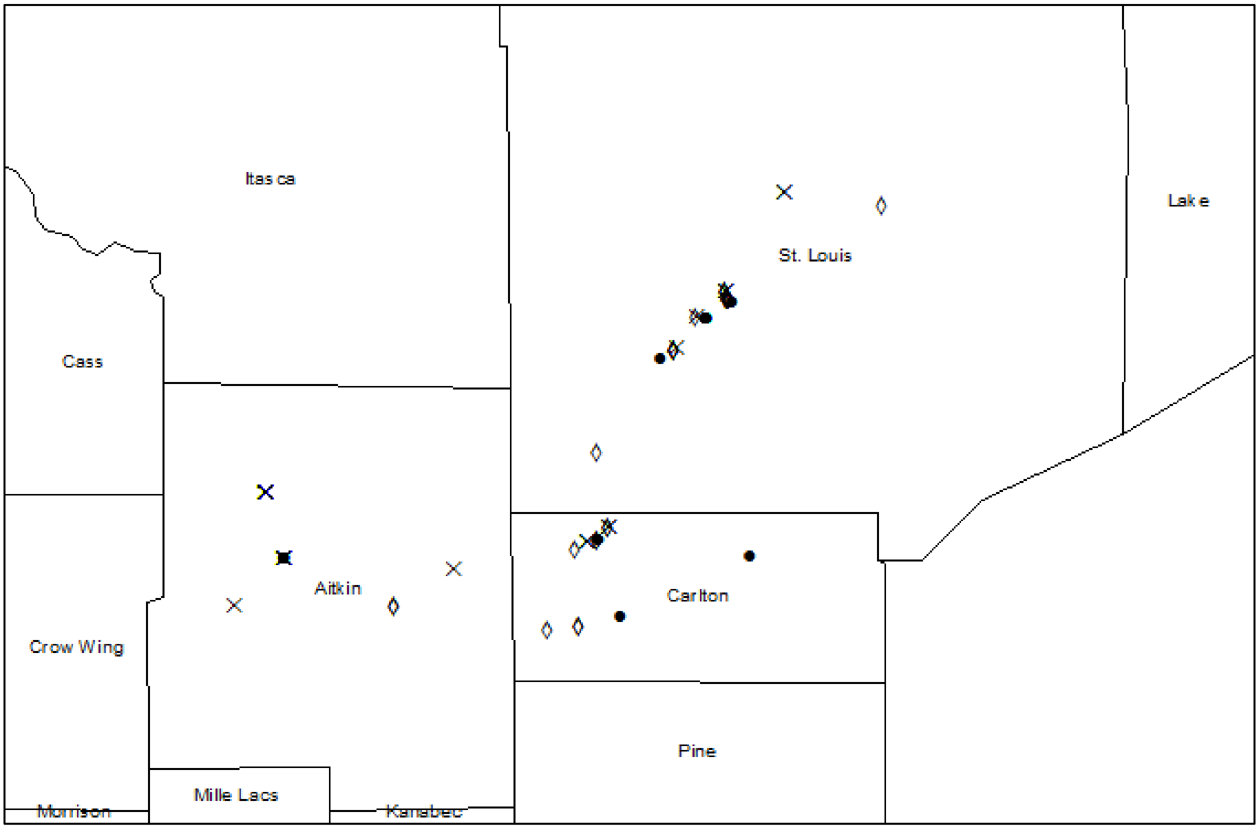

Figure 6. Sampling sites during 2014 and 2015 in the east-central management region of Minnesota mapped based on putative population assignment from Program Structure. Program Structure found greatest support for 3 genetic clusters within the east-central region, indicated symbolically with circles, diamonds, and crosses. The observed clusters do not have a strong spatial pattern, which is indicative of a recent bottleneck or mixing of previously isolated populations, but not structure.

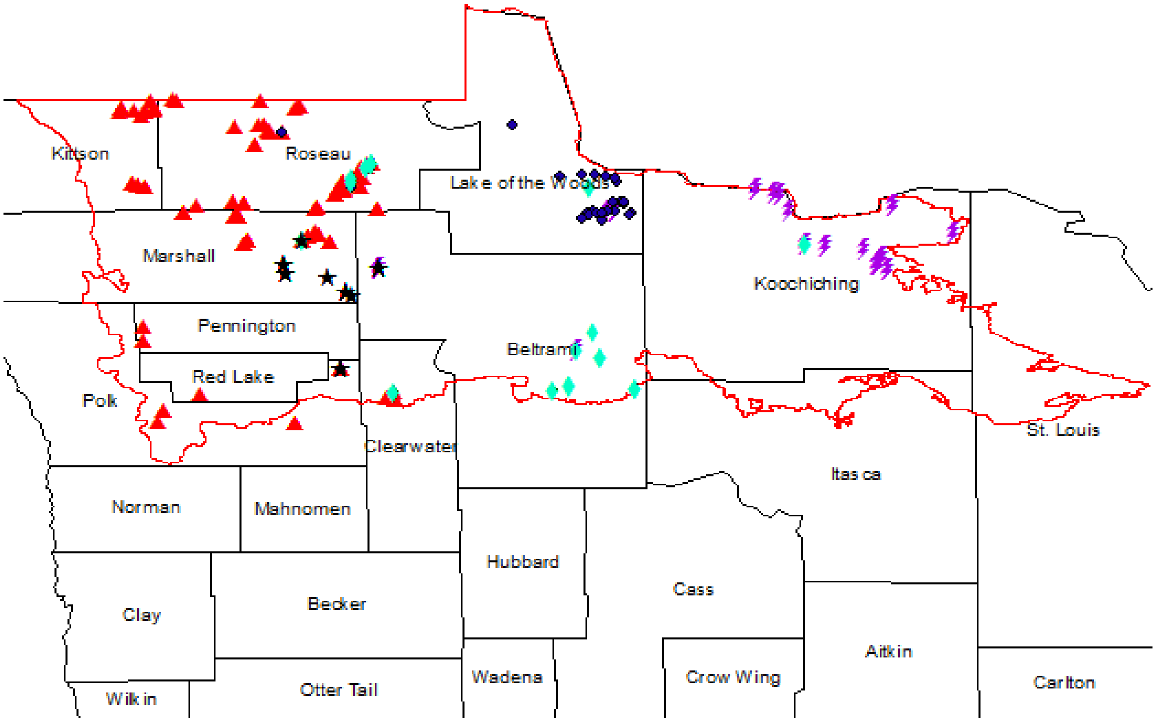

Figure 7. Genetic clusters in the northwest sharp-tailed grouse management region of Minnesota as identified by Program Structure from samples collected during 2014 and 2015. Each set of symbols represents a distinct genetic cluster. Some intermixing of the clusters occurs, but most of the genetic population identities are the same as their sampling location (e.g., all lightning bolts are in Koochiching County). The exception is the cluster centered in Beltrami County, indicating migrants from that area into other areas (i.e., there are diamonds elsewhere in the region).

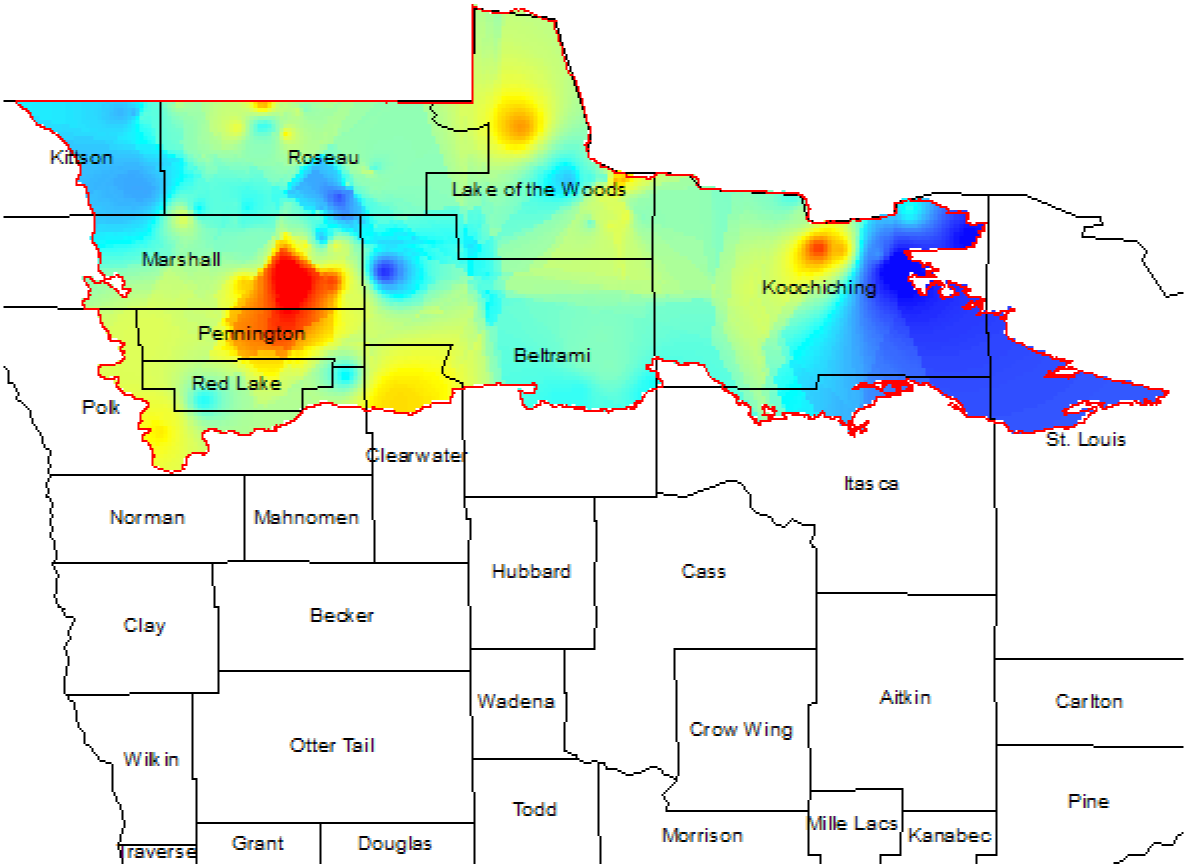

Figure 8. Sharp-tailed grouse subpopulation isolation analysis for the northwest management region in Minnesota, indicating areas with limited migration into or out of the local region. This analysis was based on HotSpot Analysis of F_{IS} values and samples collected during 2014 and 2015. Limited movements are indicated in red (e.g., a foci in Pennington and Marshall counties), but more movement than expected occurred along the eastern and western borders of the range (blue), with intermediate values occurring in yellow. Habitat is most likely marginal in the far eastern part of the range, resulting in more searching and movement, whereas birds in areas with better habitat likely settle closer to their natal areas.

SHARP-TAILED GROUSE RESPONSE TO FALL PRESCRIBED FIRE AND MOWING

Charlotte Roy and Lindsey Shartell

SUMMARY OF FINDINGS

We began a 2-year pilot study in 2015 to examine sharp-tailed grouse (i.e., sharptail, *Tympanuchus phasianellus*) responses to habitat management in the fall (mid-August through November). Our study area included the northwest (NW) and east-central (EC) sharp-tailed grouse populations in Minnesota, but only one site was managed in the fall in the EC region during the pilot study. We studied responses to prescribed fire and mechanical treatment (i.e., mowing) using a Before-After-Control-Impact (BACI) design. In falls of 2015 and 2016, we measured sharp-tailed grouse use and vegetation at 15 managed and 14 control sites prior to and following management. Managed areas included 7 mowing treatments and 8 prescribed burns, ranging in size from 12 to 664 ac (4.9–269 ha) and totaling 1,812 ac (733.3 ha). We also conducted surveys of sharp-tailed grouse use and vegetation at an additional 18 control sites and 27 sites that were planned to be managed, but for which management could not be completed because of unfavorable weather and site conditions.

We conducted surveys of sharp-tailed grouse use 0–28 (mean 10.3) days before management (PRE), 1 week after (1WK), 1 month after (1MO), and 1 year after (1YR) management by conducting fecal pellet transects and documenting sharptails observed at the site. In the NW, we detected sharp-tailed grouse pellets at 2 of the 14 treatment sites and 3 of the 13 control sites prior to treatment. Following treatment, sharp-tailed grouse pellets were detected in ≥1 fall survey (1WK, 1MO) at 9 treatment sites and 3 control sites. Sharptails were observed at only 1 treatment site and at no control sites in PRE surveys, but in later fall surveys (1WK, 1MO), sharptails were observed at 4 treatment and 2 control sites. In 1YR surveys, which have yet to be completed for sites managed during fall 2016, naïve occupancy of sites treated in fall 2015 was higher than before management, but control sites remained unchanged from pretreatment values.

We have developed a proposal to continue the study for another 3 years but anticipate additional data collection may be necessary to understand the variables that influence sharp-tailed grouse responses to these types of management actions.

INTRODUCTION

Sharp-tailed grouse rely on early successional habitats of open grass and brushland. Historically, these habitats were created and maintained through periodic wildfire. More recently, fire suppression has played a role in reducing habitat for sharp-tailed grouse (Berg 1997). Prescribed fire has become an important management tool for maintaining open grass and brushlands habitats, but it can be difficult to implement effectively or safely under many conditions (e.g., too wet, windy, humid, dry) and can require considerable staff and resources to execute. Thus, wildlife managers supplement prescribed burning with mechanical habitat management tools (e.g., shearing, mowing) to maintain early successional habitats. Although mechanical treatments set succession back, they may not produce the same wildlife response as fire does. Wildlife managers have expressed concern that sharp-tailed grouse are not responding to management in the way they would expect if habitat were limiting.

Fall may be a particularly important season for management because juvenile sharptails disperse to surrounding habitat in the fall. Currently, most prescribed burns on state and other lands in the sharp-tailed grouse range occur in the spring (Roy and Shartell, unpubl. data from DNR Wildlife Managers). Region 1 (R1) regularly conducts fall burning, however Regions 2 and 3 (R2/3) have not been burning in the fall because of concerns about peat fires during drier conditions and challenges mobilizing a large number of fire-qualified staff on short notice during the fall (R1 has a Roving Crew to assist with prescribed fire treatments and R2 does not). This study aims to measure the response of sharptails to prescribed burning and mechanical treatments in the fall, as compared to untreated controls.

Historically, fires occurred throughout the year and maintained early successional habitats, such as open grass and brushland, on the landscape. Grassland fires were started by lightning during the growing season, and Native Americans set fires during both the spring and fall dormant seasons in both grasslands and forests to aid hunting (see review in Knapp et al. 2009). Stand replacing fires occurred at 0-10 year intervals in grass and shrub vegetation types, and in forest and woodland types, understory fires occurred at 0-10 year intervals, with more severe, stand-replacement fires occurring at less frequent intervals in Minnesota (Brown and Smith 2000).

Native Americans referred to the sharp-tailed grouse as the “fire grouse” or “fire bird” because of their association with habitats frequently burned, and kept open, by fire. Sharptails have been shown to respond to prescribed fire treatments. Kirsch and Kruse (1973) found that the numbers of broods hatched per 100 acres was higher in 2 burned areas compared to an unburned control 1 year after spring prescribed fires. Sexton and Gillespie (1979) reported that sharptails switched leks just 2 days after a spring burn, abandoning the former dancing ground in favor of the recently burned site 480 m to the north. Sharptails have also been observed returning to leks to dance the day after a burn (J. Provost, pers. comm.).

Burn season may have an effect on the response of sharptails to prescribed fire treatments. Burns conducted in the fall might attract dispersing juveniles searching for habitat. Numerous bird species are known to be attracted to fire, smoke, and recently burned areas (Smith 2000); smoke, flames, and dark burned ground could provide strong visual cues about habitat creation and its direction from a large distance. Young sharptails disperse during September and October (Gratson 1988), typically <6 km from brood rearing areas near nest sites. Sites burned in the fall are not followed by regrowth of vegetation during winter (Kruse and Higgins 1990) and could serve as lek sites the following spring. Sharp-tailed grouse also resume dancing at leks in the fall; Hamerstrom and Hamerstrom (1951) suggested that these fall dances, which include young males, might establish leks for the following spring.

Similar long-distance cues to habitat creation and maintenance are not provided by mechanical treatments. Thus, we might expect wildlife responses to management lacking these cues to be delayed or muted. In Florida shrub-grassland, burned plots were colonized by birds sooner than the mechanically treated plots, in which shrubs were chopped (Fitzgerald and Tanner 1992); birds were observed in burned plots the next day but not for months in chopped plots. Species richness and abundance remained lower in winter chop plots than in burned and control plots throughout this study. Fitzgerald and Tanner (1992) suggested that this was because burned plots provided more complex structure than mechanically treated plots.

Sharp-tailed grouse densities and responses to management treatments have been measured with numerous methods, but pellet counts are the simplest to execute. Pellet counts along transects have been shown to be indicative of the relative abundance of greater sage grouse

(*Centrocercus urophasianus*) (Hanser et al. 2011), density of red grouse (*Lagopus lagopus scoticus)* (Evans et al. 2007), and habitat use of red grouse (Savory 1978). Pellet counts along transects in plots have been used to compare sage-grouse responses to mechanical and chemical treatments (Dahlgren et al. 2006). Schroeder and Vander Haegen (2014) used pellet counts along circular transects to examine the effects of wind farms on sage-grouse.

OBJECTIVES

1. To compare sharp-tailed grouse use prior to and following fall management within burn, mow, and control treatments.
2. To relate vegetation metrics to differences in sharp-tailed grouse use of burn, mow, and control treatments.

Hypotheses

1. Sharp-tailed grouse use will increase following burning or mowing, with burned sites showing a greater increase in sharptail use than mowed sites, and both treatments having greater sharptail use than controls.
2. Vegetation composition and structure will influence the use of treatment and control sites by sharp-tailed grouse, with increased use in early successional habitats.

METHODS

Study Areas

Our pilot study included the northwest and east-central regions of Minnesota where sharp-tailed grouse occur. Treated study sites were mainly on state lands, however 1 site owned and managed by The Nature Conservancy (TNC) and 2 private land sites were included as well.

In 2015, we conducted pre-treatment surveys at 23 sites that were planned to be managed and 19 control sites. Of these, 10 sites (6 mows and 4 prescribed burns) were treated (Table 1) in the NW and management was not completed at any sites in the EC region. In 2016 we conducted pre-treatment surveys at 19 sites that were planned for management and 13 control sites. Of these, 4 sites (1 mow and 3 prescribed burns) were treated in the NW (2016 was an unusually wet year which restricted management opportunities) and 1 site was burned in the EC region.

Data Collection & Experimental Design

Treatment sites varied in size, date of management, vegetative composition, surrounding landscape, and local sharptail density. We attempted to match treatments in each DNR work area or sub-work area (some work areas are very large) with a control site of similar size and successional stage (e.g., crude habitat classification, visual assessment of percent cover shrubs and herbaceous vegetation, and average shrub height) *a priori* as determined by inspection of aerial imagery, conversations with managers, and site visits. Control sites were identified $\leq$6 km from treatment sites when possible (based on dispersal distances of young males in the fall; Gratson 1988). Control sites helped account for changes related to seasonal progression (i.e., changes in habitat use, social behavior, and vegetation) not related to management. Dahlgren et al. (2006) implemented a similar design to account for temporal differences in the application of management treatments for sage grouse.

We surveyed treatment and control sites as close as possible in time, both before and after treatment (Smith 2002, also see Morrison et al. 2001:118-130). We walked systematically spaced parallel transects with a starting point placed on the site boundary and the transect traversing the treatment capturing both edge and interior portions. The sampling rate was standardized to 10 m of transect/ac (25 m/ha), with transects at least 150 m apart, based on placement of pellet transects in other studies (Evans et al. 2007, but half as dense as Dahlgren et al. 2006, Hanser et al. 2011). We counted sharptail pellet piles $\leq$0.5 m from the transect, removing all pellets encountered (Evans et al. 2007, Schroeder and Vander Haegen 2014). At each pellet pile we recorded pellet freshness and vegetation category (i.e., grass, shrub, forb, grass-shrub mix, grass-forb mix, etc.). We also recorded all sharp-tailed grouse observed (heard, flushed, tracks seen) at the site while walking transects.

We sampled transects 4 times at each site—once before treatment, targeting measurements within 2 weeks of treatment (PRE), and 3 times after treatment; one week after treatment (1WK), one month after treatment (1MO), and one year after treatment (1YR). Matched treatment and control sites were sampled within 14 days of each other. We also surveyed sites during the spring of 2016, but our early results indicated that sharptail use of managed sites in spring could be misleading at sites without a lek due to focused activity at lek sites in the spring. Thus, spring use of managed sites may have little relationship to fall habitat use, so we discontinued spring surveys in 2017.

To adjust naïve occupancy rates for detection differences among treatment groups, vegetation categories, and other sources, we conducted pellet detection assessments. We accomplished this by surveying transects with pellets placed in known locations (but unknown to observers) and estimated detection probabilities for each vegetation and management category. Dahlgren et al. (2006) reported detectability of pellets along transects to be very high and similar in different types of vegetative cover. However, their study was conducted on sage grouse in sage brush, and sharp-tailed grouse habitats in Minnesota differ considerably in vegetative composition and structure.

We sampled vegetation within treatments using point-intercept sampling (Levy and Madden 1933, Dahlgren et al. 2006) to determine percent cover and average height of broad vegetation classes (i.e., tree, shrub, forb, and graminoid) before and after treatment. We sampled vegetation along 20-m transects placed perpendicular to the pellet transect, with the number of transects based on the size of the site. We marked the start of each vegetation transect using ground staples with numbered aluminum tags and flagging, and we used Global Positioning System (GPS) coordinates to allow re-measurement following treatment. For each vegetation class we recorded maximum height every 0.5 m for a total of 40 points per transect. We used a pole with graduated measurements every dm to determine the type of vegetation intercepted (touching the pole) and the highest point at which each vegetation class touched the pole. We also recorded whether the vegetation was dead/dormant, combining those categories because it was unclear due to natural plant senescence whether vegetation was dormant or dead in late-fall surveys. Following treatment, we classified cut vegetation as dead/dormant, recorded height, and noted that the vegetation was cut. If no vegetation was present, the substrate type was recorded. For the purpose of this study, moss and lichen were considered a substrate type rather than vegetation. Vegetation metrics were determined for each study site. Proportion of cover in each class and average maximum height were compared among treatment types and between sites with and without sharptail use. In our preliminary analysis, we included both live and dead vegetation and used the maximum height of vegetation at each point. Significant differences were tested for using Tukey's Honest Significant Difference test.

RESULTS AND DISCUSSION

Sharp-tailed grouse pellets were detected on transects at 2 (14%) of the 14 treatment sites and 3 (23%) of the 13 control sites prior to treatment in the NW (Table 2). Following treatment, sharp-tailed grouse pellets were detected in >1 fall survey (1WK, 1MO) at 9 treatment sites (64%) and 3 control sites (23%). Sharptail observations on transects prior to treatment exhibited similar patterns, with detections at only 1 treatment site (7%) and no control sites (0%). In later fall surveys (1WK, 1MO), however, sharptails were observed at 4 treatment sites (29%) and 2 control sites (15%, Table 3). In 1YR surveys (completed for 2015 sites to date), we detected pellets on transects at 3 (30%) of 10 treatment sites and 2 (22%) of 9 control sites, and sharptails were observed on transects at 2 treatment sites (20%) and 1 control site (11%). Naïve occupancy of treated sites was higher 1YR later, but occupancy of control sites remained unchanged (Figure 1).

Our pellet survey results thus far suggest that our methods are capturing sharptail use of treatment and control sites. Naïve occupancy rates (i.e., site use) from data collected thus far suggest increases in sharptail use of sites following management (Figure 1). Although occupancy and detection are confounded in naïve estimates for the 1WK, 1MO, and spring (SP) surveys (due to treatment effects on screening cover), surveys conducted 1 year (1YR) following treatments should have similar detection rates to pre-treatment measurements due to regrowth of vegetation the next growing season, especially in burn sites. Thus, the PRE vs. 1YR comparison should be reasonably straightforward and informative (e.g., Figure 2), whereas results from other time comparisons are more tenuous to interpret from naïve occupancy rates. Nevertheless, demonstrating that managed sites are used after management directly addresses manager concerns.

General field observations of vegetation prior to treatment indicated that mowing might be applied to sites at a later successional stage than prescribed fire. However, there were no significant differences observed when averaging sites by treatment, possibly due to the low sample size and high variability of sites. Despite this, mow sites tended to have a lower mean proportion of grass cover, greater mean proportion of forb and shrub cover, and taller shrubs than burn sites (Table 4).

Control sites had lower graminoid height in 1MO surveys than PRE surveys, which was likely the result of vegetation senescence (Table 5). One year later, we did not detect differences in vegetation cover or height at control sites compared to pre-treatment measurements (Table 5). At sites that were mowed, shrub cover and graminoid, forb, and shrub height were lower in 1MO surveys, but in 1YR measurements only shrub height differed from PRE survey measurements. At sites that were burned, graminoid cover, forb cover, and graminoid height were lower in 1MO surveys, but in 1YR surveys no differences were detected (Table 5). Sites occupied by sharp-tailed grouse did not differ in vegetation cover or height from unoccupied sites during PRE or 1YR surveys (Table 6).

This report includes the fall surveys for the second year of data collection but not the 1YR surveys that will be conducted in fall 2017. Results presented in this report are preliminary and subject to revision. We anticipate that 5 years of data collection may be necessary to understand the complex responses of sharp-tailed grouse to fall management treatments and associated vegetation changes. This 2 year pilot study provided data to inform development of a proposal for continuing the study. Managers throughout sharptail range in Minnesota have expressed a need for this type of information to more effectively manage for sharptails. Given the current sharptail population concerns in the east-central region, information on the effectiveness of various management options would be helpful for decision-making with finite resources for management. Managers in the northwest region are also interested in this information to ensure that their management actions are as effective as possible.

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Table 1. Management activities completed for sharp-tailed grouse habitat during falls 2015 and 2016 and associated control sites, in order of treatment date.

Site name	Work area	Treatment	Treatment date	Treatment ac (ha)	Control ac (ha)
Roseau River	Roseau River	Mow	28 Aug–16 Sep 15	31 (12.5)	28 (11.3)
Skull Lake	Karlstad	Burn	1 Sep 2015	90 (36.4)	70 (28.3)
Halma	Karlstad	Mow	16–23 Sep 2015	41 (16.6)	39 (15.8)
Red Lake mow	Red Lake	Mow	22 Sep 2015	12 (4.9)	22 (8.9)
Spooner	Baudette	Mow	28 Sep 2015	22 (8.9)	26 (10.5)
Caribou	Karlstad	Burn	28 Sep 2015	664 (268.7)	No control
TL 2015 burn	Thief Lake	Burn	28 Sep 2015	58 (23.5)	31 (12.5)
Red Lake burn	Red Lake	Burn	19 Oct 2015	152 (61.5)	176 (71.2)
Prosper	Baudette	Mow	19–30 Oct 2015	63 (25.5)	201 (81.3)
TL mow	Thief Lake	Mow	30 Oct 2015	20 (8.1)	19 (7.7)
TL 2016 burn	Thief Lake	Burn	1 Sep 2016	31 (12.5)	37 (15.0)
Noracre	Roseau	Burn	14 Sep 2016	71 (28.7)	22 (8.9)
Roseau brush	Roseau	Mow	27 Sep – 7 Oct 16	23 (9.3)	29 (11.7)
Espelie	Thief River Falls	Burn	3 Oct 2016	443 (179.3)	460 (186.2)
Hasty Brook*	Cloquet	Burn	16 Nov 2016	91 (36.8)	90 (36.4)

* Hasty Brook was the only site where management was completed in the east-central sharptail region during the 2-year pilot study, and because of snowfall after treatment, post-treatment surveys were incomplete.

Table 2. Sharp-tailed grouse pellet detections at treatment and control sites in northwest Minnesota. Surveys were conducted before (PRE), 1 week (1WK), 1 month (1MO), the spring (SP), and 1 year (1YR) after treatment. The number of pellet detections on transect are indicated numerically, and pellets detected off-transect are indicated with an OT, indicative of site use not captured in sampling. An asterisk indicates that snow impeded detection of pellets, and T indicates that tracks were detected in snow. Surveys with confirmed sharptail use through any source of sign at the site during a survey are highlighted in gray. NS indicates that a survey has not yet been completed for sites managed during fall 2016.

Fecal pellets		Treatment					Control			
Site	PRE	1WK	1MO	SP	1YR	PRE	1WK	1MO	SP	1YR
Red Lake mow	0	0	0	0	2	0	0	0	0	1
Thief Lake mow	0	0	0*	0	0	0	0	0*	0	0
Spooner mow	0	0	2	3	0	0	0	0	0	0
Roseau 2015 mow	2 OT	1 OT	1	0	0	0	0	0	0	0
Halma mow	0	0	0	0	1 OT	1	1	2	0	0
TL 2015 burn	1 OT	0	1	0	1 OT	0	0	0	0	0
Skull Lake burn	0	1	0	0	1	0	0	0	4	0
Red Lake burn	0	0	0	0	0	0	0	0*	0	0
Prosper mow	0	1	0*	0	2	1	11	T*	11	5 4 OT
Caribou burn	1	2	1 OT	1	0	.	.	.	.	-
TL 2016 burn	0	1	4 7 OT	NS	NS	0	0	0	NS	NS
Noracre burn	0	9 3 OT	0	NS	NS	0	0	0	NS	NS
Espelie burn	1	6	18 31 OT	NS	NS	1 1 OT	1 3 OT	4 5 OT	NS	NS
Roseau 2016 mow	1 OT	0	0	NS	NS	0	0	0	NS	NS

Table 3. The number of sharp-tailed grouse observed at treatment and control sites in northwest Minnesota. Surveys were conducted before (PRE), 1 week (1WK), 1 month (1MO), the spring (SP), and 1 year (1YR) after treatment. Sharp-tailed grouse observed while off-transect are indicated with OT, indicating site use not captured in sampling. Surveys with confirmed sharptail use through observations of any birds at the site during a survey are highlighted in gray. NS indicates a survey has not been completed for sites managed in fall 2016.

Grouse observations`		Treatment					Control			
Site	PRE	1WK	1MO	SP	1YR	PRE	1WK	1MO	SP	1YR
Red Lake mow	0	0	0	0	0	0	0	0	0	0
Thief Lake mow	0	0	0	0	0	0	0	0	0	0
Spooner mow	0	0	11	3	3 OT	0	0	0	0	0
Roseau 2015 mow	2 OT	5OT	2OT	1 OT	0	0	0	0	0	0
Halma mow	0	0	1	0	0	0	2	0	0	0
TL 2015 burn	4	0	0	0	0	0	0	0	0	0
Skull Lake burn	0	0	0	0	0	0	0	0	0	0
Red Lake burn	0	0	0	0	0	0	0	0	0	0
Prosper mow	0	0	0	0	1	0	0	0	4	12-20
Caribou burn	0	5	13	0	2	-	-	-	-	-
TL 2016 burn	0	0	0	NS	NS	0	0	0	NS	NS
Noracre burn	0	0	0	NS	NS	0	0	0	NS	NS
Espelie burn	0	1	2 OT	NS	NS	5 OT	1	7 OT	NS	NS
Roseau 2016 mow	6 OT	0	0	NS	NS	0	0	0	NS	NS

Table 4. Mean pre-treatment vegetation cover and height for 4 vegetation classes at control (*n* = 13), mow (*n* = 7), and burn (*n* = 7) sites sampled for sharp-tailed grouse use in northwestern Minnesota during 2015 and 2016.

	Control	Mow	Burn
Cover (proportion)			
Graminoid	0.92	0.88	0.96
Forb	0.33	0.46	0.24
Shrub	0.34	0.36	0.21
Tree	0.05	0.04	0.09
Height (m)			
Graminoid	0.52	0.51	0.56
Forb	0.34	0.37	0.33
Shrub	1.09	1.30	0.74
Tree	2.50	1.63	1.98

Table 5. Change in mean vegetation cover and height from PRE treatment to 1MO (control *n* = 13, mow *n* = 7, and burn *n* = 7) and 1YR (control *n* = 9, mow *n* = 6, and burn *n* = 4) surveys in northwest Minnesota. Comparisons to 1YR surveys exclude sites managed in 2016, thus results are preliminary and subject to change with additional data collection. Significant differences ($P < 0.05$) between pre and post measurements are indicated with an asterisk.

	Control	Control	Mow	Mow	Burn	Burn
	1MO	1YR	1MO	1YR	1MO	1YR
Cover (proportion)						
Graminoid	-0.01	0.02	-0.32	-0.03	-0.38	-0.03
Forb	-0.14	-0.02	-0.35	0.01	-0.18*	0.13
Shrub	-0.06	0.01	-0.30*	-0.07	-0.06	-0.05
Tree	-0.01	0.02	-0.04	-0.04	-0.03	-0.03
Height (m)						
Graminoid	-0.11*	-0.01	-0.37*	-0.08	-0.19*	0.04
Forb	-0.04	-0.03	-0.18*	-0.09	0.10	-0.09
Shrub	0.11	-0.10	-1.05*	-0.77*	0.02	-0.04
Tree	-0.32	0.01	-0.25	-1.03	-0.02	-0.21

Table 6. Mean cover and height at sites occupied and unoccupied by sharp-tailed grouse during PRE (occupied *n* = 8, unoccupied *n* = 19) and 1YR (occupied *n* = 9, unoccupied *n* = 10) surveys in northwest Minnesota. 1YR surveys exclude sites managed in 2016, thus results are preliminary and subject to change with additional data collection. No significant differences (P < 0.05) between occupied and unoccupied sites were observed.

Sharptail occupancy	PRE unoccupied	PRE occupied	1YR unoccupied	1YR occupied
Cover (proportion)				
Graminoid	0.92	0.92	0.90	0.90
Forb	0.36	0.30	0.33	0.41
Shrub	0.34	0.27	0.37	0.29
Tree	0.04	0.09	0.04	0.02
Height (m)				
Graminoid	0.51	0.58	0.54	0.49
Forb	0.35	0.33	0.33	0.28
Shrub	0.97	1.24	0.90	0.72
Tree	1.71	3.32	1.93	1.41

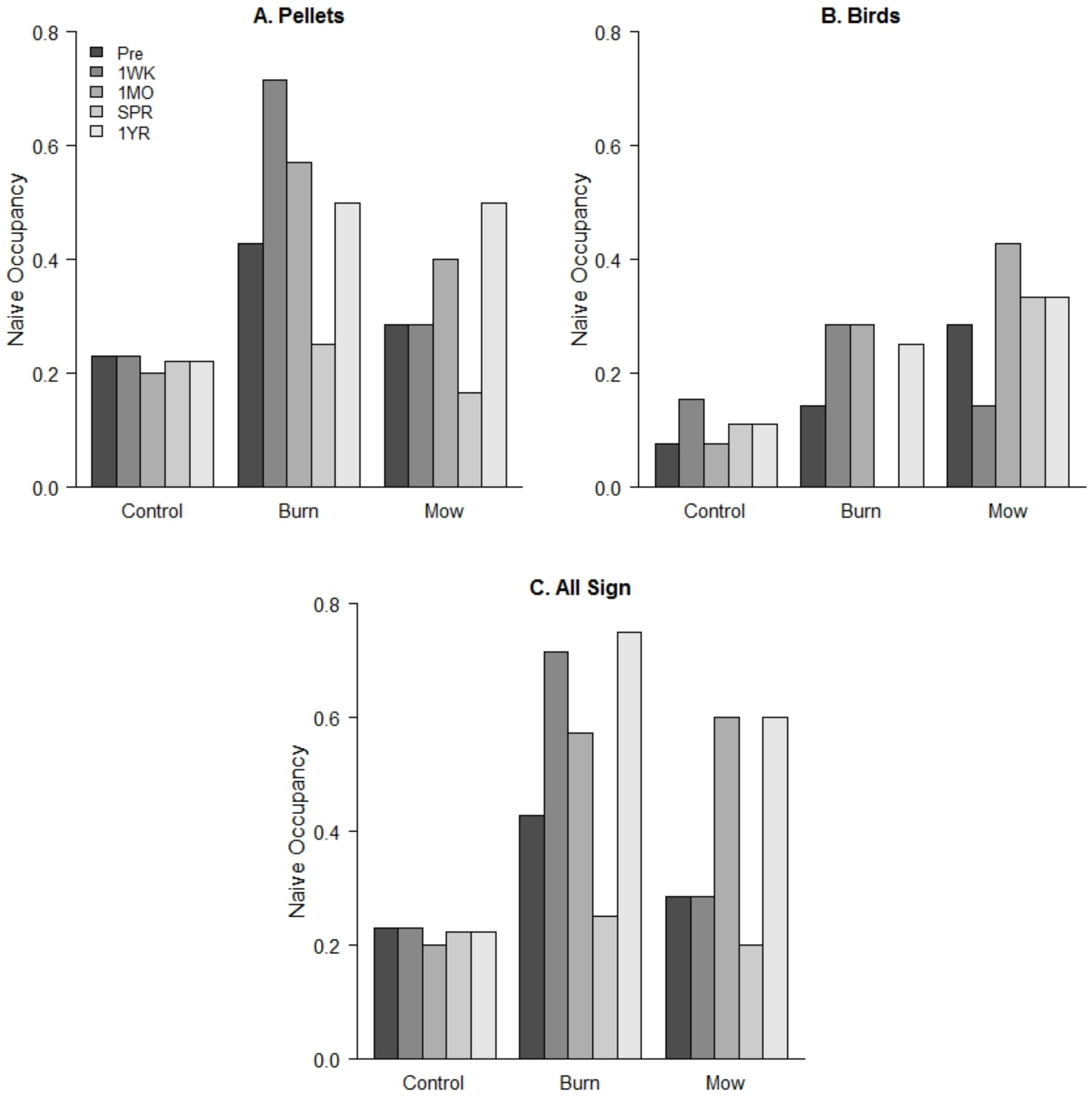

Figure 1. Naïve occupancy for sharptail pellets (A), sharptail observations (B), and all sign (includes off-transect detections), C) during surveys conducted before (PRE), 1 week (1WK), 1 month (1MO), the spring (SP), and 1 year (1YR) after treatment during 2015 in the northwest study area of Minnesota to assess the effects of prescribed burning and mowing.

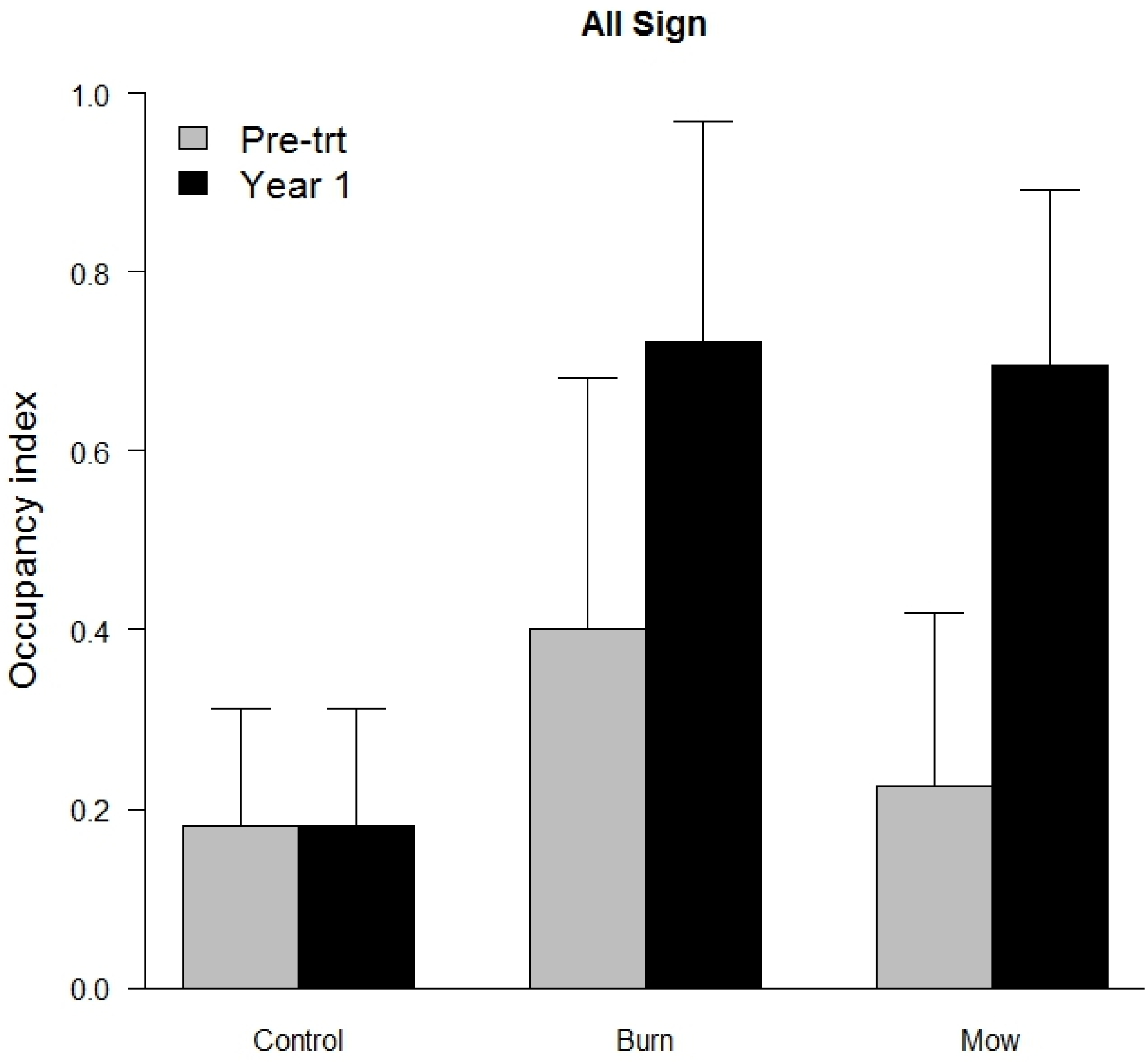

Figure 2. Mean naïve occupancy index at 10 sites managed for sharp-tailed grouse in northwestern Minnesota during 2015 based on a logistic regression model with an offset for transect length.

USE OF MANAGED FOREST OPENINGS BY AMERICAN WOODCOCK

Lindsey Shartell

SUMMARY OF FINDINGS

American woodcock (*Scolopax minor*) were surveyed at permanently managed forest wildlife openings in Minnesota. Singing ground surveys conducted from mid-April through May 2016 indicated that 59% of openings were occupied by singing male woodcock. Roosting ground surveys conducted from June through August 2016 indicated that 23% of openings had confirmed roosting and 71% of openings had woodcock activity in or over the opening. In addition to woodcock surveys, vegetation transects were collected within openings to assess the relationship of vegetation to woodcock use and management of habitat in openings. Information from this pilot study helped to inform the development of a targeted research project to assess management practices and woodcock use of forest openings. This information will guide wildlife managers in creating optimal singing and roosting habitat for woodcock in forest-dominated areas.

BACKGROUND

The American woodcock is a popular migratory game bird and a Species of Greatest Conservation Need in Minnesota (MN DNR 2015). In 2015, Minnesota had an estimated 13,500 active woodcock hunters harvesting 25,600 woodcock, ranking Minnesota third highest in the country for both woodcock hunter and harvest numbers (Seamans and Rau 2016). Annual woodcock surveys have indicated a long-term (1968-2016) decline in singing male numbers across the full breeding range (Seamans and Rau 2016). These declines have been attributed to the loss of open and early successional forest and shrub habitat due to succession, lack of disturbance, and development (Dessecker and McAuley 2001).

Woodcock require a variety of habitat components including dense young forests or shrublands and open singing and roosting grounds (Wildlife Management Institute 2009). Woodcock move frequently between these habitat types, often being found in forests during the day and open sites at night (Sheldon 1967). In the spring, male woodcock use openings as breeding sites, called singing grounds, where they perform their courtship ritual. Females nest and raise broods in the forest surrounding these openings (Sheldon 1967). In the summer, woodcock make evening crepuscular flights to open habitats to roost. Open roosting grounds provide the benefit of reduced predation risk (Masse et al. 2013). Historically, disturbance by fire, wind, Native American activities, flooding, and beavers created openings and early successional habitat for woodcock (DeGraaf and Yamasaki 2003). Many of these disturbances that created and maintained open areas are now prevented. Pastures, fields, agricultural sites, and recent clearcuts (Hale and Gregg 1978, Long and Locher 2013) can all serve as open habitat for woodcock, but in areas dominated by forest cover, managed forest wildlife openings are often used to provide this habitat component.

The secretive nature and cryptic coloration of the woodcock makes it difficult to estimate population size and management effects. There have been past studies assessing the use of openings by woodcock, but most have been focused on wintering grounds (for example

Glasgow1958, Stribling and Doerr 1985, Berdeen and Krementz 1998). Fewer studies have explored woodcock use of summer roosting grounds in the northern part of the range (though see Sheldon 1961, Sepik and Derleth 1993, Masse et al. 2013), and even fewer have incorporated habitat characteristics and management into studies of use. Researchers have also studied the use of aspen clearcuts in Wisconsin and young pine plantations in Arkansas by woodcock in spring and summer, finding that woodcock do utilize these areas (Hale and Gregg 1978, Long and Locher 2013). Additional research comparing the use and characteristics of temporary openings such as clearcut harvests to permanent openings would improve our understanding and provide context for management in Minnesota.

The Upper Great Lakes Woodcock and Young Forest Initiative published best management practices for woodcock in 2009. Their recommendations call for establishing 8 singing grounds at least 0.5 acres in size and 1 roosting field at least 5 acres in size per 100 acres of land (Wildlife Management Institute 2009). Open sites should cover not more than 20 percent of the area, and the remaining land should consist of abundant feeding, nesting, and brood-rearing habitat (Wildlife Management Institute 2009). They also suggest that recent clearcuts can be used by woodcock as singing grounds for “several years” and as roosting grounds for “at least one year” after harvest (Wildlife Management Institute 2009). Assessing the use of recent clearcuts with known harvest dates is needed to better understand how long they can serve as open areas for singing and roosting woodcock.

OBJECTIVES

1. Assess woodcock use of managed forest wildlife openings with differing management history (time since mowing).
2. Relate opening size and configuration, vegetation composition and height, and surrounding landscape to woodcock use and/or management history.
3. Develop recommendations to improve the current management of forest wildlife openings.

METHODS

Singing ground surveys for American woodcock were conducted from mid-April through May 2016 in forest openings within the Grand Rapids, Cloquet, and Red Lake work areas. Surveys followed Singing Ground Survey (SGS) protocol where possible (Seamans and Rau 2016). Surveys generally took place 15 to 60 minutes after sunset, when temperature was above 40 F, and there was no heavy precipitation or strong wind. Openings in close proximity were grouped to allow surveying multiple openings per evening. At each opening observers recorded their GPS location (UTM coordinates), time of sunset, cloud cover, temperature, wind speed, precipitation, and any noise disturbance present at the time of the survey. Observers listened for and recorded the number of different woodcock heard peenting or observed displaying (heard and/or seen) within the opening during a listening period of at least 5 minutes. Observers also recorded other observations of woodcock (not within the opening) along with time and approximate location (direction and distance) of the woodcock.

Roosting ground surveys were conducted June through August 2016 using crepuscular flight surveys and spotlighting (Glasgow 1958, Berdeen and Krementz 1998). The observer was positioned on the edge of the opening and recorded the number of woodcock observed flying into the opening or heard peenting (when not seen). Surveys were conducted from 20 minutes before sunset to 40 minutes after sunset (a one hour period). Observers recorded their GPS location (UTM coordinates), time of sunset, cloud cover, temperature, wind speed, precipitation, and any noise disturbance present at the time of the survey. After the survey window, observers systematically walked openings using spotlights and recorded the number of woodcock flushed or spotted.

Vegetation within forest openings was sampled along 2 transects using a line intersect/intercept method (Canfield 1941). The first transect (Transect A) was placed across the widest part of the opening from edge to edge (as determined in GIS and in the field) and the second transect (Transect B) was placed perpendicular to the first crossing the opening from edge to edge. The transect start and end points were marked using a flag and flagging to aid resampling, UTM coordinates were taken at each (using point averaging to increase accuracy). A measuring tape was stretched tight from the starting point to the end point and secured in place by rebar. The direction of the transect (azimuth) from the start point facing the end point was recorded, and a photo of the site from the start point facing the end point of the transect was taken. Observers also described the habitat across the entire opening (e.g., number of trees, distribution of trees, percent shrub cover) and the surrounding habitat by type (e.g. upland forest, lowland forest, upland shrub), tree or shrub species, and coarse age class (young, middle, old). Vegetation was sampled along the right edge of the measuring tape (from the start point looking towards the end point). For each change in cover, the start distance to the nearest tenth of a meter (e.g., 1.1 m, 5.8 m), the cover type code, abundance, and height class was recorded (Table 1). For shrubs and trees taller than 1.5 m, the actual height to the nearest meter was recorded.

To assess the use of openings in this study by other wildlife (e.g. deer, bear) the presence of scat encountered within 0.5 m of the transect was recorded along with the distance along the transect and suspected species.

PRELIMINARY RESULTS/DISCUSSION

In the 2016 pilot project, singing ground surveys were conducted at 94 openings, with singing males observed at 55 openings (59%). The majority of occupied openings (42 of 55) had only 1 male present, 10 openings had 2 males, and 3 openings had 3 males. These findings suggest that a binary presence/absence assessment of opening use would be most appropriate. Twelve openings were surveyed for singing males on multiple occasions. Woodcock occupancy (presence or absence) remained the same at 10 of these openings (6 present, 4 absent), with the 2 remaining openings having woodcock present in the first visit but not in the second. Repeated surveys on multiple occasions would be ideal, however limited field staff and a short window of activity in the evening and breeding season to conduct surveys makes it difficult to both maximize the sample size of openings and conduct repeated surveys at all sites. Since woodcock use will be assessed as presence/absence for singing ground surveys, effort can be reduced by repeating surveys only at sites without woodcock use to determine if errors of omission exist.

Roosting ground surveys were conducted at 65 openings, and roosting woodcock were spotlighted and flushed at 15 openings (23%). In addition, woodcock were observed flying, landing in, or flushing from 46 openings (71%). Both flight and spotlighting surveys appear to provide useful information on woodcock use. Roosting surveys were not repeated due to time limits and sample size, but other research has found that the frequency of roosting field use by individual woodcock varies by month and by age and sex (Sepik and Derleth 1993). Sepik and Derleth (1993) found the highest frequencies of roosting field use in June and July. However, there was no significant relationship found between date and woodcock observed at roosting openings in this preliminary study in which surveys were conducted from June through August. Roosting surveys can only be conducted at one site per observer per evening, thus making repeated surveys difficult to accomplish with limited field staff. Nevertheless, roosting surveys should be repeated to determine how roosting use varies. Repeated surveys may decrease the total number of openings that can be included, however we currently have no information on how woodcock use of roosting grounds in this study varies. If repeated surveys are possible they would provide important information on the variability in woodcock use of roosting openings and the need to repeat surveys in future studies.

Exploratory data analysis was used to assess the independent variables collected. Kernel density plots and histograms were used to explore the distribution of variables to assess their value as predictors. Opening size (ac) and perimeter (m) were highly skewed with large outliers due to larger forest harvest sites, however the ratio of perimeter to acres was more normally distributed and might serve as a better variable in modeling. Plots for proportion in 9 cover type classes (grass, herbaceous, woody, shrub, tree, coarse woody debris, bare ground, moss, and other) and 6 height classes (0-3cm, 3-10cm, 10-30cm, 30-50cm, 0.5-1.0m, 1.0-3.0m. and >3.0m), as well as combinations of these, were explored. Most openings were dominated by grasses with few shrubs and trees. The cover of herbaceous vegetation seemed to be the most informative variable. Vegetation in the 2 shortest (<10cm) and 3 tallest (>0.5m) size classes was typically rare. The 10-30cm and 30-50cm classes had good variation and would seems most promising as variables. It was noted anecdotally in the pilot that sites heavily invaded by the exotic plant common tansy (*Tanacetum vulgare*) tended to have no woodcock present. This could have important implications for management and prevention of invasive species spread in wildlife openings.

Time since mowing was not always known for sites included in the pilot study. Most sites had recent mowing within 2 years (73 of 94 singing and 47 of 65 roosting openings). Future research should balance sample size and include more sites with longer time since mowing to allow for this important management consideration to be assessed. Excluding forest harvest sites from analysis of woodcock use of openings may be necessary since harvest sites differed markedly from openings in both size and cover, and represent only a small portion of the total sample size (8 sites). Anecdotal and qualitative comparison of harvest sites and openings would be more feasible for this study.

Weather conditions including cloud cover, temperature, wind speed, and precipitation were poorly distributed, partly due to restrictions for surveying conditions. Consequently, no significant relationships were found between woodcock observed and these metrics for either singing or roosting ground data. Date of survey and time since full moon also did not show a relationship with woodcock observed in the pilot data.

Forest wildlife openings are often clustered along hunter walking trails, and openings in the pilot study showed this clustering. To assess if woodcock use of openings showed spatial autocorrelation, Moran's I in ArcGIS was used. The number of woodcock per opening from singing ground surveys showed no spatial autocorrelation (I=0.05, P=0.38). Number of woodcock flushed in roosting ground surveys showed no spatial autocorrelation (I=-0.009, P=0.94), however minimum number of woodcock flying, landing in, or flushing showed a clustered pattern (I=0.21, Z=2.16, P=0.03). Due to the arrangement of openings, spatial autocorrelation should be tested for in future samples and included in modeling when present.

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Table 1. Codes used to describe vegetation composition and structure along transects sampled within managed forest openings Minnesota during 2016.

Cover type codes	Abundance codes	Height codes
G = Graminoid	R = Rare, < 25%	0 = < 3 cm
H = Herbaceous	S = Sparse, 25-50%	1 = 3-10 cm
W = Woody (<0.5 m tall)	M = Moderate, 50-75%	2 = 10-30 cm
S = Shrub (0.5 - 2 m tall)	D = Dense, > 75%	3 = 30-50 cm
T = Tree (>2 m tall)		4 = 0.5 - 1.5 m
B = Bare ground		
C = Coarse woody debris		

ECOLOGY AND POPULATION DYNAMICS OF BLACK BEARS IN MINNESOTA

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SUMMARY OF FINDINGS

During April 2016–March 2017, we monitored 14 black bears (*Ursus americanus)* previously radiocollared 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 Training Center (southern fringe), and a site at the northwestern (NW) edge of the range. Additionally, we collared 19 more bears (among a total of 28 captured) in the CNF. The young, male-biased capture sample in the CNF was indicative of heavy hunting pressure. Hunting has been the primary source of mortality in all areas, although vehicle collisions have been a significant source of mortality for bears wandering off Camp Ripley, which is flanked by highways. In the 2016 hunting season, 20−23% (depending on fate of 1 bear that disappeared) of collared bears were shot, even though hunters were asked to avoid killing collared bears, and they were all marked with conspicuously large, colorful ear tags.

Reproduction was strongly affected by food supply. The NW area had the highest reproductive rate, due to early maturity, large litters, and litter intervals rarely exceeding 2 years. Camp Ripley bears matured early but had the highest proportion of 3-year litter intervals. Litter sizes of 3 were most common in NW and CNF, whereas litter sizes of 2 were most common in VNP; in Camp Ripley, 3-year-old mothers all had litters of 2, whereas older mothers had an equal proportion of 2- and 3-cub litters.

Camera traps set outside den sites revealed dates of initial den emergence (22 Feb−24 Mar) and final departure from the den site (11 Mar−20 Apr). Bears spent 1−41 days going in and out of the den after they first emerged, before departing the area. Much of the activity outside the den—and an apparent motivation for coming out—was to collect dry bedding material. We suggest that early, warm springs, with melting snow, may prompt bears to emerge early from wet dens.

INTRODUCTION AND STUDY AREAS

Telemetry-based research on black bears was initiated by the Minnesota Department of Natural Resources (MNDNR) in 1981, and has been ongoing continuously 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*

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tremuloides, P. grandidentata) of varying ages. Camp Ripley Training Center, a National Guard facility at the southern periphery of the bear range, was added as a second study site in 1991. Camp Ripley is unhunted, but bears may be killed by hunters when they range outside Camp, which they often do in the fall. Oaks (*Quercus* sp.) are plentiful within Camp, and cornfields border the site. 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 VNP.

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 MNDNR 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, until recently, most other parts of the bear range have had stable or declining bear numbers; and (4) hunting pressure in this area is unregulated (it is within the no-quota zone, so there is no restriction on hunting licenses).

We used these 4 study sites to examine spatial variation in bear population dynamics and ecology to help inform bear management.

OBJECTIVES

1. Compare sources of bear mortality in different parts of the bear range.
2. Quantify temporal and spatial variation in cub production and survival.
3. Understand factors affecting emergence and departure from dens.

METHODS

During May−August, 2016, we captured bears in the CNF with barrel traps and immobilized them with ketamine-xylazine. During December–March, we visited all radiocollared bears once or twice at their den site and immobilized them with Telazol. For all handling, we measured and weighed bears, assessed body condition, took blood and hair samples, and extracted a vestigial first premolar to estimate age on all bears whose age was unknown (i.e., first handling of bears older than cubs). We changed or refit the collar, as necessary. We collared all new females and larger males that we thought would not disperse from the study area.

This year we used mainly GPS-Iridium collars (Telonics Inc., Mesa, AZ) or VHF collars with an attached GPS pod (Telemetry Solutions, Concord, CA), except in VNP where we used only VHF collars. All collared bears had brightly-colored, cattle-size ear tags (7x6 cm; Dalton Ltd., UK) that would be plainly visible to hunters. Bears that were not collared had small inconspicuous ear tags.

We 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. Licensed hunters could legally shoot collared bears, although they were asked not to. 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, and this request was also made through news releases. Requests to hunters to voluntarily not shoot collared bears have been made through the news media and MNDNR hunting regulations and website since 2001, although the individual letters to hunters was not initiated until 2011.

We assessed reproduction by observing cubs in March dens. We sexed and weighed cubs without drugging them. We quantified 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 monitored heart rates of a subset of bears using a new Insertable Cardiac Monitor developed for human heart patients (Reveal LINQ^{TM}, Medtronic Inc., Minneapolis, MN). 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 (see Laske et al. 2014). These data are not presented in this report, but will be used to inform our research questions about factors affecting den emergence.

We set remote cameras (camera traps; Reconyx, Inc., Holmen, WI) outside bear dens to gain information about dates and behaviors of bears emerging from dens and departing from the den site. Bears that emerged from dens <48 hours after our den visit were excluded from the analysis.

RESULTS AND DISCUSSION

Radiocollaring and Monitoring

As of April 2016, the start of the current year's work, we were monitoring 16 radiocollared bears: 1 in the CNF, 6 at Camp Ripley, 3 in VNP, and 6 in the NW (Table 1). Two NW bears lost their collars (1 breakaway link degraded after the bear could not be handled in the den, and 1 yearling removed the collar put on in the den). We captured 28 new bears (21M; 7F) in the CNF, and collared 19. We selectively collared all females and larger males. Only 4 bears were >5 years old (Figure 2). The heavy skew toward males and young age structure of the captured bears suggests that this area has been subjected to heavy hunting, and many of the bears had immigrated from elsewhere.

Mortality

Since 1981 we have recorded the cause of death for 367 radiocollared bears, 78% of which died (or likely died) from legal hunting (Table 2). Vehicle collisions are another significant source of mortality at Camp Ripley, which is flanked by 2 highways.

Despite our request (for the past 16 years) not to shoot collared bears (with large ear tags), 6 or 7 collared bears were shot by licensed hunters during September 2016 (Table 1). This represents 20−23% of the collared bears monitored at the time. The actual mortality rate is somewhat higher, as most of the collared bears at Camp Ripley were on the reserve, not available to hunters. Conversely, all 3 VNP bears were outside the park during the fall and photographed at hunters' baits (Figure 3). The high harvest mortality rate was a reflection of poor fall foods, and thus ready attraction to hunters' baits.

The bears shot by hunters were from all 4 study sites (Table 1), so the sample represents a large portion of the bear range. But since we are also aware of hunters who passed up shooting collared bears (observed or photographed at their baits), as per our request, it seems likely that the population at large was subjected to an even higher mortality rate than the collared bears.

However, among 18 bears that we ear-tagged but did not collar this year, none were reported killed by hunters. We suspect that the reporting rate of tags is less than for collars, but if they were subjected to the same mortality rate as collared bears, it seems somewhat enigmatic that ~4 would not be reported, especially since we only requested that hunters refrain from shooting bears with collars and large ear tags (bears without collars had very small ear tags that would be difficult to see before shooting). One explanation is that only 2 of the 18 ear-tagged bears were >2 years old, so in a year when many bears were at hunters' baits, hunters could select for larger bears. Only 1 of the hunter-killed collared bears was <4 years old.

Reproduction

Since 1982, within the 4 study areas, we have checked 290 litters with 745 cubs ($\bar{x}$ = 2.6 cubs/litter), of which 50.7% were male (Tables 3–6). The sex ratio of cubs has become female-biased in all study areas except the CNF, where we have checked an average of only 1 litter per year for the past 10 years. The increased collaring in the CNF this year will provide a larger sample of reproductive bears in the future.

At Camp Ripley, all 5 collared females produced cubs in 2015, so none produced in 2016; all but 1 of these produced cubs in 2017. The 1 bear that did not produce cubs was unusual: since 2005, when this bear was 3 years old, she has produced a litter every-other year (6 litters, 15 cubs), and her body condition was more than ample to produce cubs this year (44% body fat, 365 lbs in Dec; 324 lbs in Mar). We know that this bear has been a nuisance, and has been shot at, although we did not see any obvious injuries, other than a healing wound from where we removed an archery broadhead from her shoulder last year. Overall, bears at Camp Ripley, despite being large, have had a higher rate of missed litters (3-year litter intervals) than bears in the other study sites (Table 7).

One of 2 collared females in NW produced cubs in 2017 (the other had yearlings). Both collared bears in VNP were too young to have cubs (both 3 years old; no VNP bears have produced cubs at 3). One 6-year-old bear in the CNF produced her first cubs; 3 CNF 4-year-olds did not have a first litter yet. However, 2 bears caught in the CNF had cubs with them in summer 2016, 1 of which was 4 and 1 was only 3 years old. Of 91 female bears in the CNF monitored through 3 years old, this is only the third to have cubs at this young age.

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 4). This is somewhat ironic in terms of Minnesota's bear management, given that the NW study site is outside "core" bear range and, accordingly, is within a management zone where bear hunting license sales are unrestricted (no-quota). The NW site contains not only agricultural crops consumed by bears, but also an abundance of natural foods, especially along the edges of woodlots (Ditmer et al. 2015). In all areas except the NW, reproductive rates were higher for ≥7-year-old bears than 4- to 6-year-olds because many bears in this younger age group either had not yet reproduced or just had their first litter, which tended to be smaller (fewer cubs). The most striking differences among study sites were in the reproductive rates of these 4–6 year-olds (Figure 4).

Bears in the CNF and NW produced more 3-cub litters than 2-cub litters, whereas 2-cub litters were most common at Camp Ripley and VNP (Figure 5). The relatively small litter sizes at Camp Ripley were due to many of those bears producing cubs when only 3 years old (all 3-year-old mothers had litters of 2 cubs). Eliminating these bears, litter sizes of 2 and 3 cubs were about equal at Camp Ripley (Figure 5).

Age of first reproduction was dramatically different among areas. By 4 years of age, >80% of bears at Camp Ripley and in the NW had produced surviving cubs (observed in the den at 1 year; Figure 6). Only 37% of bears on the CNF produced surviving cubs by 4 years old and no bears at VNP produced cubs by 4 years of age. Camp Ripley bears sacrificed litter size for earlier age of reproduction (Figures 5 and 6). NW bears had both large litters and early age of first reproduction, so were most prolific of all the sites.

Mortality of cubs during their first year of life averaged 19% (annual range 0–31% for years with at least 10 cubs monitored), with mortality of male cubs (25%) exceeding that of females (16%; $\chi^2 = 6.38$, $P = 0.01$). The timing and causes of cub mortality are unknown.

Camera Trap Photos at Dens

We obtained camera-trap photos of bears that yielded dates of natural emergence and departure from 14 dens: 1 in 2015, 6 in 2016, and 7 in 2017. Dates of first emergence ranged from 22 February to 24 March (Figure 7). After first emergence (which we defined as completely exiting the den, not just poking their head out), bears remained at the den site for 1−41 days. This span of time is similar to that reported by Miller et al. (2016; 0−47 days) for 21 black bear dens monitored with camera traps in Utah. Between the time of emergence and eventual departure from the den site, bears moved back and forth between the den site and outside the den.

When outside the den, but before leaving the vicinity of the den (defined as beyond the detection of the remote cameras), bears were involved in the following principal behaviors: raking more bedding material into the den, stretching/walking, laying in the sun, eating snow or drinking water, monitoring cubs playing and climbing trees. We thus interpret the period between den emergence and departure to be a time where bears: (1) attempt to stay dry in the den while snow is melting and causing some discomfort; (2) regain muscle strength; (3) warm body temperature; and (4) rehydrate. Often, in March, we observed bears poking their head outside the den to lick snow while not coming completely out of the den (Figure 8).

Miller et al. (2016) quantitatively assessed the proportion of time that bears in Utah invested in various activities outside the den, most of which was simply standing or walking, suggesting that muscular activity was important. Although we have not yet quantified time by activity, the photographs in our study indicate a substantial investment in gathering more bedding material. In fact, it often appeared that the primary reason for coming out of the den was to get more bedding, apparently because the den had gotten wet—in some cases, the photos showed that bears had gotten wet from water in their dens (Figure 9). Bears in Utah also commonly augmented their nests, although they spent comparatively little time doing so. Miller et al. (2016) did not mention whether dens in Utah were wet.

The span of time over which individual bears departed the den site (11 March to 20 April) was even wider than the span of dates of first den emergence times. We suspect that bears employed different thresholds for leaving. Bears with young cubs tended to stay until cubs were mobile and able to climb trees. Solitary bears or mothers with yearlings often waited for most of the snow to melt; however, some did not. In one case, an adult male bear was in an excavated den that seemed to have gotten wet, and it left the vicinity of the den only 1 day after first emerging, despite the surrounding area being totally snow-covered (Figure 10). The photos show that the bear was wet, and there was no obvious place near the den to obtain more bedding material (grass, leaves or conifer boughs). Conversely, bears that were able to rake in copious bedding material had more incentive to remain. We speculate that with warmer weather in late winter, issues of wet dens will become an increasing concern for bears, and those that cannot rake in dry bedding material will be prompted to leave the den site earlier than they might otherwise choose. In an extreme case, 1 study bear was flooded from its den during a thaw in December. The water pooled and froze in the den, and the bear apparently sat outside the den all winter. Possibly as a reaction to flooding underground dens, we note that an unusually low proportion of CNF bears denned underground (2 males, 2 females of 15 total dens = 27% in 2016; 56% of females denned underground in the 1980s; Garshelis 1987).

Above-ground dens may be drier but also expose the bear to direct stresses of the weather (e.g., rain or snow on their back) and possible confrontations with predators. In one case a dog visited an occupied den under a root mass, but remained outside. In another unusual case, a bear in a brush pile left to retrieve a leg from a dead deer. It simply rolled around on top of the leg, but never ate it. Shortly after the bear departed the den, a wolf came by and devoured the deer leg (Figure 11).

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Table 1. Fates of radiocollared black bears in Chippewa National Forest (CNF), Camp Ripley, Voyageurs National Park (VNP), and northwestern Minnesota (NW) study sites, April 2016−March 2017.

	CNF	Camp Ripley	VNP	NW
Collared sample April 2016	1	6	3	6
Trapped	19			
Killed in vehicle collision	1			
Killed by Minnesota hunter[a]	3 or 4[b]	1	1	1
Natural mortality				
Dropped radiocollar				2
Failed radiocollar	0 or 1[b]			1[c]
Collared in den				
Collared sample April 2016	15	5	2	2

[a] Hunters were asked not to shoot collared bears (although it was still legal).
[b] One GPS-Iridium collared bear disappeared after the first week of the hunting season (either shot and not reported or collar failed; categorized as 'likely shot by hunter' in Table 2).
[c] One GPS-Iridium collared bear disappeared in mid-July; we suspect that the radiocollar failed.

Table 2. Causes of mortality of radiocollared black bears ≥1 year old in 4 Minnesota study sites, 1981–2017. 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[a]	230	13	16	14	273
Likely shot by hunter[b]	9	1	0	4	14
Shot as nuisance	22	2	1	3	28
Vehicle collision	13	9	1	3	26
Other human-caused death	9	1	0	0	10
Natural mortality	8[c]	3	5	0	16[c]
Died from unknown causes	4	2	0	3	9
Total deaths	295	31	23	27	376

[a] Since 2001, the MNDNR has asked hunters not to shoot collared bears, so the proportion killed due to this cause is no longer representative of the population at large.
[b] Lost track of during the bear hunting season, or collar seemingly removed by a hunter.
[c] 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, Minnesota during March, 1982–2017. High hunting mortality of radiocollared bears severely reduced the sample size in recent years.

Year	Litters checked	Number of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 year[a]
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%	—[b]
2015	0	0	—	—	—
2016	0	0	—	—	—
2017	1	3	3.0	—	—
Overall	180	475	2.6	53%	19%

[a] Cubs that were absent from their mother's den as yearlings were considered dead.
[b] Mother was killed by a hunter so status of cubs unknown.

Table 4. Black bear cubs examined in dens in northwestern Minnesota during March, 2007–2017.

Year	Litters checked	Number of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 year
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%	33%
2015	2	5	2.5	60%	0%
2016	2	6	3.0	50%	0%
2017	1	3	3.0	0%	
Overall	31	86	2.8	43%	17%[a]

[a] 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 Training Center, Minnesota, during March 1992–2017.

Year	Litters checked	Number of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 year[a]
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[b]	—[b]	—	—	—
2015	6	15	2.5	20%	10%
2016	0	0	—	—	—
2017	4	10	2.5	60%	—
Overall	50	119	2.4	49%	20%

[a] Blanks indicate no cubs were born to collared females or collared mothers with cubs died before the subsequent den visit to assess cub survival.

[b] Cubs heard, litter not handled. Camera set outside den indicated that all cubs died. This litter not included in total.

Table 6. Black bear cubs examined in dens in Voyageurs National Park, Minnesota, during March 1999–2017. All adult collared females were killed by hunters in fall 2007, so no reproductive data were obtained during 2008–2009.

Year	Litters checked	Number of cubs	Mean cubs/litter	% Male cubs	Mortality after 1 year[a]
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%	0%
2015	0	0	—	—	—
2016	0[b]	0	—	—	—
2017	0	0	—	—	—
Overall	29	65	2.2	48%	25%

[a] 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.
[b] One bear that likely had cubs was not checked because access to her den was precluded by poor ice conditions.

Table 7. Intervals between surviving litters for black bears within 4 study sites in Minnesota (see Figure 1) through March 2017 (CNF since 1981, Camp Ripley since 1991, VNP since 1997, NW since 2007). Cubs are generally born in January and remain with their mother for about 17 months, so the normal reproductive interval is 2 years. Reproductive intervals here include only litters where at least 1 cub survived through the next denning period (1 year), so intervals <2 years are impossible.

Study area	2-year reproductive intervals	≥3-year reproductive intervals	% intervals ≥3 years
CNF	110	8[a]	7%
Camp Ripley	26	5	16%
VNP	15	1	6%
NW	17	0[b]	0%

[a] Including the only case of an interval spanning >3 years, due to whole litter loss followed by a non-reproductive year.
[b] Excluding 1 missed litter (3-year interval) that was due to the bear leaving the den after disturbance and aborting the litter.

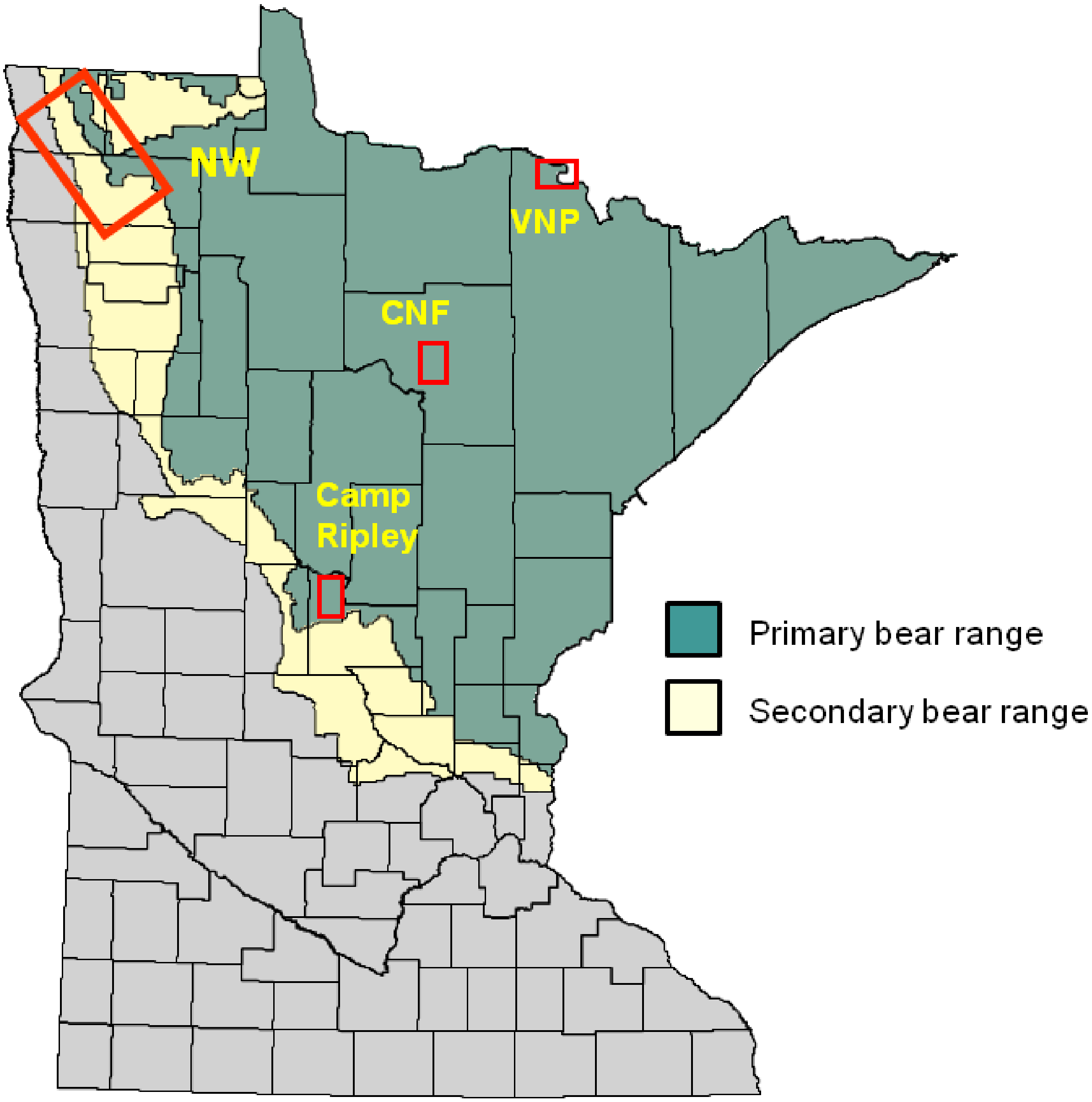

Figure 1. Location of 4 study sites within Minnesota's bear range: CNF (Chippewa National Forest, central bear range; 1981–2017); VNP (Voyageurs National Park, northern fringe of range; 1997–2017); Camp Ripley Military Reserve (near southern edge of range; 1991–2017); NW (northwestern fringe of range; 2007–2017).

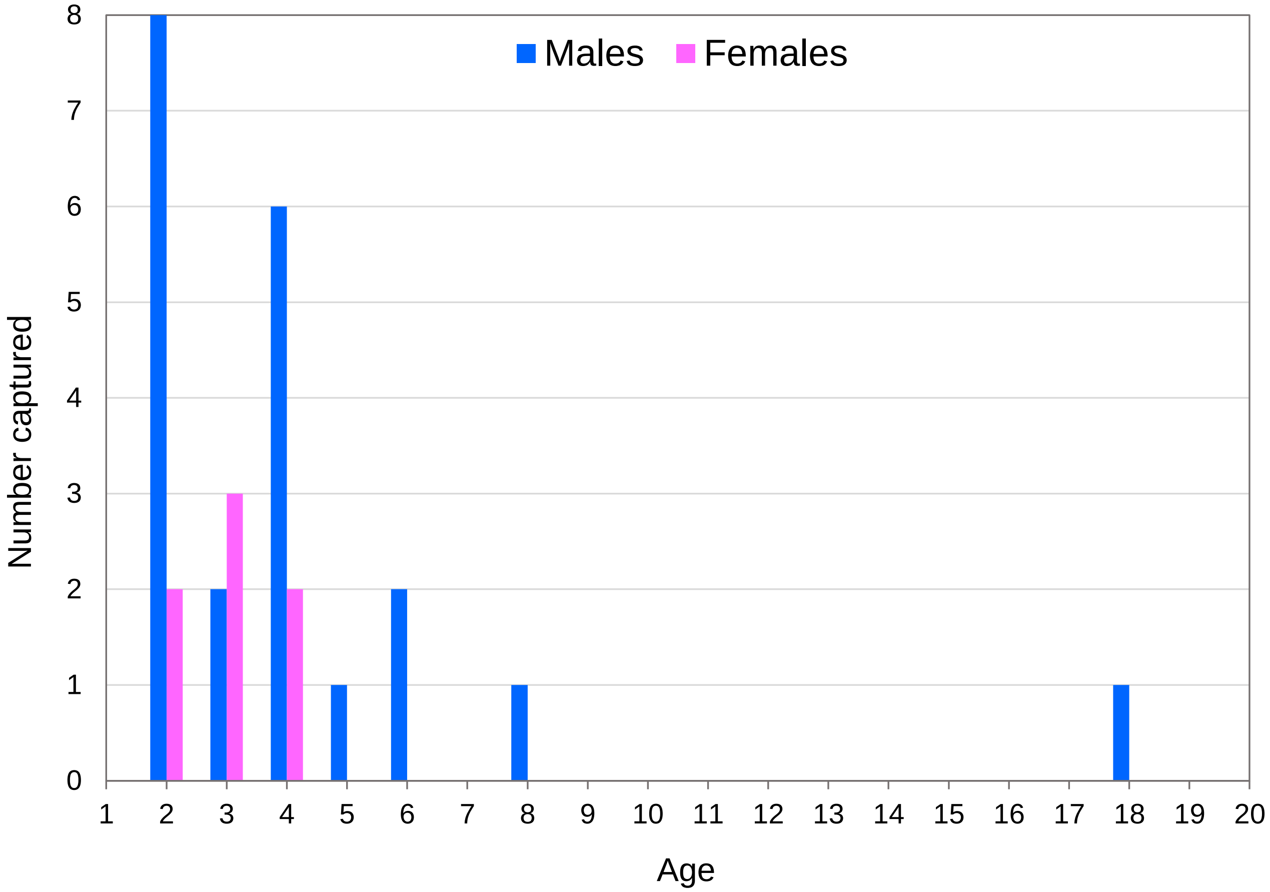

Figure 2. Bears captured by sex and age in the CNF, Minnesota, May−Aug, 2016. All 7 females were collared. Ages were not known at time of capture, so among males, we selected bears to collar based on weight and capture location (not collared: 7 of 8 2-year-olds, 1 4-year-old, 1 6 year-old).

Figure 3. All 3 collared bears at VNP, in Minnesota, visited hunters' baits well outside the park during the 2016 hunting season. A hunter selected the adult female (top). Hunters passed up on the 2 subadult females. (Trail camera photos courtesy of K. Keeler).

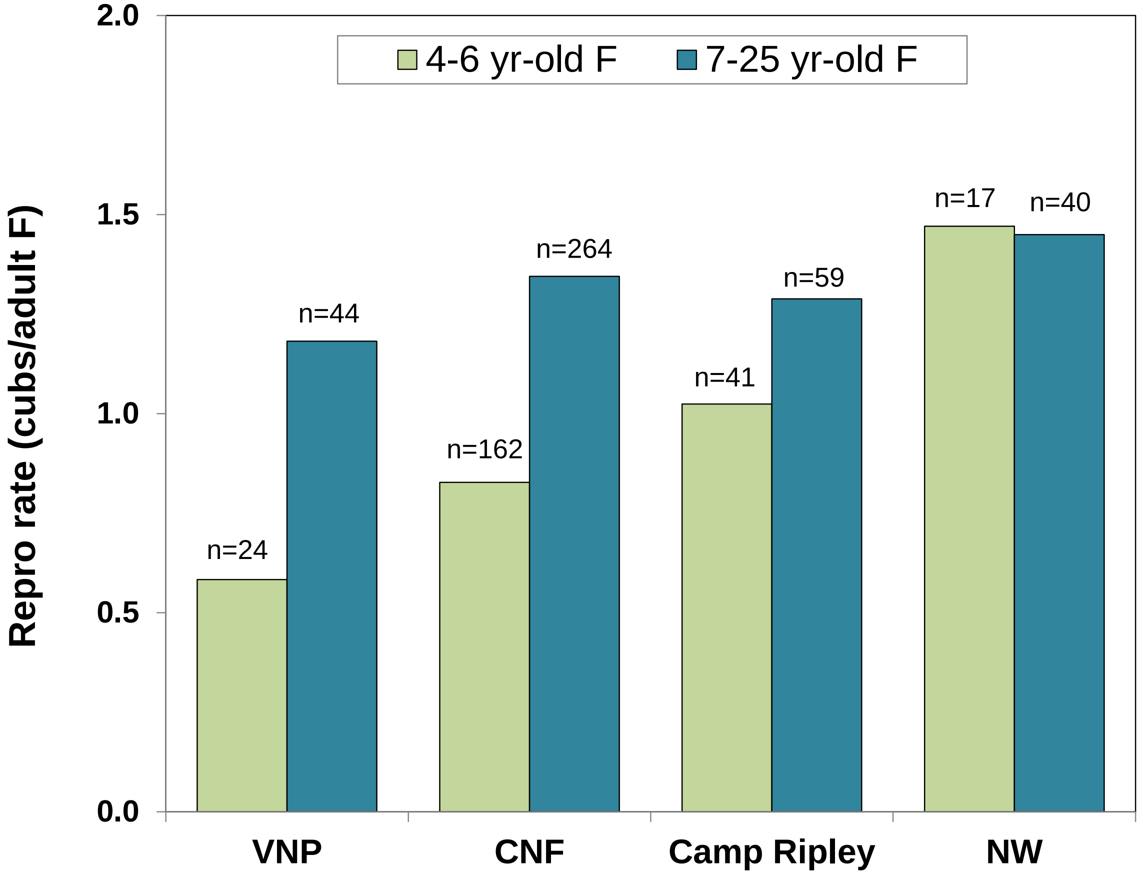

Figure 4. Reproductive rates of radiocollared bears within 4 study sites (see Figure 1) through March 2017 (VNP, Minnesota, since 1997, CNF since 1981, Camp Ripley since 1991, NW since 2007). Data include only litters that survived 1 year (even if some cubs in the litter died). Sample sizes refer to the number of female bear-years of monitoring in each area for each age group. Some bears in CNF, Camp Ripley, and NW produced cubs at 3 years old, but are not included here.

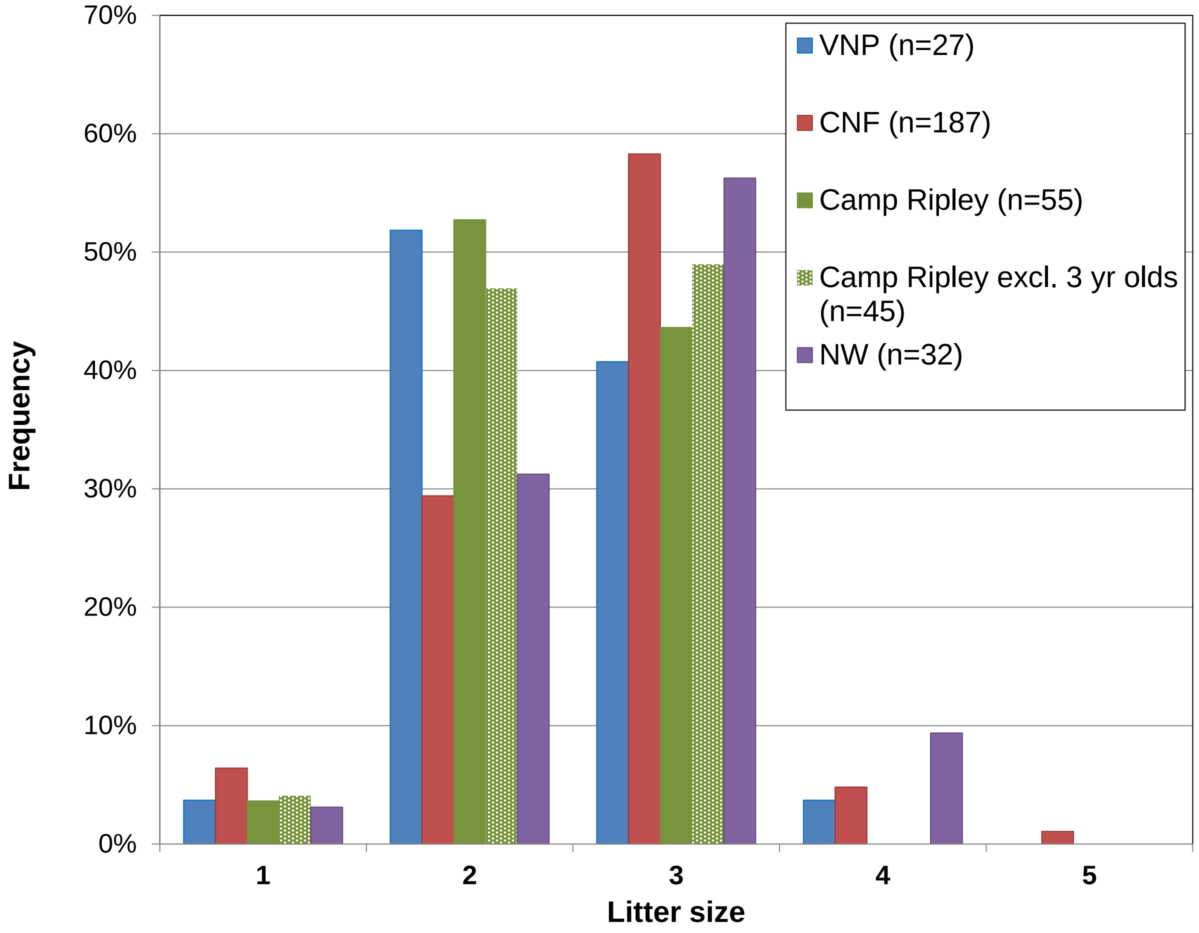

Figure 5. Frequency of cub litter sizes (examined in natal dens in March) within 4 study sites (see Figure 1) through March 2017 in Minnesota. Data include only litters that survived 1 year (even if some cubs in the litter died). Camp Ripley data are shown for mothers of all ages, as well as excluding 3-year-old mothers. For the other sites, elimination of 3-year-olds did not make a difference.

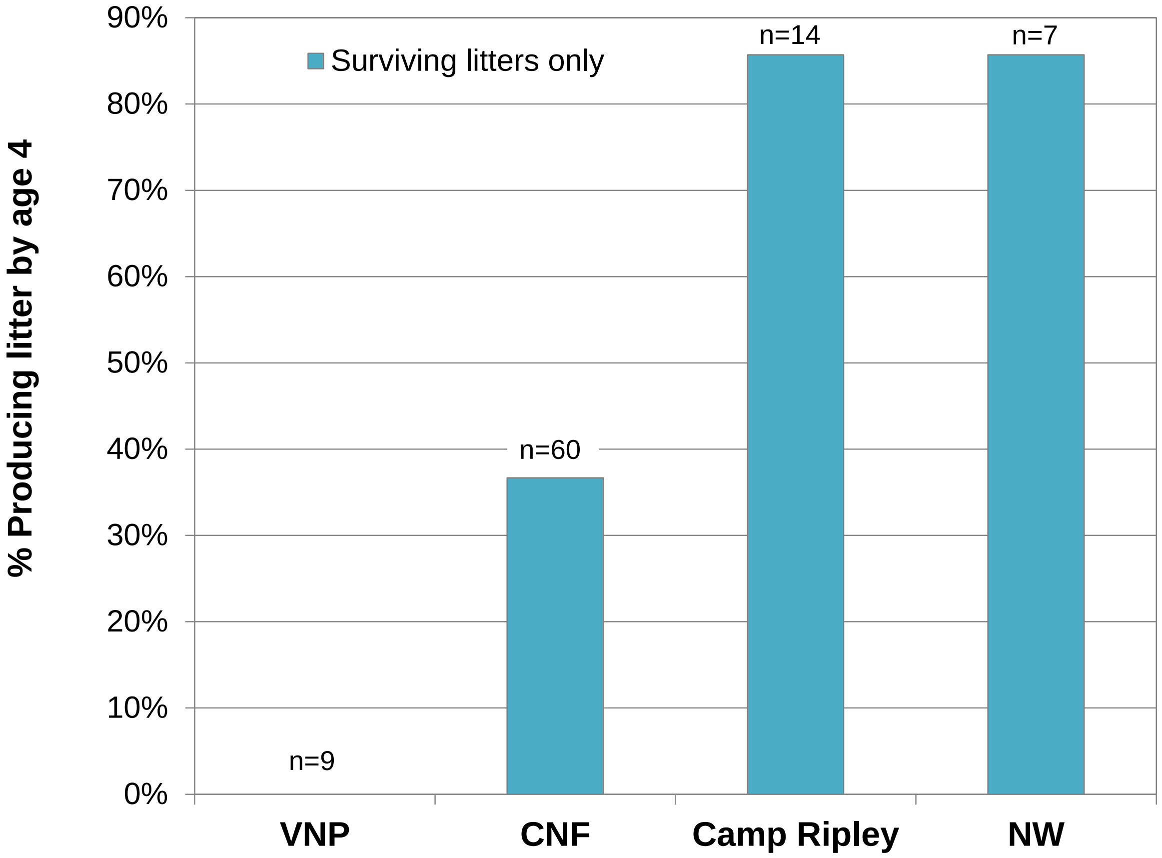

Figure 6. Percent of radiocollared females on each Minnesota study site that produced a surviving litter of cubs by 4 years old. Births of cubs were detected in natal dens in March each year (through March 2017). A surviving litter was one in which at least 1 yearling was present in the mother's den the next winter. Note that no females in VNP produced cubs by 4 years of age.

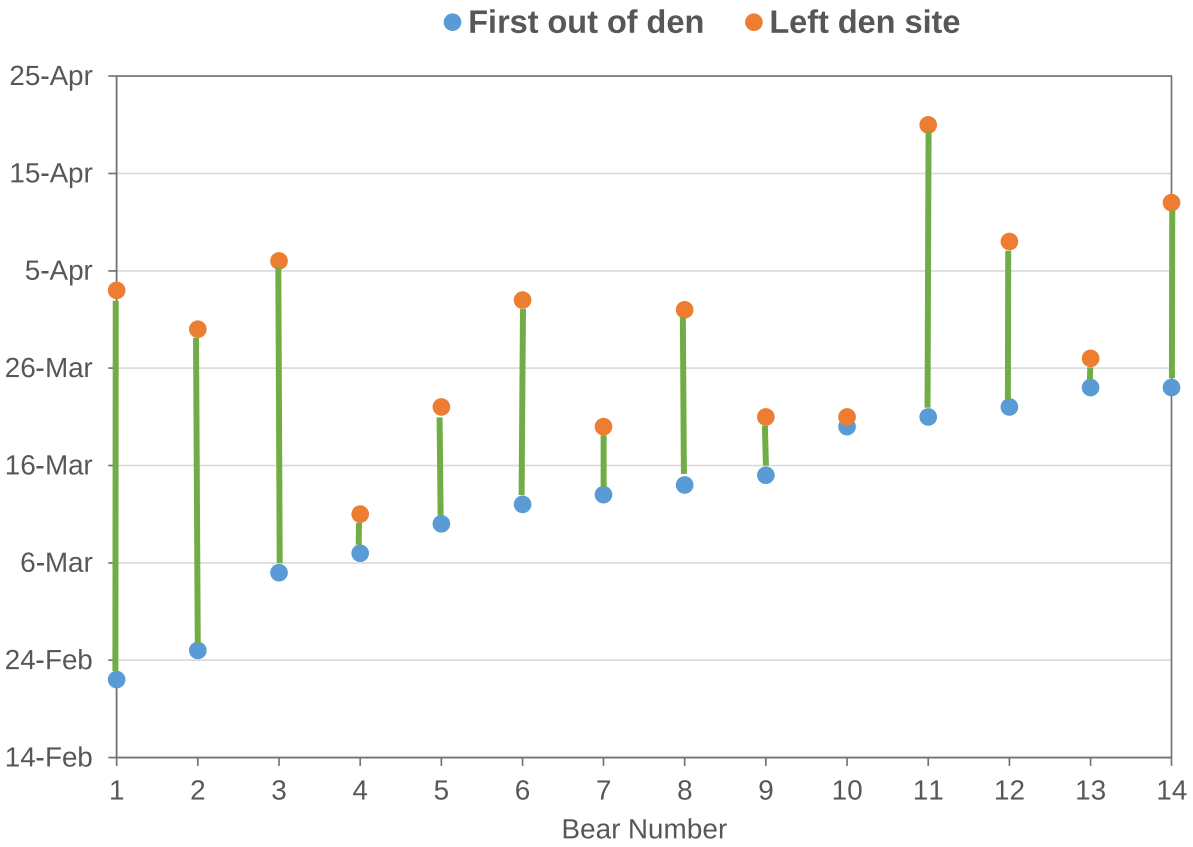

Figure 7. Dates of first emergence from dens, and eventual departure from the den site, for 14 radiocollared black bears monitored with remote cameras in Minnesota, 2015−2017. Green lines show the time period (1−41 days) that the bear remained at the den site following initial emergence.

Figure 8. Hibernating bears do not eat or drink through the winter, but in the month before leaving the den site, they sought to rehydrate, as shown by this camera-trap photo of a CNF bear emerging from its underground den to lick snow, March 12, 2017 Minnesota.

Figure 9. Camera-trap photos of Minnesota’s CNF bears emerging from dens showed that they frequently sought dry bedding material as melting snow leaked into their dens. Top row: mother with cubs in nest den pulling in spruce boughs. Middle row: mother with wet hind end coming out on March 22, 2017, to pull in a small balsam twig. Bottom row: same mother coming out again a week later, with water dripping from her hind end as well as that of her yearling; last photo shows mother rearranging the nest at the front of the den (original nest was in the rear) while the yearlings explored outside.

Figure 10. Camera trap photo of a large adult male in an underground den in the CNF in Minnesota, first peering out of its den on March 14, 2017, seemingly to assess the conditions outside (we did not consider this to be den emergence because it did not come completely out of the den). A week later, clearly very wet (bottom photo), the bear fully emerged, and left a day later, despite the area being still totally snow covered.

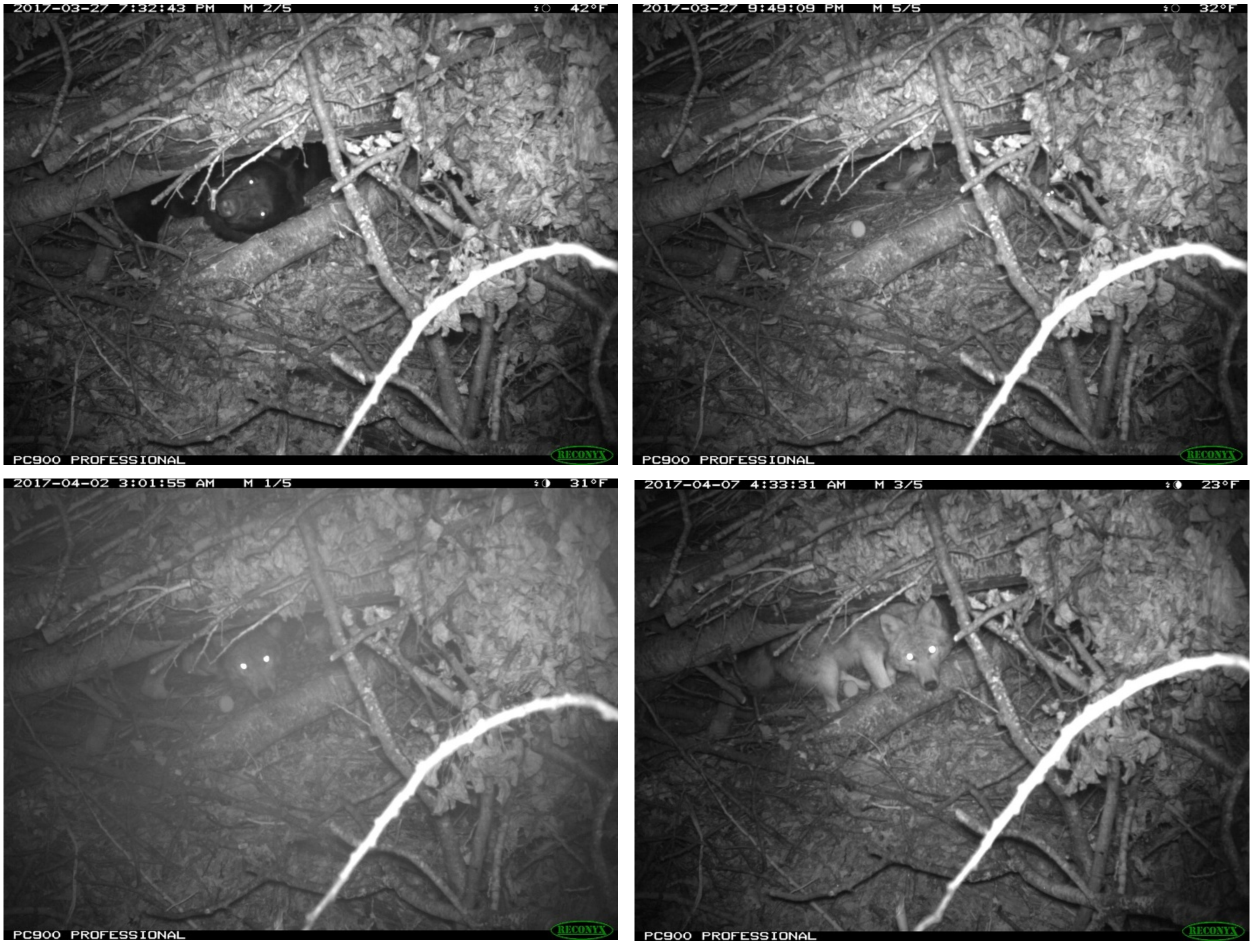

Figure 11. A young male bear, denned in a brush pile in the CNF in Minnesota, retrieved a leg of a dead deer and rolled around on it (top left; leg visible top right after bear left den, March 27, 2017). A few days after the bear departed, a wolf entered the den and ate the leg (bottom left, April 2). Five days later a wolf entered the den again, even though the deer leg was gone.

HAVE CHANGES IN FOREST COMPOSITION ALTERED FOOD ABUNDANCE, HABITAT USE, AND REPRODUCTION IN BEARS?

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SUMMARY OF FINDINGS

During the summer of 2016, we measured abundance (coverage) of black bear (*Ursus americanus*) food-producing plants and their production of fruits and nuts in 12 main forest types in and around the Chippewa National Forest, north of Grand Rapids, MN. We used the same methodology in the same area that was sampled through the 1980s and found that in most forest types, key bear foods were greatly reduced. This was true for both the abundance of the plants as well as the fruit production, resulting in greatly diminished biomass of bear foods in the forest. This change was likely due, in part, to changes in forest age and composition; however, even in young stands (e.g., 5−15 year-old aspen), bear foods were greatly reduced compared to the same stand types in the 1980s. Unexpectedly, we did not witness obvious changes in body condition between bears in the 1980s and those we captured this summer (n = 28). One explanation may be that bears are now exploiting more human-related foods. We observed, for example, that 70% of bears with GPS radiocollars (10 of 14) selected areas in proximity to hunters' baits in the fall.

INTRODUCTION

Population growth of black bears (*Ursus americanus*) in Minnesota is affected by hunting pressure, food availability, and by the interaction between these (bears are more susceptible to hunting when natural food supplies are low; Noyce and Garshelis 1997, Garshelis and Noyce 2008). Food availability affects bear body mass and condition, which in turn influences reproduction (Noyce and Garshelis 1994, Costello et al. 2003) and thus the resilience of the population to hunting (Kontio et al. 1998). Bear foods can vary enormously year-to-year due to year-specific environmental conditions (Noyce and Coy 1990). Additionally, bear foods on the landscape are subject to long-term trends with changes in forest age (canopy closure) and forest composition. For example, on an island in Washington state, reduced food supplies associated with forest succession led to a crash in black bear numbers (Lindzey et al. 1986). In the only other example of "bottom-up" control of bear numbers that we know of, grizzly bears (*Ursus arctos*) in southern British Columbia, Canada, relied heavily on huckleberries (*Vaccinium membranaceum)* and their body condition and population fitness was strongly affected by production of this single food item (McLellan 2011). When the forest was young, grizzly bear reproduction and density at this site were among the highest in North America, but as trees shaded out the huckleberries, bear reproduction plunged (in just 10 years) to one of the lowest in North America, and bear density declined by 36% (McLellan 2015).

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Here we examine long-term changes in availability of bear foods, and their effects on a bear population in Minnesota. In 1981, we initiated a long-term study of bears in the Chippewa National Forest (CNF), near the geographic center of the bear range. From the early 1980s through the early 1990s, we studied bear use of different habitat types, and also examined their body condition and reproduction (Noyce and Garshelis 1994). Concurrently, we collected data on bear food production by habitat type (Noyce and Coy 1990). Since then, forests on the CNF study site appear to have changed radically: food-rich stands of young aspen (*Populus* spp.) and young pine (*Pinus* spp.) plantations, which were common in the 1980s, have matured, and much of the upland forest is now dominated by maple (*Acer spp.*), which produces little food for bears. Concurrently, the CNF population declined drastically (MN DNR, unpublished data); we do not know whether this was attributable solely to over-hunting, or if habitat-related changes also played a role. Population modelling suggests that despite similar harvest pressure, the statewide population appears to be growing much more slowly than it did in the 1980s (Garshelis and Tri 2017). This study aims to investigate whether changes in habitat composition are affecting population growth.

OBJECTIVES

1. Quantify and assess changes in natural food abundance over the past 30 years.
2. Ascertain how bears have reacted to changes in food abundance.

METHODS

This study is being conducted primarily in the Chippewa National Forest, north of Grand Rapids, Minnesota. The ~130-mi^2 study area also includes a patchwork of state, county, and private lands, which are managed differently (e.g., more timber cutting) than the CNF.

This study involves the collection of 2 general types of data: (1) availability of bear foods by habitat type, and (2) responses of bears to altered food supplies. We will make comparisons from the early study period (1980s) to the present (2015–17). Data collection involves: (1) assessing fruit production in all major habitats; (2) capturing and fitting bears with GPS-Iridium collars to assess habitat use, movements, and home range size; and (3) visiting bears in their winter dens to measure body condition and reproduction. Results of den visits are addressed in a companion report (Garshelis et al. 2017).

Availability of Bear Foods

We visually assessed fruit production of 19 species (or species groups; Table 1) during the fruiting season (July–Aug, 2016) within fairly homogeneous stands of 12 different types of forests (the predominant forest types on the study area; Table 2). We situated 12 circular plots (3-m radius) within each stand such that 1 row of 4 were along the edge of the stand (if there was a clear edge), where light penetration was greatest and fruit production expected to be highest. We separately rated abundance (areal coverage within the plot) on a 0−4 scale (0= absent; 4= 67−100%), and production of fruits (0 = no fruit; 4 = bumper crop) for each of these bear food species. We matched our scale to that of Noyce and Coy (1990), who sampled in the same area in the same way during 1984−1989. We estimated biomass of each type of fruit within each stand using Noyce and Coy's (1990) counts of fruits and measurements of mass corresponding to each productivity rating. We used Kruskal-Wallis tests to compare biomass estimates in this study with that of Noyce and Coy during the 1980s.

Bear Responses to Foods

During May–Aug 2016 we captured a sample of bears in barrel traps and immobilized them with ketamine-xylazine. We measured, weighed, assessed body condition using bone prominence and skin-fold thickness (Noyce et al. 2002), and quantified body fat with bioelectrical impedance analysis. We extracted a vestigial first premolar to estimate age. We ear-tagged all captured bears and collared all females and larger (older) males that we thought would not disperse from the study area. We used GPS-Iridium collars (Telonics Inc., Mesa, AZ) or VHF collars with an attached GPS pod (Telemetry Solutions, Concord, CA 94520), programmed to obtain locations at 2-hour intervals.

We postulated that diminished natural food supplies (compared to the 1980s) could affect bears in a number of ways: (1) result in reduced body fat and general condition; (2) prompt bears to expand home ranges; (3) prompt bears to select different habitats; (4) entice bears to rely more on human-related foods. We will test each of these hypotheses, but for this report we only deal with the last one. As an initial investigation into the use of human-related foods, we investigated bears' use of hunters' baits. Licensed hunters are permitted to set baits in mid-August, about 2 weeks prior to the start of the hunting season on September 1. They must register the location of their baits, so we were able to overlay these sites with the GPS locations of bears over the same time frame (12 Aug–15 Oct). We compared the proximity of bear locations to hunters' baits and to random points in the study to create resource selection functions (RSF; Manly et al. 2002). We used RSFs to quantify the degree of attraction of each individual bear to hunters' baits for 3 periods within the day (diurnal, nocturnal, crepuscular). We generated 95% kernel density estimates (Worton 1989) for GPS locations of each bear within these daily periods to delineate period-specific home ranges, and generated 1 random point per hectare within each home range as the availability data for the RSF. We controlled for confounding variables such as habitat type, distance to roads and trails, and how recently a timber harvest occurred in the area.

RESULTS

Bear Foods

We conducted fruit surveys in 102 stands in 2016 (Table 2). These, combined with the 68 stands sampled in 2015, showed a significant decline in both the abundance and productivity of bear foods since the 1980s. Raspberry and sarsaparilla, 2 important early-summer foods whose abundance (coverage) varies year to year, were much less abundant in 2015 and 2016 compared to even the lowest years of the 1980s (Figure 1). Conversely, the abundance of beaked hazel and round-leaf dogwood, both tall woody shrubs that do not die-back over winter, did not vary much in abundance year to year, and showed no trend in abundance across decades (Figure 2). However, fruit production of all species varied enormously year to year (Figures 3–4). Although beaked hazel remained abundant in the study area, hazelnut production was extremely low in both 2015 and 2016 (Figure 4).

Combining abundance and productivity scores, we derived biomass estimates (kg/ha) that were extraordinarily low in both 2015 and 2016; this was true even for young forests, which were also much less common on the landscape. Red pine plantations (8−20 years old), previously a robust source of raspberries, blackberries, and chokecherries, had almost nonexistent bear foods the past 2 years (Table 3, Figure 5). Likewise, regenerating aspen stands (5−15 years), a previous source of abundant raspberries, round-leaf dogwood, and hazelnuts, had greatly diminished biomass in 2015 and 2016 (Table 3, Figure 6). Overall, hazelnut biomass in the forest in 2015 and 2016 was about an order of magnitude less than the average in the 1980s, and even less than the poorest year of that decade (1985, Table 3).

Bear Responses to Foods

We captured 28 bears (21M; 7F), and collared 19 (12M; 7F); 1 was killed in a collision with a car, 3 GPS units failed, and 3 bears were shot by hunters. We have not yet completed a rigorous comparison of sex- and age-specific weights or body condition with bears from the

1980s, but our impression is that despite the poor foods, recently captured bears were in good condition, especially in winter. One explanation for this may be their use of hunters' baits: 70% of bears (10 of 14 with adequate data during the fall) exhibited a significant selection for proximity to bait piles. Both females (88% of bear-time period combinations, *n* = 7 bears) and males (63%, *n* = 7) selected for hunters' baits during at least 1 time period (Figure 7). Male and female confidence intervals on RSF coefficients overlapped, but males intersected zero, whereas females did not.

Four bears (3M, 1F) consistently used areas distant from hunters' bait piles (positive coefficients, Figure 7). One of these (#6026) migrated 75 miles to corn fields near Brainerd and remained there for nearly 2 months (mid-Aug to mid-Oct) before returning to the study area (Figure 8). Another (#6005) was known to visit birdfeeders and dumpsters. A third (#6015) lived in a lowland swamp, where people generally do not hunt; notably, this bear's yearlings were much smaller (34 and 40 lbs) than those of a nearby female who visited baits (Figure 7, #6016; 72 and 91-lb yearlings). The fourth bear that stayed away from baits (#6007) was particularly thin in its den.

DISCUSSION

This study provided strong evidence of a reduction of bear foods on the Chippewa National Forest since the 1980s. Some of the reduction in biomass was due to especially poor production of fruits in 2015 and 2016, and some due to diminished abundance of some key fruit-producing plants. Through further research we aim to discern why this occurred. Our initial hypothesis appears at least partly correct, that increased forest age and altered forest composition, with less light penetration and less edge, has altered both abundance and productivity of many bear foods. Notably, we intentionally situated some of our sampling plots along the forest edge, and we observed that this is where fruit abundance was highest. The fruit surveys in the 1980s did not do this, so we only compared our interior forest plots to the data from the 1980s. Beyond the effects of more mature forests, we found that even young stands (e.g., aspen regeneration; Table 3), produced a low biomass of key foods in 2015 and 2016.

Possibly in response to the extraordinary reduction in natural bear foods, bears readily took advantage of human-related food sources, especially hunters' baits. We cannot conduct a comparable analysis of proximity of bear locations to hunters' baits for the 1980s because those bait locations are unavailable (and also, the 1980s bears had VHF collars, which were located by airplane weekly, versus every 2 hours for GPS collars). Therefore, the high use of hunters' baits that we observed here may or may not represent a change in bear behavior. Notably, only 3 of the bait-using bears were shot by hunters because we asked hunters not to shoot collared bears (all of which were prominently marked with large, colorful ear tags).

The migratory movement of one bear to a cornfield this year was not novel; in fact, during the 1980s we observed this commonly (averaging ~40% of bears each year, range = 3−87% among years; mean male movement 16 miles; Noyce and Garshelis 2011). However, most migrations in the 1980s were to southerly oak stands, not human-related food sources. We hope to investigate possible changes in use of human-related foods through stable isotope analysis of hair samples collected in the 1980−1990s versus the present.

ACKNOWLEDGMENTS

We thank the field assistants who helped capture bears and survey bear foods: Daniel Dewey, Charles Fortier, Sean Konkolics, and Michael McMahon. This project was funded in part by the Wildlife Restoration (Pittman-Robertson) Program grant W-68-D-15.

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Table 1. Bear food-producing plants sampled in the Chippewa National Forest, Minnesota 2015 and 2016.

Common name	Scientific name
Sarsaparilla	*Aralia nudicaulis*
American Spikenard	*Aralia racemosa*
Currant	*Ribes spp.*
Gooseberry	*Ribes spp.*
Blueberry	*Vaccinium spp.*
Red raspberry	*Rubus idaeus*
Common blackberry	*Rubus allegheniensis*
Red elderberry	*Sambucus racemosa*
Juneberry	*Amelanchier spp.*
Pin cherry	*Prunus pensylvanica*
Chokecherry	*Prunus virginiana*
Wild plum	*Prunus americana*
Alder-leaved buckthorn	*Rhamnus alnifolia*
Highbush cranberry	*Viburnum trilobum*
Downy arrowwood	*Viburnum rafinesquianum*
Beaked hazel	*Corylus cornuta*
Pagoda dogwood	*Cornus alternifolia*
Red osier dogwood	*Cornus sericea*
Round-leaved dogwood	*Cornus rugosa*

Table 2. Forest stands sampled for availability of bear foods in the Chippewa National Forest, Minnesota 2015 and 2016.

Stand type	2015	2016
Aspen (mature: 30+ years)	9	10
Aspen regeneration (5−15 years old)	12	10
Birch	6	10
Black ash	— [a]	8
Black spruce−tamarack	2	10
Cedar	5	8
Clearcut	— [a]	8
Lowland aspen	4	8
Lowland deciduous shrub	4	5
Maple	8	9
Pine (mature: 35+ years)	12	10
Pine plantation (8−20 years old)	6	6

[a] Not sampled

Table 3. Estimated biomass (kg/ha) for important summer and fall bear foods on the CNF, Minnesota, in key forest types where they tend to occur, comparing the 1980s to 2010s (2015 and 2016). In each decadal comparison shown, the mean for 2015 and 2016 was significantly less than that of the 1980s ($P<0.05$, Kruskal-Wallis test).

Food species	Stand type	Mean		Lowest yearly mean (year)		Highest yearly mean (year)	
		1980s	2010s	1980s	2010s	1980s	2010s
Beaked hazel	Aspen regen	16.04	1.42	0.78 (1985)	0.60 (2015)	37.01 (1988)	2.17 (2016)
	Aspen	10.11	2.87	1.31 (1985)	0.60 (2015)	20.31 (1987)	4.69 (2016)
	Pine	11.56	1.02	2.09 (1985)	0.77 (2015)	22.87 (1988)	1.27 (2016)
Round-leaf dogwood	Aspen regen	5.74	3.01	0.34 (1985)	0.36 (2015)	11.99 (1987)	5.40 (2016)
Raspberry	Aspen regen	27.95	8.85	0.00 (1989)	7.97 (2016)	83.68 (1985)	9.83 (2015)
	Pine plantation	97.55	0.02	2.78 (1989)	0.00 (2015)	224.64 (1985)	0.04 (2016)
Sarsaparilla	Aspen regen	2.45	0.01	0.15 (1985)	0.00 (2015)	4.16 (1986)	0.03 (2016)

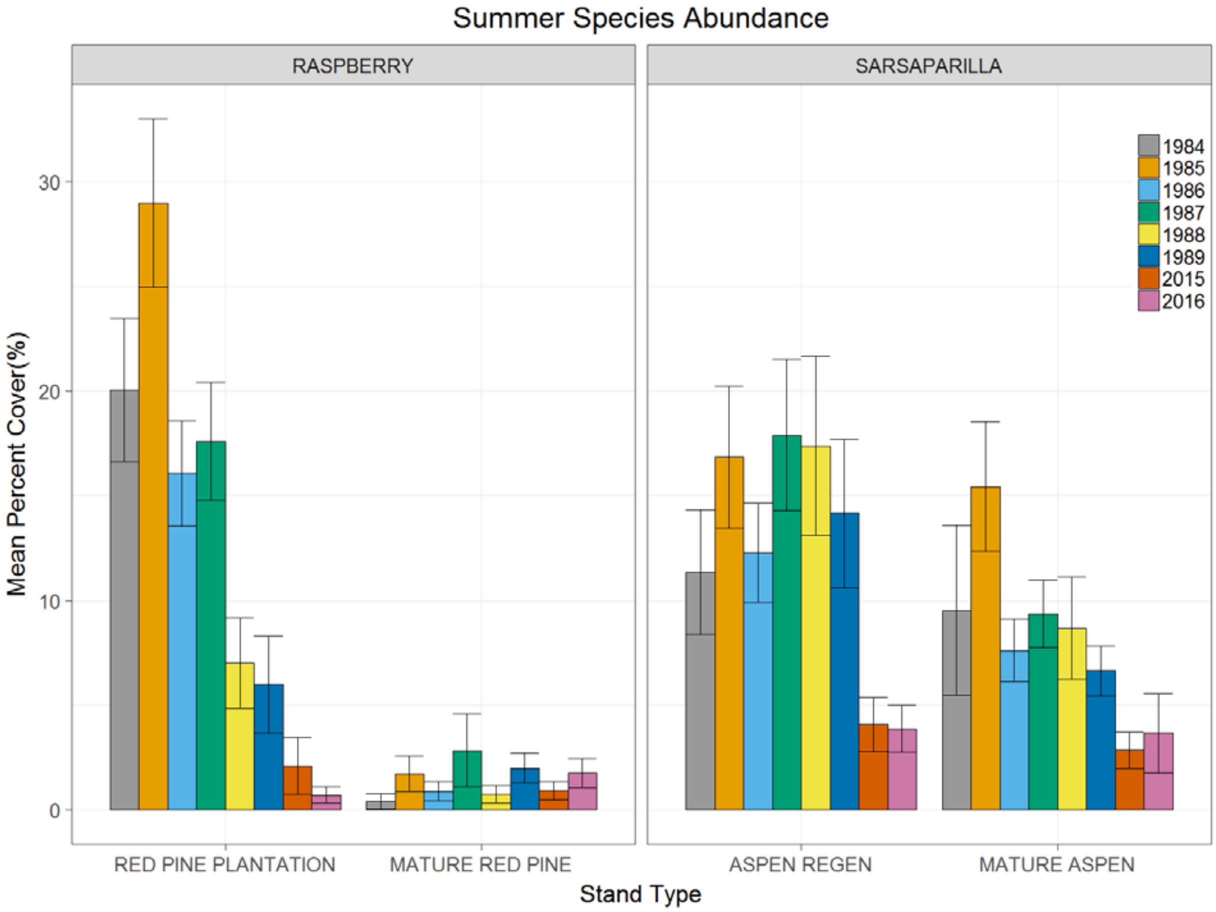

Figure 1. Yearly mean percent cover (±SE) of raspberry and sarsaparilla, important early summer bear foods, in forest stand types where they are commonly found in the CNF, Minnesota, 1984–2016.

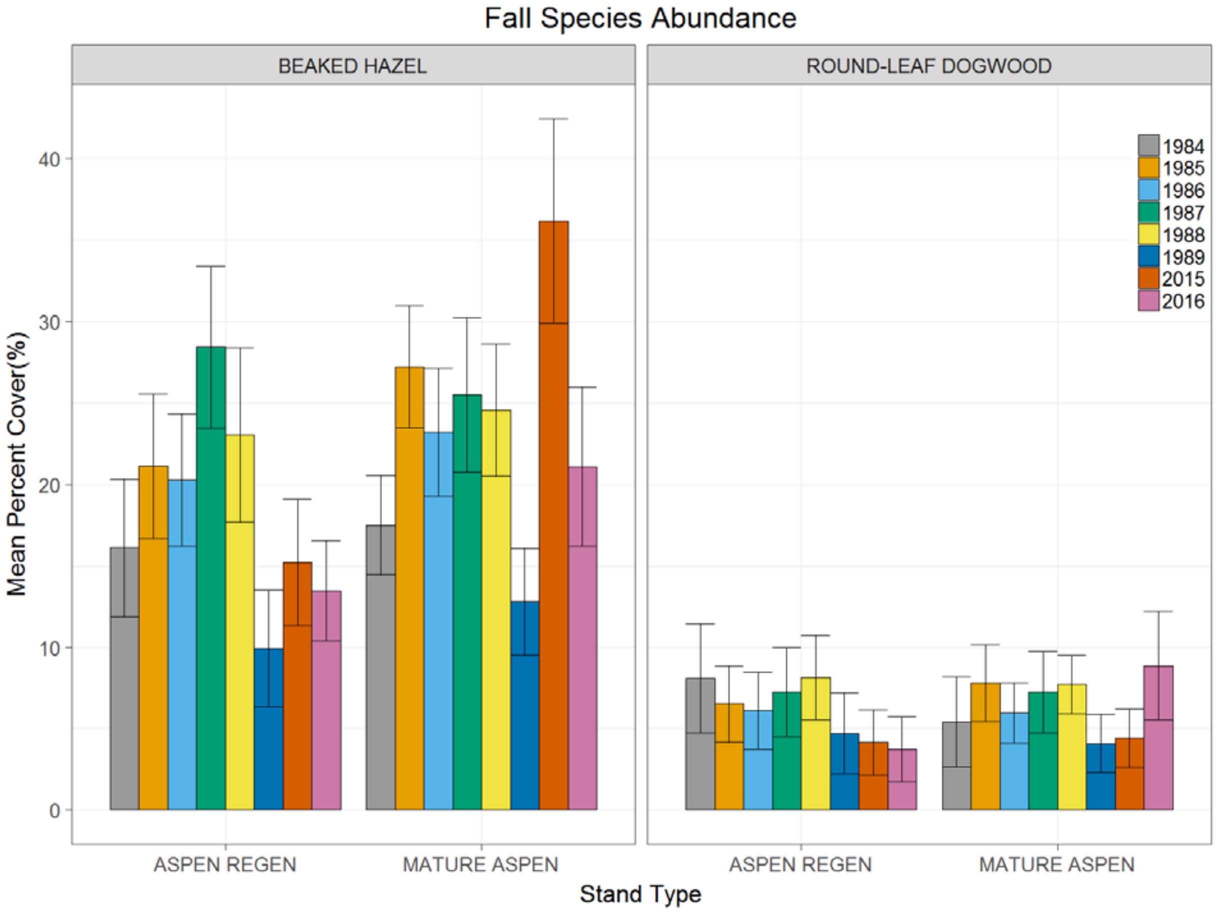

Figure 2. Yearly mean percent cover (±SE) of beaked hazel and round-leaf dogwood, important fall bear foods, in forest stand types where they are commonly found in the CNF, Minnesota 1984–2016.

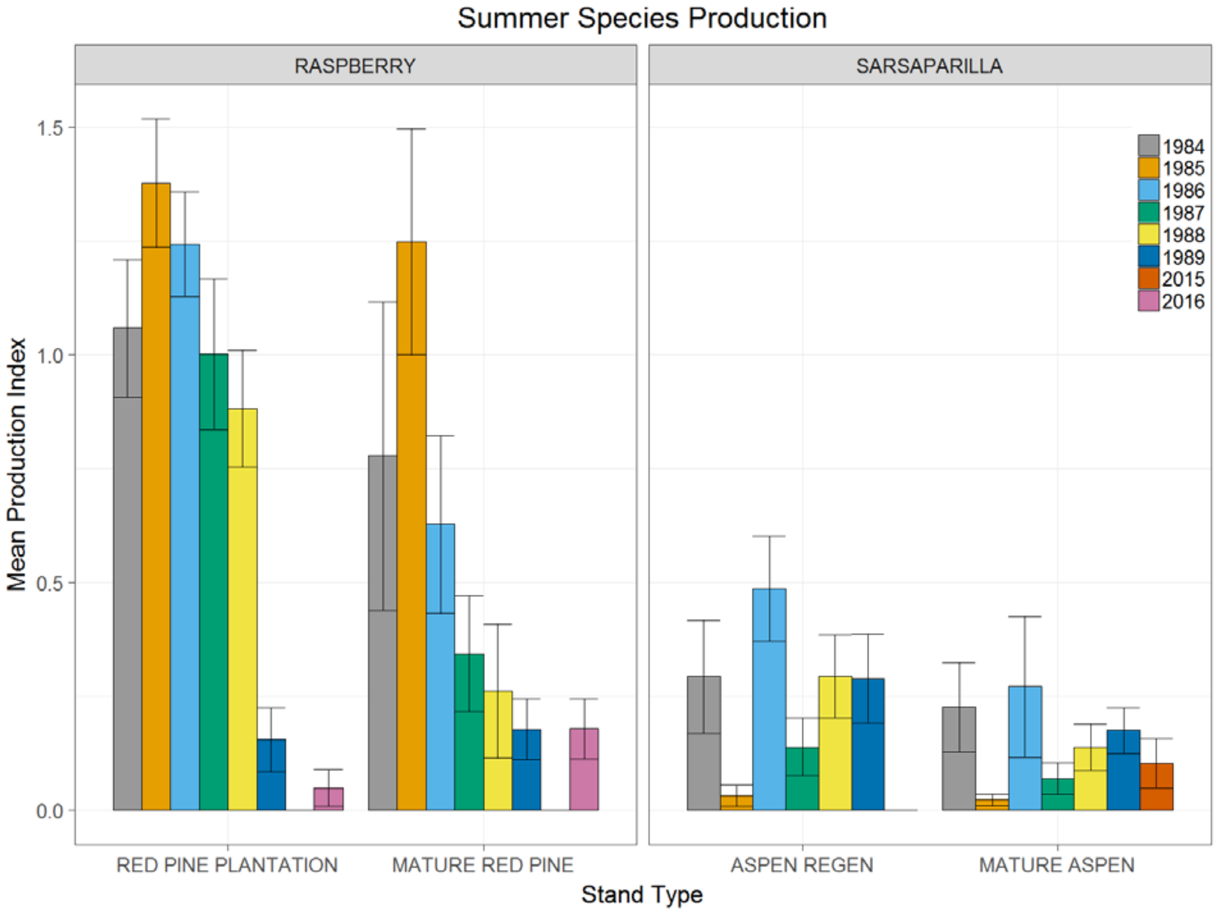

Figure 3. Yearly mean production index (0–4 scale; ±SE) of raspberry and sarsaparilla, important early summer bear foods, in forest stand types where they are commonly found in the CNF, Minnesota 1984–2016.

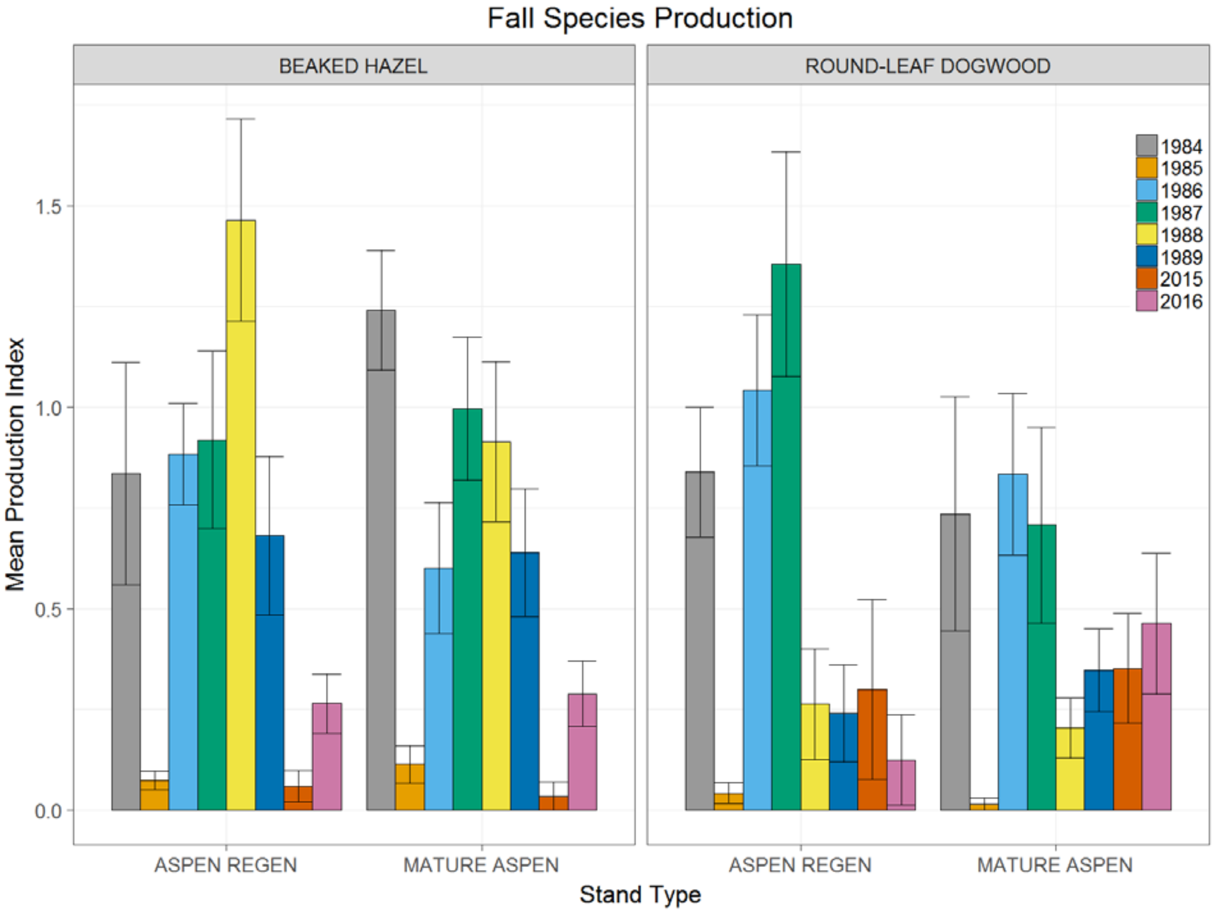

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Figure 4. Yearly mean production index (0–4 scale; ±SE) of beaked hazel and round-leaf dogwood, important fall bear foods, in forest stand types where they are commonly found in the CNF, Minnesota 1984–2016.

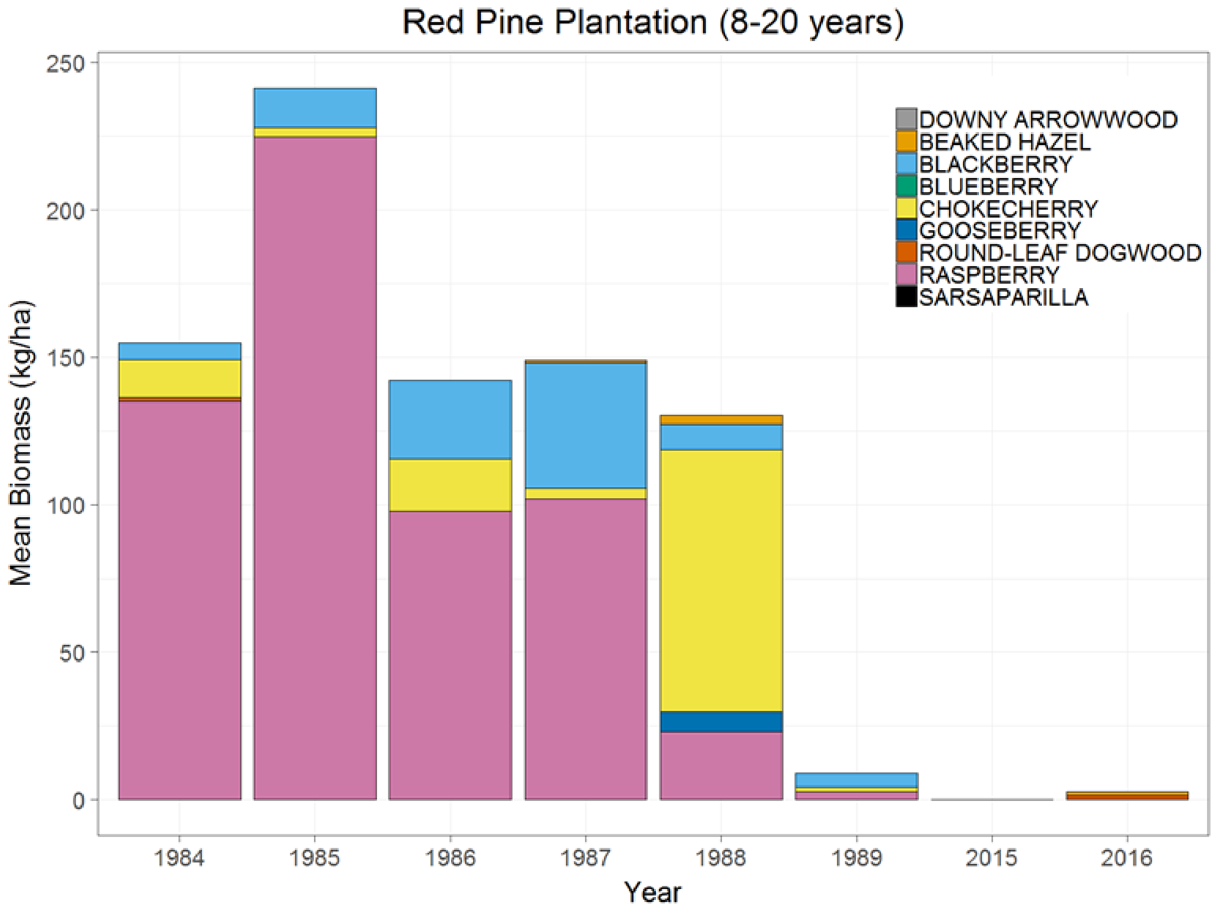

Figure 5. Yearly mean biomass (kg/ha) of bear foods in red pine plantations in the CNF, Minnesota 1984–2016.

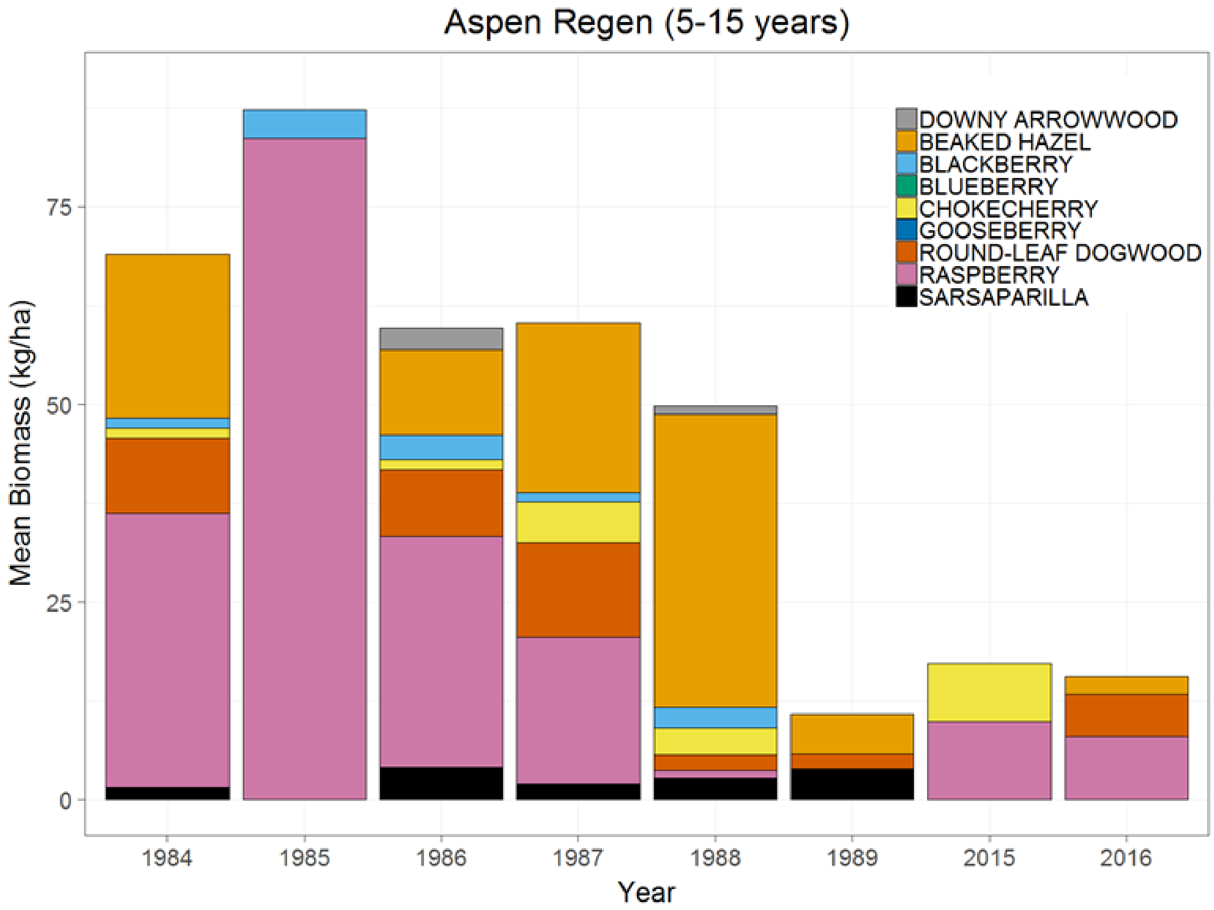

Figure 6. Yearly mean biomass (kg/ha) of bear foods in regenerating aspen stands in the CNF, Minnesota 1984–2016.

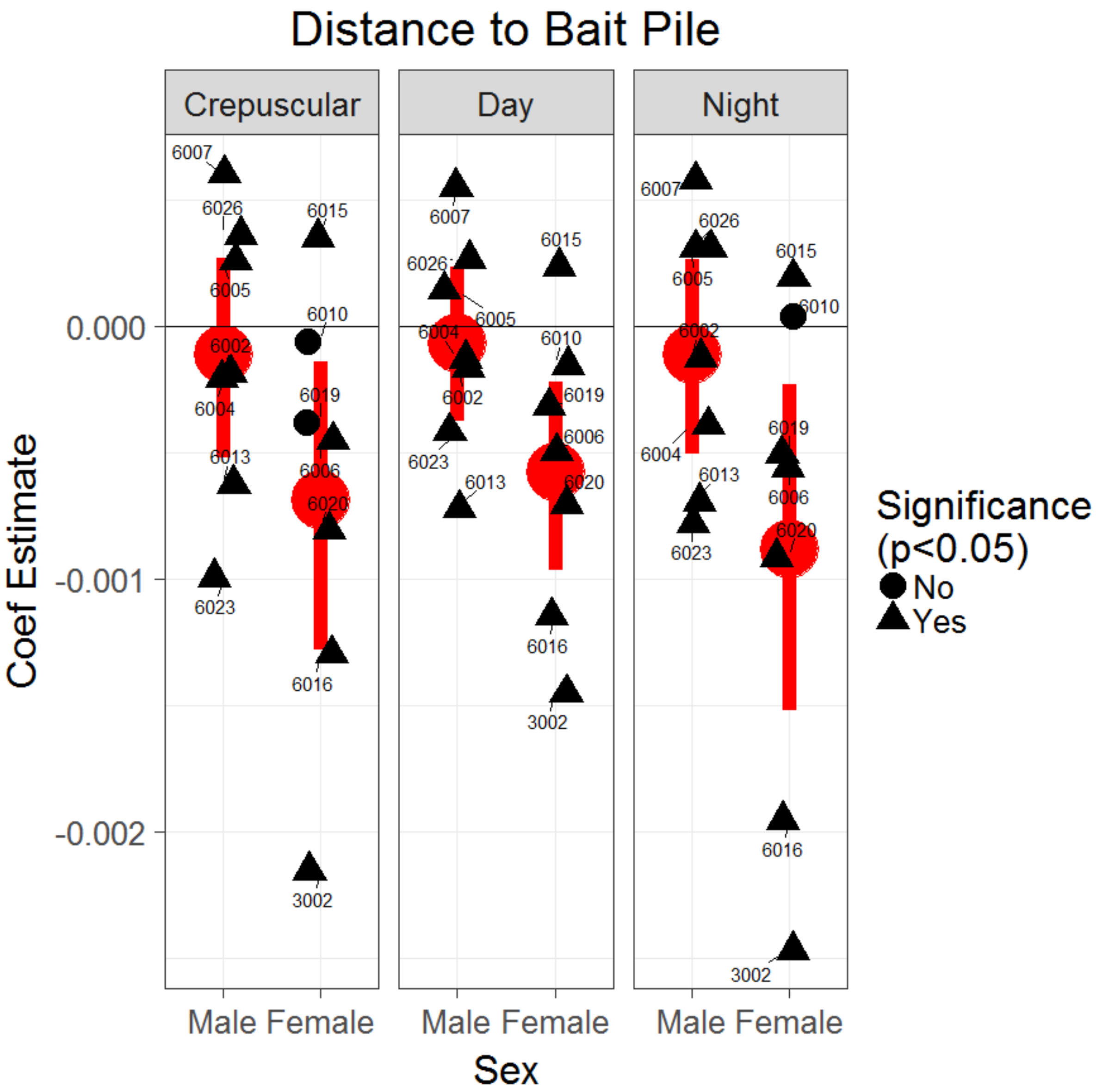

Figure 7. Resource selection function (RSF) coefficient estimates for the effect of distance to hunters' bait piles on habitat selection of black bears in the CNF, Minnesota, 12 Aug–15 Oct (the period during which hunters were allowed to maintain baits), 2016. RSFs are divided by sex and 3 periods of the day. The more negative the coefficient, the closer the bear's GPS locations were to bait piles versus random points in its home range (individual bear identification numbers shown). Four bears with positive coefficients in all 3 time periods used habitats away from baits. Red points and lines represent bootstrapped means and 95% CIs.

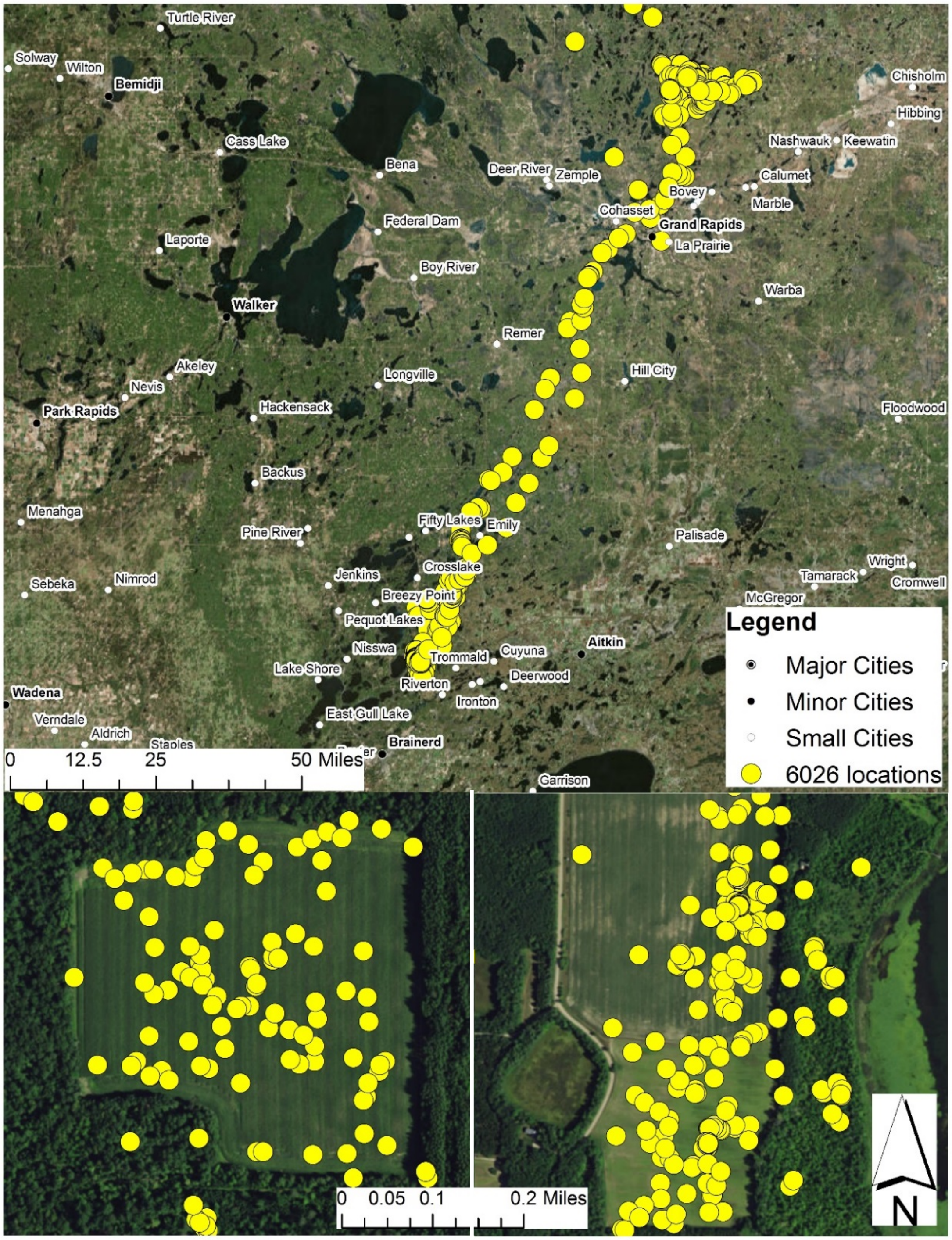

Figure 8. Long-distance migration of a GPS-collared male black bear from north of Grand Rapids to near Brainerd, Minnesota during autumn 2016. In mid-August, this bear left his summer home range and traveled south along a corridor of oak forest with poor acorn production (top panel). He spent 2 months primarily feeding in 2 corn fields <2 miles apart (bottom panels) before returning in mid-October, via the same path, to his home range to den.

EFFECTS OF HAIR-SNARE SUBSAMPLING ON SPATIALLY EXPLICIT CAPTURE–RECAPTURE POPULATION ESTIMATES OF BLACK BEARS

Nick Gondek[1], David L. Garshelis, Karen V. Noyce, and John R. Fieberg[1]

SUMMARY OF FINDINGS

Genetic mark-recapture studies estimate animal abundance using non-invasive DNA identification methods to "capture" and subsequently "recapture" individuals that leave genetic material at trap sites. Due to the cost of genotypic analysis, researchers often choose to process only a subsample of this genetic material. Traditional (non-spatial) mark-recapture estimators of abundance have been shown to be biased in this case, especially when individuals display a behavioral trap response following initial capture. Less is known about the impact of subsampling genetic mark-recapture data when using spatially explicit capture-recapture (SECR) models to estimate abundance. We are exploring the effect of subsampling on SECR estimators using hair-snare data obtained from a 2012 genetic mark-recapture study of black bears (*Ursus americanus)* from the Chippewa National Forest, north-central Minnesota. Non-proportional subsampling may be preferable to simple random sampling, despite the inherent violations of SECR assumptions that may result.

INTRODUCTION

Mark–recapture studies are routinely used by wildlife managers to estimate animal abundance. Especially in the case of endangered species and game animals, abundance and its associated temporal trends are of critical importance for making informed management decisions. Hair snares offer a minimally invasive technique for obtaining capture and recapture samples. Specifically, hair left at the trap can be genotyped to identify individuals. However, the number of samples left at barbed wire hair traps typically far exceeds the budget allotted for genetic analysis, in part because a single animal often leaves hair on multiple barbs as it passes in and out of the corral of wire. Thus, it is common practice to genotype only a subsample of hair, knowing that much of it is redundant. Subsampling has been shown to negatively bias density estimates in the context of Huggins mark-recapture models because trap-shy animals are inadequately represented (Augustine et al. 2014). Less is known regarding the impact of subsampling on spatially-explicit capture–recapture models (SECR).

A fundamental difference between SECR and non-spatial capture–recapture models relates to how the estimators make use of multiple captures of the same individual during the same time period (trapping session) at different traps. Whereas non-spatial models collapse these into a single capture event, SECR models use multiple captures within a session to inform parameters that quantify individual movement characteristics (Borchers 2012, Royle et al. 2013). Thus,

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samples that are redundant in a non-spatial model may be of critical importance to SECR models. Researchers may limit the number of genetically analyzed hair samples from a single trap within a single session because multiple visits by the same individual, hours or days apart, are impossible to distinguish from a single visit with genetic sampling alone; hence, multiple samples from the same individual at the same site-session are not informative and not worth the cost. However, if sites are visited by multiple animals, and the data from these sites are subsampled non-randomly or non-proportionally, then the spatial distribution of *processed samples* may result in a biased estimator of the spatial distribution of *visits*; hence, SECR-based density estimators may also be biased. A number of studies have examined the effects of subsampling hair samples on non-spatial capture–recapture estimates of black bears (Tredick et al. 2007, Dreher et al. 2009, Laufenberg et al. 2013), but we are not aware of any that have investigated effects on SECR-based estimates.

OBJECTIVES

1. Compare abundance and density estimates and precision obtained from the 2012 genetic capture–mark–recapture (CMR) using different subsamples of data (i.e., derive the best estimates from the existing data).
2. Provide guidance for study design of future genetic CMR estimates of bears in Minnesota.

METHODS

Data Collection

We used data from a 2012 genetic mark-recapture study of American black bears (*Ursus americanus*) mainly within the Chippewa National Forest, northern Minnesota (Noyce and Garshelis 2013). We obtained bear hair samples from 121 stationary traps, spaced within grid cells of 1 mi^2 and checked 6 times at 10-day intervals (trapping sessions) during May–July. Traps were constructed of 2 strands of barbed wire encircling a suspended bait of bacon and scent lures. We collected bear hair from 2,784 barbs that occurred in 1,642 separate clusters of 1–11 adjacent barbs (considering both upper and lower strands of wire). We considered each cluster (not the individual barbs) a sampling unit, meaning that at most, we sampled only 1 barb from each cluster (although a single barb with hair, not adjacent to any other barb with hair, was also considered a cluster). Of these 1,642 clusters, 1,113 were sent to a genetics laboratory for genotypic analysis, and 1,019 samples were successfully linked to specific individuals.

Spatially-Explicit Capture–Recapture Estimators

Detection probabilities in spatial mark-recapture models are assumed to decrease as a function of distance between each trap and an individual's activity center, a latent variable in the model. There are a number of detection functions that can be used to model detection probabilities. We fit a half-normal detection function (the default), which in its most simple form has 2 parameters: g0, which determines the detection probability at the activity center, and σ, which controls how quickly detection probabilities decrease with distance from the activity center. Intuitively, σ will depend on how much individuals move and will thus be related to home range size.

Either of these parameters can, in turn, be modeled as a function of covariates (e.g., sex) or time (sampling session). There are 2 options for how parameters, and thus detection probabilities, may depend on time–parameters can vary linearly (on a transformed scale), denoted by T, or they may vary in an unstructured way, allowing each sampling session to vary independently, denoted by t. In addition, models can allow for a "behavioral effect," whereby parameters for recapture probabilities differ from those for initial capture probabilities. Models that allow parameters to change following an initial capture are denoted using the following syntax: b indicates a behavioral effect that applies to future capture probabilities at any site, and b_k indicates the behavioral effect only applies to future capture probabilities at the site (*k*) where the animal was previously captured.

We fit 4 SECR models to the full data set, each with a different combination of explanatory variables: g0 ~ 1 (i.e., capture probabilities only depend on the distance between a bear's activity center and the trap location), g0 ~ t, g0 ~ b_k, and g0 ~ b_k + t. For all models, we also assumed that σ varied by sex. Models were fit using the Program R (R Core Team 2015), package 'secr' for fitting SECR models and packages 'foreach' and 'doParallel' for optimization of model fitting using parallel processing (Revolution Analytics and Weston 2015a,b; Efford 2017).

Subsampling Methods

We are exploring 2 subsampling strategies: simple random sampling (SRS) and a subsampling method that gives preference to unique site-sessions, which we refer to as site-session preferred (SSP). With SRS, *n* samples were chosen at random from the set of hair clusters pooled across the different sites and trapping sessions. Alternatively, with SSP, we tried to maximize the number of unique site-sessions represented in the subsample. Let *m* represent the number of unique site-sessions with hair in the full dataset. If $m \leq n$, we randomly choose *n* unique site-sessions, with 1 sample randomly selected from each of these site-sessions (in the survey sampling literature, this is referred to as a 2-stage cluster sample). When $n > m$, we chose 1 sample at random from each unique site-session and then took a second simple random subsample of size $n – m$ from the remaining clusters (from the pooled data) to give a total of *n* samples. This approach attempts to serve as a compromise between minimizing the amount of redundant data (accomplished by sampling an equal, or near equal, number of observations from each unique site-session) and maximizing the representativeness of the sample (as accomplished by SRS).

We considered 3 subsample sizes, *n* = 250, 550, and 850. For each subsample, we determined the number of unique combinations of (individual x site x session). We subtracted this number from *n* to determine the amount of redundant data in the subsample. We are in the process of developing R code that will allow us to fit the same 4 SECR models to each subsampled data set. This will allow us to compare density estimates from the full dataset to those obtained with various levels of subsampling.

RESULTS AND DISCUSSION

SECR Models Fit to Full Dataset

The best-fitting SECR model for the full hair-snare data set included a trap-specific behavior response and an unstructured time covariate, g0 ~ b_k + t. Estimates of recapture probabilities were substantially greater than original capture probabilities (Figure 1). Capture and recapture probabilities also varied considerably among the different sampling sessions and decreased more quickly with distance for females than for males (Figure 1). Using this model we estimated there were 12.4 bears per 100 mi^2 (95% CI = 9.06–16.87, Table 1).

Effects of Subsampling on Redundancy

We collected data from m = 377 unique site-sessions with hair. As the SSP subsampling strategy initially collects 1 sample from each unique site-session, subsamples with $n \leq 377$ will not include any redundant data. The same is not true for SRS, which may include multiple samples from the same individual at the same trap, particularly for site-sessions with many clusters of hair. We found that, at low sample sizes, SSP selected far fewer redundant samples than SRS, and that this advantage diminished as sample size increased (Figure 2).

Future Work

Our simulations suggest that using a subsampling strategy that maximizes the number of unique site-sessions can reduce the likelihood of analyzing redundant samples, but more work is required to determine whether this benefit outweighs the inherent loss of movement information incurred using this method. In either case, the optimal strategy likely depends on the characteristics of the observed study population with respect to the spacing of the traps. Further simulation is needed before we can make general conclusions and recommendations regarding the effect of subsampling methodology on SECR estimates. In the future, we plan to fit SECR models to the subsampled datasets. In addition, we plan to simulate genetic mark-recapture data with varying degrees of behavioral responses to evaluate subsampling methods across a range of scenarios where the true population size is known.

ACKNOWLEDGMENTS

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Table 1. Density estimates and Aikaike Information Criterion (AICc) scores associated with spatially-explicit capture–recapture models fit to data from 1,019 genetically-identified hair samples from black bears that visited hair-snares in Minnesota, May–July, 2012. ΔAICc represents the difference from the lowest scoring model and the compared model. All models were fit using Program R and package secr. For capture probabilities, notation 'b_k' represents trap-specific behavior,'*t*' represents a non-linear time effect, and 1 indicates an intercept-only model (i.e., detection depends only on distance from the animal's activity center to the trap). All 4 models assumed the scale parameter varied by sex, σ ~ sex.

Model	AICc	ΔAICc	Bears/100 mi^2 (95% CI)
$g0 \sim b_k + t$	3082	0	12.36 (9.06–16.87)
$g0 \sim b_k$	3129	47	12.31 (9.02–16.80)
$g0 \sim t$	3507	425	12.54 (9.22–17.08)
$g0 \sim 1$	3570	488	12.54 (9.21–17.07)

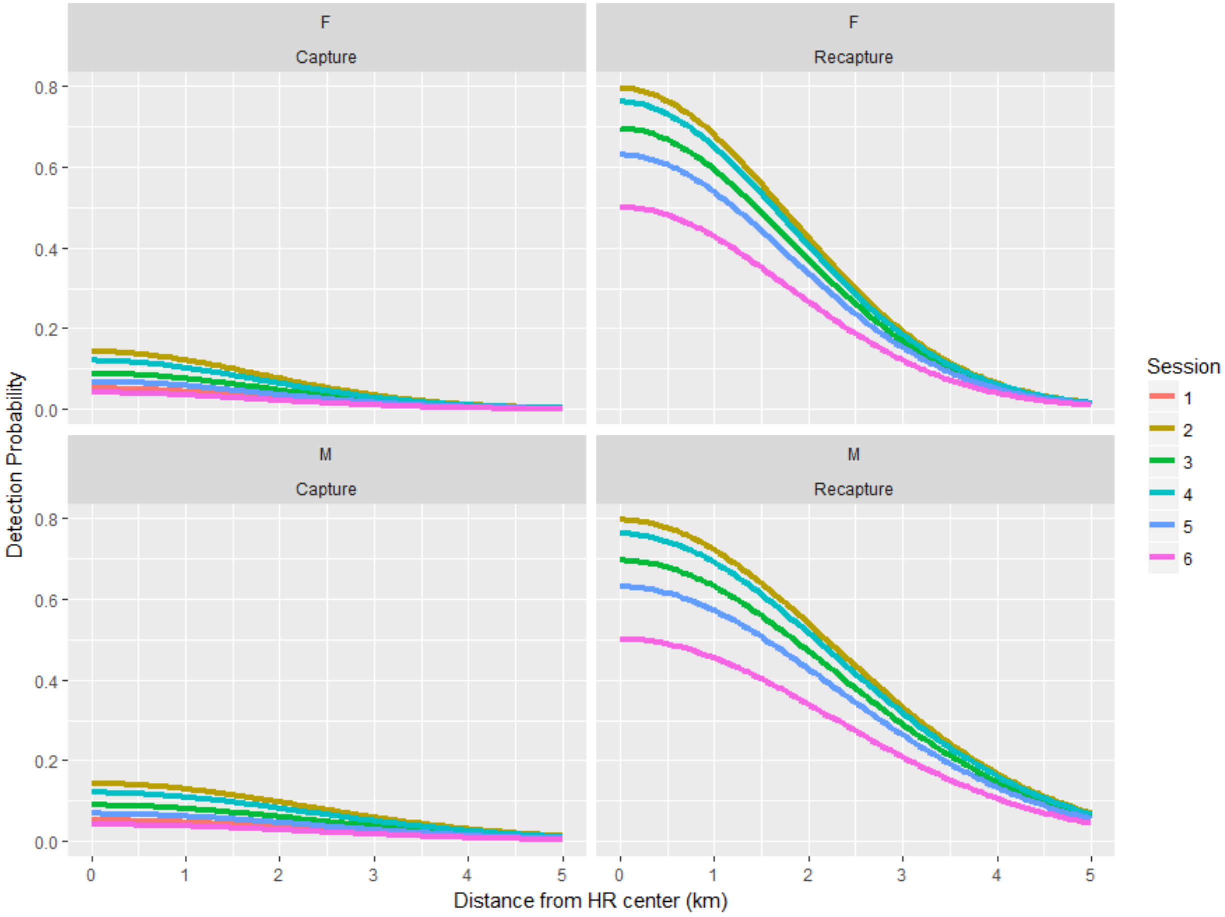

Figure 1. Estimated capture and recapture probabilities for male (M) and female (F) bears modeled as a function of the distance between a bear's estimated home range (HR) center (based on spatial distribution of recapture data) and a given trap. Estimates were obtained using model g0 ~ b_k + t, σ ~ sex. In all graphs, the 6 lines represent, from top to bottom, periods 2, 4, 3, 5, 1 (no recaptures for 1st period), and 6. Data are from a bear hair-snaring study in the Chippewa National Forest, Minnesota, in 2012.

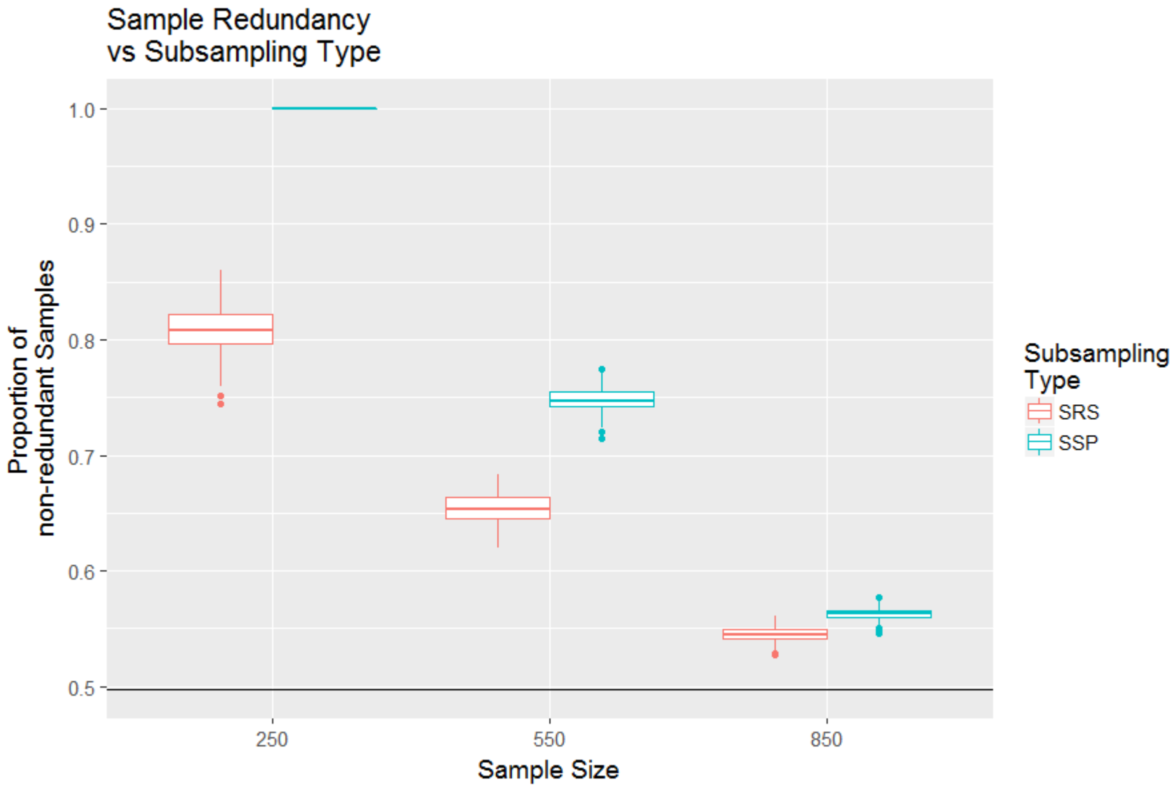

Figure 2. Proportion of non-redundant samples (i.e., different site-sessions) versus sample size when using simple random sampling (SRS) or site-session preferred sampling (SSP). Horizontal line near 0.5 represents the proportion of non-redundant samples in the full data set of genetically analyzed samples (which itself was already a subsample of 1,642 clusters of barbs with hair). When sample size is smaller than number of traps on the trapping grid, all samples chosen using SPP will, by definition, be non-redundant. As *n* increases, the difference in sample redundancy between SRS and SPP diminishes, converging to 0 when the full data set is utilized (i.e., when *n* = 1019). Data are from a bear hair-snaring study in the Chippewa National Forest, Minnesota, in 2012.

EVALUATION OF DESIGN AND ANALYSIS OF A CAMERA-BASED MULTI-SPECIES OCCUPANCY SURVEY OF CARNIVORES IN MINNESOTA

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SUMMARY OF FINDINGS

Camera-based surveys are increasingly being used to monitor wildlife species across large areas and a diverse range of habitats. We initiated a study in a forested area of northern Minnesota to assess various design and analysis questions related to use of remotely-triggered cameras for simultaneously monitoring the occurrence of multiple species of carnivores. In spring and fall 2016, we deployed 100 cameras in an area equivalent to 20 townships, with 5 cameras placed in each 9.65- x 9.65-km township. To test different lures and strategies for camera placement, we conducted a 2 x 2 factorial experiment following a randomized complete block design: four cameras were placed at randomly selected locations within forested areas, and were assigned one of 2 lures (salmon oil or a liquid version of the fatty-acid scent used in tablet-form on the Minnesota Department of Natural Resources (MNDNR) scent-station survey) and one of 2 different placement strategies (on the closest suitable tree within 5 m from the randomly selected point, or at a user-chosen location within 90 m of the randomly selected point). We deployed an additional camera, without a lure, on a secondary road or trail within a forested area of each township. All cameras were active for a minimum of 6 weeks, and we recorded ~680,000 photos in the spring and ~370,000 in the fall. Among carnivores, black bears (*Ursus americanus*) and bobcats (*Lynx rufus*) were detected at a greater number of sites in spring than in fall, whereas coyotes (*Canis latrans*), red (*Vulpes vulpes*) and gray (*Urocyon cinereoargenteus*) foxes, martens (*Martes americana*), fishers (*Pekania pennanti*), raccoons (*Procyon lotor*), and striped skunks (*Mephitis mephitis*) were detected more frequently in fall. Gray wolves (*C. lupus*) were detected at a similar number of sites in both seasons, whereas badgers (*Taxidea taxus*) and weasels (*Mustela spp.*) were detected only in the fall and at few sites. We also frequently detected several non-carnivore species, including white-tailed deer (*Odocoileus virginianus*), red squirrels (*Tamiasciurus hudsonicus*), snowshoe hares (*Lepus americanus*), and, more rarely, porcupines (*Erethizon dorsatum)* and moose (*Alces alces*). More detailed analysis of the data is pending.

INTRODUCTION

Monitoring programs designed to track the distribution and actual or relative abundance of carnivores can be important for determining population status and for quantifying the effects of harvest, habitat change, and environmental variability on populations. The Minnesota Department of Natural Resources (MNDNR) currently relies on 2 track-based surveys (scent station and snow-track surveys) to monitor trends in a suite of 14 carnivores/furbearers. The data from these surveys have provided rough estimates of trend for many species, although interpretation must always be qualified with acknowledgement of 2 key, but untested, assumptions, namely that detection rates do not exhibit significant temporal or spatial trends

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and that road-based surveys adequately represent population-wide trends. Logistical challenges with conducting these surveys have also increased in the last decade due to loss of survey collaborators from other natural resource agencies, increased traffic or paving/plowing of roads, and less reliable snow in early winter. In the past decade, several key carnivore species had declined (e.g., fishers, martens, bears) and management intensity had increased on wolves. Given the importance of monitoring these species, statistical uncertainties with existing surveys, and increasing logistical challenges, we felt it was an opportune time to consider alternative ways to monitor carnivore populations. Camera surveys are an attractive option because they provide a means to estimate detection rates with little if any additional field effort, are less dependent on specific environmental conditions, and are more amenable to use of ‘citizen scientists’ with little formal training (photos can be verified by trained staff). Thus, remote cameras are increasingly being used or considered for large-scale multi-species occupancy surveys (e.g., O’Brien et al. 2010, Pettorelli et al. 2010, Ahumada et al. 2011, Kays et al. 2011, Fisher and Burton 2012).

Camera-based surveys are not new to wildlife monitoring (Kays and Slauson 2008, Kucera and Barrett 2011), but the simultaneous development of improved remotely-triggered cameras, rigorous analytical methods, and reduced costs have bolstered their applied value. As evidenced by their use in monitoring a wide array of carnivores in different landscapes (e.g., see Table 5.1 in Kays and Slauson 2008), cameras are a non-invasive tool well-suited to detect species that may be difficult to trap and handle, occur at low densities, or have nocturnal and secretive habits.

Occupancy models (sensu MacKenzie et al. 2002, MacKenzie et al. 2006) are commonly used in wildlife monitoring programs, often in conjunction with camera traps, due to their flexibility, sound statistical framework, and close connection to population estimation. Taking advantage of repeated sampling (in space or time), occupancy models can provide unbiased estimates of occupancy probabilities that adjust for imperfect detection (i.e., failure to detect a species when it is present in a certain area). Failing to account for imperfect detection can lead to misleading estimates of spatial and temporal trends in occurrence (Guillera-Arroita et al. 2014a), and as a result, poor management and conservation decisions. While there are several important assumptions that must be met to apply occupancy models, the approach is not dependent on a specific tool or method to detect animals.

General survey design guidance for occupancy surveys is available (e.g., MacKenzie and Royle 2005, MacKenzie et al. 2006, Bailey et al. 2007, Guillera-Arroita and Lahoz-Monfort 2012, Guillera-Arroita et al. 2014b), but ideally study designs should be tailored to features of the target species and study area to avoid violation of model assumptions (e.g., independent detections and constant occupancy status), which can lead to biased estimators of detection and occupancy rates or require complex modelling approaches for sound statistical inference. Not surprisingly, occupancy modelling is an emerging and fast-moving field, and we expect new methods to be developed and guidance on their use to continually evolve in the coming years (Rota et al, 2016; Broms et al, 2016; Tobler et al, 2015; Ovaskainen et al, 2016).

Implementing a camera-based occupancy survey requires consideration of a variety of design and analysis options. While we do not delve into the details of each here, we highlight the following considerations: 1) camera selection and settings (Swann et al. 2004, Kays and Slauson 2008, Damm et al. 2010, Swann et al. 2011, Meek et al. 2012, Rovero et al. 2013, Weingarth et al. 2013, Wellington et al. 2013); 2) camera positioning; 3) whether to use baits/lures, and if so, which ones (Kays and Slauson 2008, Schlexer 2008, Du Preez et al. 2014); 4) time of year, which can affect species’ behaviour and ‘availability’ as well as likelihood of meeting methodological assumptions (e.g., Kendall and White 2009, Rota et al. 2009); 5) number of cameras; 6) camera spacing and consideration of spatial correlation among sites (e.g., Sargeant et al. 2005, Hines et al. 2010, Magoun et al. 2010, Aing et al. 2011, Guillera-Arroita et al. 2011, Dorazio and Rodriguez 2012, Johnson et al. 2013); 7) whether or how best to discretize (e.g., hours, days, weeks) the temporally-continuous data from cameras into multiple survey occasions (e.g., Guillera-Arroita et al. 2011, Bischof et al. 2014); 8) site selection (e.g., random, systematic, convenience) and whether to allow flexibility in micro-site selection; and 9) approach to data analysis (e.g., single-species versus hierarchical community models; Dorazio and Royle 2005, Dorazio et al. 2006, Kery and Royle 2008, Zipkin et al. 2009, 2010, 2012, Giovanini et al. 2013, Pacifici et al. 2014).

Optimizing survey design becomes more complicated when multiple species with varying abundance and detection rates are involved. Biological characteristics of the species, such as home range size, movement patterns, and habitat preferences show large variation among carnivores (Boitani and Powell 2012). Consequently, a sampling design optimal for one species can violate important model assumptions for another. In the case of MNDNR surveys, where the suite of target species ranges from small to medium-sized mammals, such as skunks and martens, to large, roaming species like wolves and bears, design and analysis options that best account for or address this variability will be preferred. Recent attention has been given to design of camera-based occupancy surveys targeting a community of carnivores (Hamel et al. 2013, Shannon et al. 2014), but their conclusions may not extend beyond the specifics of the biological system and analysis approaches considered therein.

OBJECTIVES

The broad objectives of this project are to:

1. Compare effects of various survey design and analysis options on the magnitude and precision of estimates of detection and occupancy rate for multiple species.
2. Assess possible logistical constraints on implementing a large-scale multi-species camera survey in Minnesota; and
3. Compare the efficacy of camera surveys to the track surveys currently being used for monitoring carnivores in Minnesota.

As noted above, there is a large array of design and analysis questions to consider when conducting a multi-species occupancy survey with cameras. Hence, we decided to use an adaptive approach to survey design, focusing year 1 efforts on 4 specific design questions: 1) timing (spring versus fall survey; survey duration); 2) lure options (salmon oil versus fatty acid scent oil); 3) site selection (cameras on trails versus randomly selected sites); and 4) strategies for camera deployment (enhanced placement versus not enhanced). Our approach to analysis will also consider the effects of using daily versus weekly survey intervals and single- versus multi-species occupancy models. Additional comparisons and analysis will be undertaken next year after results of the first analyses are completed.

STUDY AREA

In spring and fall 2016, we implemented the first camera survey in one study area located in Itasca County, north-eastern Minnesota (Figure 1). This 1872 km^2 (48 x 39 km) area is mainly covered by forests and lakes and includes a high percentage of public land, including a portion of the Chippewa National Forest (SW portion of the study area), George Washington State Forest (NE portion), Scenic State Park (NC portion) and other state and county lands interspersed throughout.

METHODS

Based on our minimum camera specifications [i.e., passive infrared (PIR) cameras with intermediate to fast trigger (<0.7 s) and recovery (<1.7 s) speeds, multi-picture capability (minimum 3) per trigger event, “no-glow” (black LED) infrared flash, and of moderate cost (maximum $200 per camera)] and a competitive bid process, the camera model we deployed was the Bushnell Trophy Cam HD Aggressor No-Glow.

Survey Timing and Duration

We considered 4 objectives in selecting the timing of our camera surveys: 1) maximize the species richness of carnivores that would be ‘available’ for detection; 2) minimize the likelihood of violating the occupancy model assumption of species’ closure during the survey; 3) minimize logistic challenges with deploying cameras; and 4) maximize ‘biological relevancy’ and consistency with timing of existing surveys and annual management decisions. Although our experience has been that winter is a good time to conduct lure-based camera surveys for many carnivores, we concluded that several species would be undetectable (e.g., bears, skunks), ongoing harvest seasons for many species would increase risk of violating closure assumptions, and deep snow could pose logistic challenges. Although summer was a potential option, we believed that more rapid desiccation of lures and rapidly changing ‘availability’ of maturing offspring made it a less desirable option than spring and fall surveys. Hence, we chose to compare camera-based surveys conducted in the spring and fall, presumably reflecting spring ‘pre-breeding’ and fall ‘pre-harvest’ populations.

Our previous experience had been that few additional species are detected after 3–4 weeks of camera deployment. Although cameras can be left out indefinitely with only minimal additional financial cost related to personnel to review photos, long surveys increase risk of violating closure assumptions through mortality, immigration, or emigration. Hence, we chose to deploy cameras for 6 weeks during the first year, specifically May 1 to June 15 and September 1 to October 15.

Lure Selection

We concluded that use of a bait or lure was likely necessary to produce sufficient detection probability for many carnivore species, especially if cameras are to be deployed using a more desirable probabilistic sampling scheme. Similar to conclusions by Fisher and Burton (2012), we believed that olfactory lures will be preferred over baits and that all species of interest in this study can likely be attracted, albeit to varying degrees, with a more logistically-practical olfactory lure.

We decided to test 2 lures the first year, limiting our consideration to attractants that were likely to be not only effective for a suite of carnivore species, but also ones that could be reasonably standardized and were expected to be commercially available into the foreseeable future, easily applied, resistant to variable weather conditions, and could be purchased and distributed without significant secondary processing. There was a vast array of potential lures to consider. Based on our goals, personal experience, examination of the literature (e.g., Schlexer 2008), and consultation with a trapping lure manufacturer, we chose to compare commercial salmon oil with a liquid version of the synthetic fatty acid scent (FAS) that has been used (in tablet form) on a long-term multi-species track survey in Minnesota (Erb 2015). Details of the lure placement protocol are discussed below; here we simply note that at each site selected for salmon oil, we deployed 473 ml (16 oz), whereas for sites selected for FAS oil, we deployed a 237-ml (8 oz) bottle that consisted of 80% mineral oil and 20% liquid FAS.

Macro-Site Selection

In the first year, our focus was on evaluating the spatial sampling design in forested habitats. To identify suitable locations for camera deployment, we used Light Detection and Ranging (LiDAR) data (e.g., see Merrick et al. 2013) collected by the State of Minnesota in 2011 (http://www.mngeo.state.mn.us/chouse/elevation/lidar.html) to identify pixels (~ 20 X 20 m) with mean tree height >3 m (10 ft) and canopy cover >50% (Figure 2; details of this process will be incorporated in future reports). We then divided the study area into 20 contiguous blocks the size of townships (9.65 x 9.65 km). To ensure a minimum distance of 1.6 km (1 mi) between cameras both within and across blocks, we constrained the randomly selected points to lie within 4 equally-spaced sub-quadrats within each block (Figure 2). We then intersected the suitable locations (pixels) identified via LIDAR with the sub-quadrats and used the *Generate Random Points* tool in ArcGIS to select one random point falling within each of the 4 sub-quadrats in each block (Figure 2).

In addition, we deployed an un-lured camera placed on a secondary trail closest to the center of each township (hereafter, *trail camera*), provided the site was at least 400 m (0.25 mi) from all primary roads and at least 1.6 km (1 mi) from other cameras (Figure 2). We loosely defined secondary roads or trails as those that did not receive year-around maintenance and were accessed primarily on foot or with off-road vehicles. Our primary intent in deploying un-lured cameras along trails was to assess whether this type of convenience sampling was more likely to detect larger carnivores, such as wolves, that often use these trails and may be more wary of lured sites.

After selecting all locations and before deploying the cameras, each site was visualized on 2015 aerial photos to help ensure all requirements for deployment were likely met, including an additional requirement that each site was a minimum of 30 m (100 ft) from any non-forested edge.

Micro-Site Selection and Covariates

Another important decision, after selecting the camera macro-sites, was how much flexibility should be allowed in determining the exact placement of the camera. Although the use of lures effectively expands the area of camera 'coverage' well beyond the actual camera, within a given forest patch one can still potentially locate a microsite where the probability of carnivore use or detection will be higher. However, allowing flexibility in micro-site selection could introduce a source of heterogeneity in detection probabilities that may be difficult to quantify objectively. Using experienced biologists, we decided to test whether expert-based choices in fact increase detection rates. We accomplished this by dividing lured cameras into 2 camera placement strategies: 1) *not enhanced*, meaning the camera was placed on a tree within a 5-m (15-ft) radius from the randomly selected point; or 2) *enhanced*, meaning the operator actively looked for an optimal deployment location within a 90-m (300-ft) radius of the randomly selected point.

At all camera stations, we recorded several vegetation characteristics (tree species diameter and dominance, shrub cover, canopy cover) and presence of game trails, natural 'bottlenecks', and other features within approximately 15 m of the final deployment location that could increase probability of detecting a carnivore. We also took a digital photo of angular (45°) canopy cover in 4 directions around the base of the camera tree, parallel and perpendicular to the camera-lure axis. While walking to each camera site (usually < 3 km), we also recorded presence of indirect carnivore sign (tracks, scats, dens). For trail cameras, we recorded trail width, ease of access (e.g., walk, ATV, vehicle), an initial index of frequency of use by humans (which we will corroborate based on human-detections by the cameras), and vegetative coverage and height on the trail surface. Other variables (e.g., distance to main roads or water, landscape configuration metrics) will be measured using GIS. Although trail cameras were not designated an enhanced versus not enhanced treatment, we allowed flexibility in final deployment location of these cameras due to the need to position the camera on a tree at the desired angle and within sufficient distance of the trail to ensure trigger activation by animals; from the original coordinate, users were allowed a distance of 45 m (150 ft) in either direction down the trail to place the camera.

Experimental Design

To test different lures and placement strategies, we conducted a 2 x 2 factorial experiment following a randomized complete block design. Along with the trail camera, 4 lured cameras were placed within each block at sites selected using the processes described above in the macro- and micro-site selection sections. Cameras at each randomly chosen site were randomly assigned 1 of 2 lure types (salmon oil or fatty acid scent oil) and 1 of 2 camera placement strategies (not enhanced or enhanced, Figure 3).

Camera Deployment and Settings

In each camera session we deployed 100 passive infrared Bushnell Trophy Cam HD Aggressor No-Glow cameras, 80 at lured sites and 20 at un-lured trail sites. The general settings for all the cameras were based on pre-deployment testing. All cameras were attached to sturdy trees with bungee straps and placed about 75 cm (30 in) above the ground. The detection area in front of the cameras was cleared of vegetation (ferns, branches, leaves) that could obstruct the viewing area or cause false triggers, especially on windy days. At lured sites, we poured the lure on a tree located 4.5 to 9 m (15 to 30 ft) from the camera tree, with a preferred distance of 6 to 7.5 m (20 to 25 ft). We aimed trail cameras at a 45° angle to the main axis of the trail to ensure more opportunity to capture images of faster moving animals. We also aimed all cameras north (ranging from northeast to northwest) when possible to reduce false triggers and blurred photos from direct sunlight.

All the cameras were programmed to record 3 mega-pixel images (color during daylight and black/white during night), with 3 'rapid-fire' pictures per trigger event and a 2-second delay between subsequent triggers. Additionally, a set of 3 rapid-fire time-lapse pictures were taken twice a day (noon and midnight) to check the functioning of the cameras and to record regular measures of daily temperature at each site. Date, time, temperature and camera Id were printed on all the images and recorded in the image metadata.

Photo Processing and Analysis

Identification of species is done using experienced personnel following the protocol described in Niedballa et al. 2016, using the *camtrapR* package (Niedballa et al. 2017) in Program R (R Core Team 2015). We will use these data to compare detection rates for the 2 lures and the 3 camera placement strategies. In addition, we will calculate cumulative species richness curves to address questions related to survey duration and timing. Lastly, we will model occurrence and detection probabilities as functions of landscape features (e.g. bottlenecks, game trails) and forest characteristics (e.g. forest type, shrub cover) to provide information on species distribution and detectability. Further details of analysis methods will be presented in future reports.

RESULTS AND DISCUSSION

Camera Function

During the first year of sampling, cameras recorded ~680,000 pictures in the spring and ~370,000 in the fall. In the spring, 75 of the 100 cameras deployed remained operational for the full session (Figure 4); one was missing (site was logged), 4 malfunctioned, and bears altered camera positioning on approximately 20 cameras, though only 9 of these were moved to an extent that the lure tree was no longer visible. Insolation paired with lack of canopy cover during the first weeks of the spring survey and growing vegetation (especially ferns) in the later weeks resulted in a large number of false triggers and, in some cases, cameras that were no longer operable (e.g., when growing vegetation filled the detection area). In the fall, 93 of the 100 cameras remained operational (Figure 4); canopy cover appeared to reduce false triggering, all ground vegetation had sprouted and could be cut, and we added a second strap to secure the cameras and minimize bear disturbance to cameras. Bears were still the main reason for cameras becoming inoperable in the fall (5 out of 7), and the reduced number of bear-related problems could be due to a decrease in the number of bear visits in the fall.

Species Detections

Coyotes, red and grey foxes, raccoons, striped skunks, martens and fishers were detected at ≥ 2 times as many sites during the fall compared to spring (Figure 5). Conversely, bears were detected at >4 times the number of sites in spring compared to fall, and bobcats were detected at 42% more sites in the spring. Badgers and weasels were detected only in the fall, at 4 and 1 sites, respectively. Gray wolf was the only species that did not show a large difference in detections between the 2 sessions (Figure 5). In the spring, black bear was the most frequently detected species, followed by red fox and coyote. In the fall, grey foxes were the most frequently detected, followed by raccoons and coyotes (Figure 5). We also frequently detected white-tailed deer, red squirrels, snowshoe hares, and on occasion, porcupines, moose, and several species of birds.

Given the higher number of issues observed during the spring, which are still being considered prior to analysis, here we present more detailed results only for the fall survey. Fall cameras were active from approximately September 1 to November 2, for a total of 4,789 'trap-nights' ($\bar{x}$ = 48, SD = 11 trap-nights per camera). Most (n=60) cameras detected between 1 and 3 carnivores species (1 species, n=21; 2 species, n=20; 3 species, n=19); the maximum number of species detected was 7 (Figure 6).

Comparison of Lures and Site-Selection Strategies

Preliminary results suggest that coyotes, raccoons, and skunks may prefer salmon oil over liquid FAS, grey foxes were more likely to be detected at cameras deployed using the enhanced strategy, whereas Gray wolves were detected more often at sites with cameras deployed using the non-enhanced placement strategy (Table 1). Macro-site selection strategies indicated strong differences in the proportion of unlured on-trail versus lured random sites at which some species were detected (Table 1). In particular, preliminary analysis suggests that black bears, fishers, martens, and raccoons were more often detected at lured, randomly-selected sites compared to unlured trails, whereas wolves were more often detected at unlured trail sites (Table 1). A sample of the pictures collected during spring 2016 sampling is shown in Figure 7.

Although many preliminary findings are generally consistent with expectations, more complete and formal analyses will be conducted and presented in future reports. During year 2, protocols will remain the same with the exception that we are employing a crossover design with respect to lure choice (i.e., a site with salmon oil in 2016 will receive FAS lure in 2017). In addition, to partially avoid false triggers in the ongoing spring survey (2017), we decided to postpone the beginning of the sampling period for 2 weeks (from 1 May to 15 May) with the hope of allowing initial canopy growth (more shading) and initial growth of lower-growing herbaceous vegetation that could thus be seen and cut in the detection area at the time of camera deployment. Although reducing trigger sensitivity may also reduce false triggers, initially we were more concerned about potential loss of animal detections from reduced sensitivity.

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Table 1. Number of fall 2016 camera sites in Itasca County, MN at which each species was detected based on a) lure type, and b) micro-site selection strategy. For macro-site selection, we report the percentage of sites where each species was detected to better compare the visitation at on-trail cameras (n=20) versus random lured sites (n=80).

	BADGER	BLACK BEAR	BOBCAT	COYOTE	FISHER	GREY FOX	MARTEN	RACCOON	RED FOX	SKUNK	WEASEL	WOLF
Lure type												
Fatty acid scented oil	2	6	3	7	12	9	10	10	5	6	0	7
Salmon oil	1	4	3	16	16	13	13	16	8	17	0	5
Micro-site selection												
Non enhanced	0	5	3	12	12	8	14	14	7	11	0	9
Enhanced	3	5	3	11	16	14	9	12	6	12	0	3
Macro-site selection (%)												
On-trail	5.0	5.0	5.0	45.0	10.0	60.0	5.0	20.0	25.0	45.0	5.0	50.0
Random	7.5	25.0	15.0	57.5	70.0	55.0	57.5	65.0	32.5	57.5	0.0	30.0

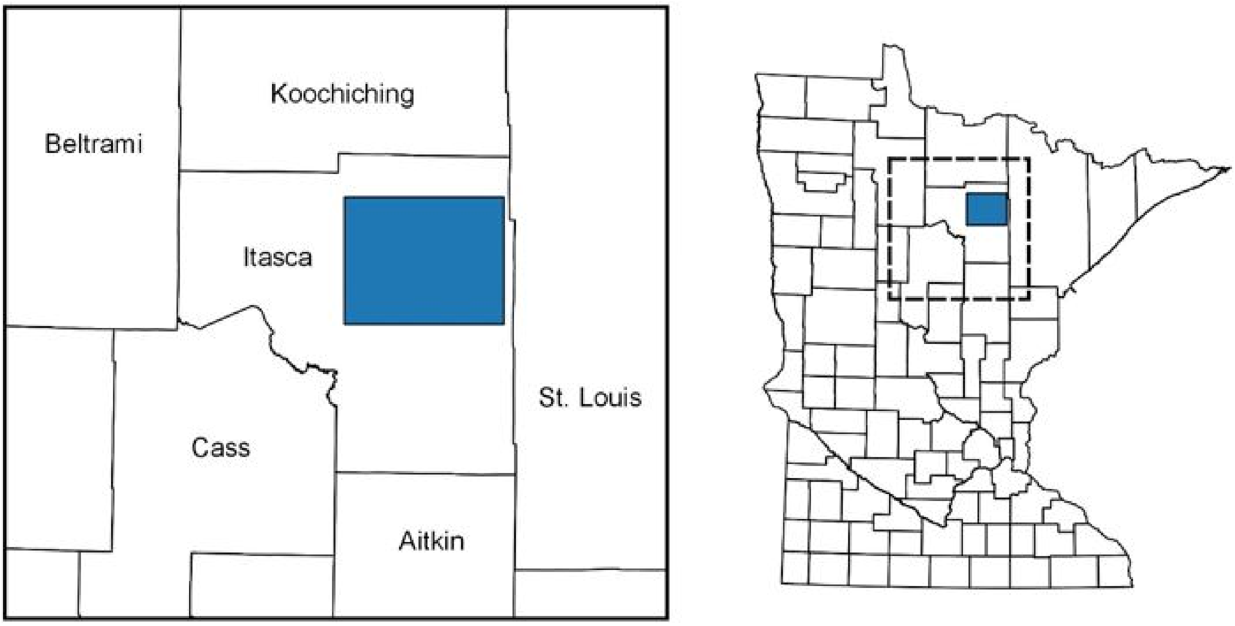


Figure 1. Location of the 2016-17 carnivore camera survey in the north-eastern portion of Itasca County, Minnesota.

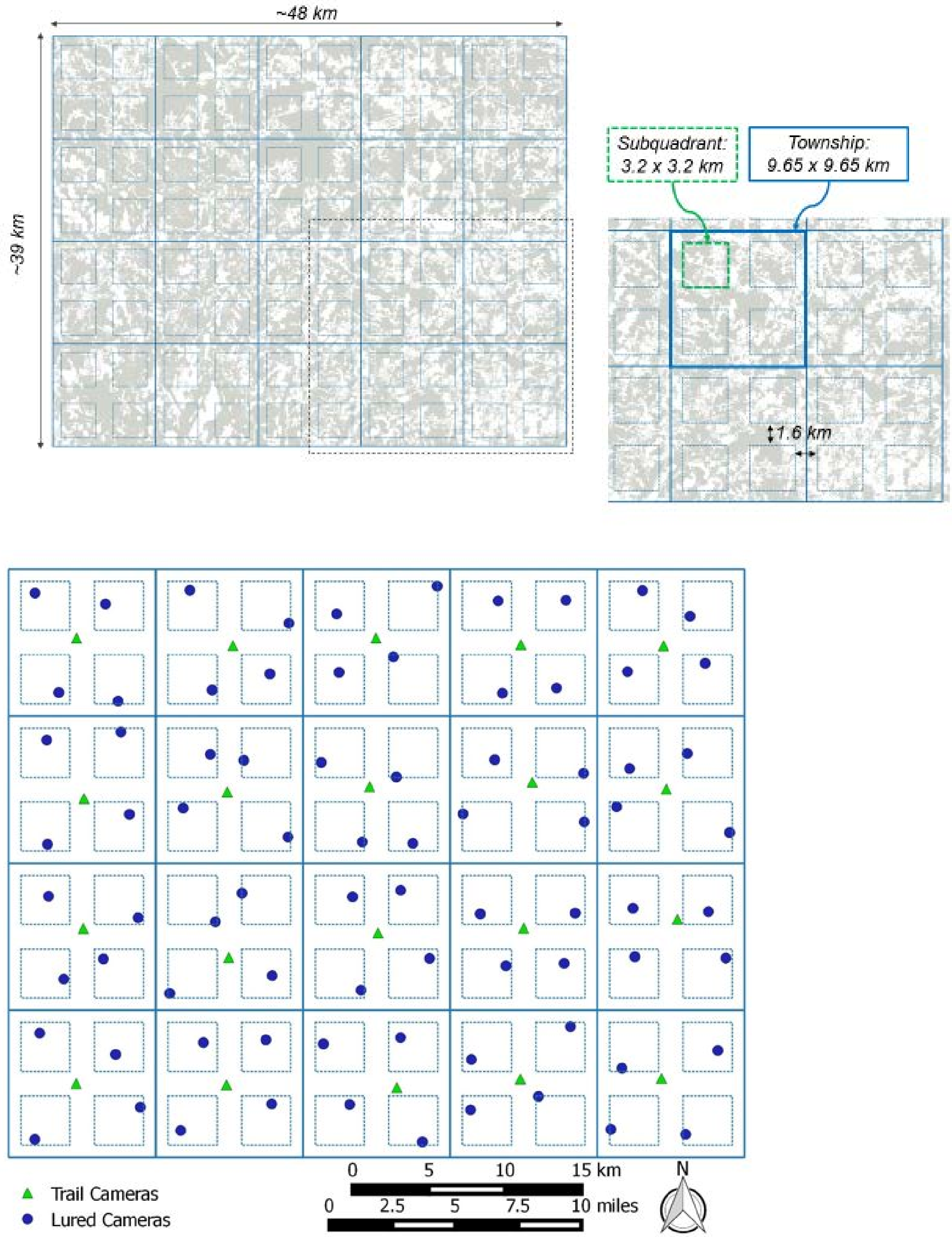


Figure 2. *Top*: Graphic of the Itasca County, MN study area showing forested habitat meeting our macro-site selection criteria in 2016 (*top:* gray areas). In each township (solid blue lines; 9.65 x 9.65 km) we defined four 3.2 x 3.2 km sub-quadrats (green dotted lines). The spacing between adjacent sub-quadrats ensured a minimum distance of 1.6 km (1 mi) between cameras subject to different treatments. *Bottom:* One location for a lured camera was then randomly selected from the suitable area within each sub-quadrat. A fifth un-lured camera was placed outside the quadrats and on a trail nearest the center of the township.

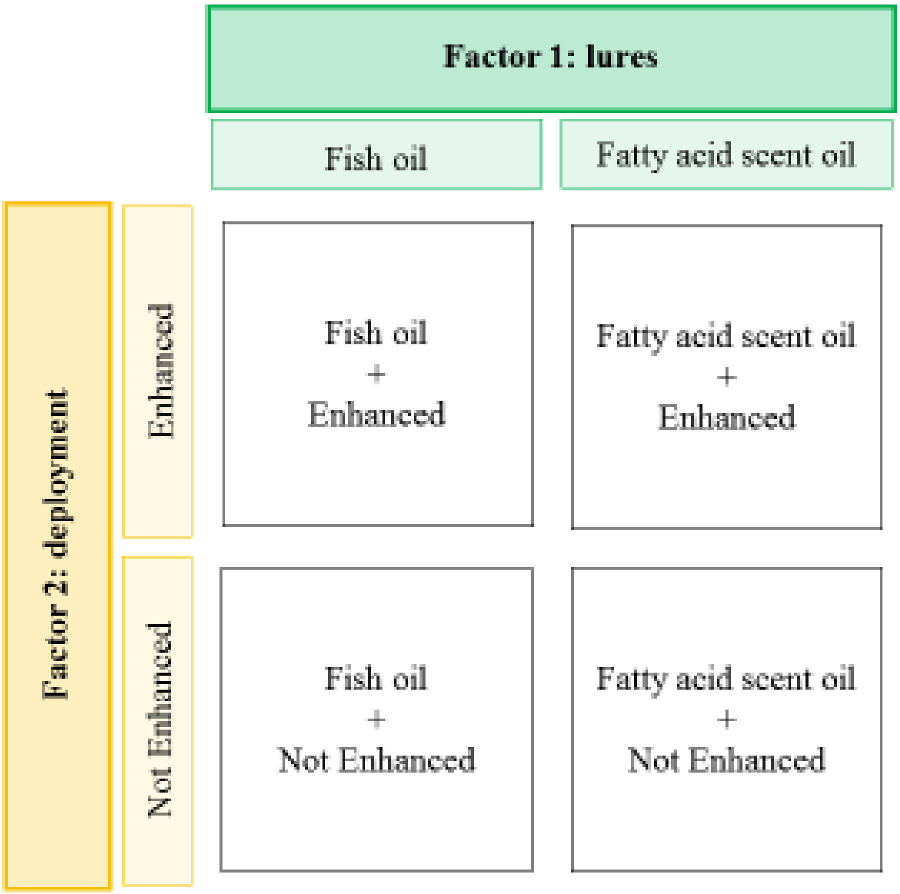


Figure 3. Factorial sampling design, 2016-17. In each of 20 townships in Itasca County, MN, 4 cameras were randomly assigned to one of 4 different treatments given by the intersection between 2 factors: lure type and camera deployment strategy. The lure factor had 2 levels: *fatty acid scent oil* and *fish oil*; the second factor, camera deployment strategy, also had 2 levels: *not enhanced* (i.e., camera placed on nearest tree to the randomly selected UTM location) and *enhanced* (i.e., camera placed at a presumably optimal location within 90 m of the randomly selected point to increase carnivore detection).

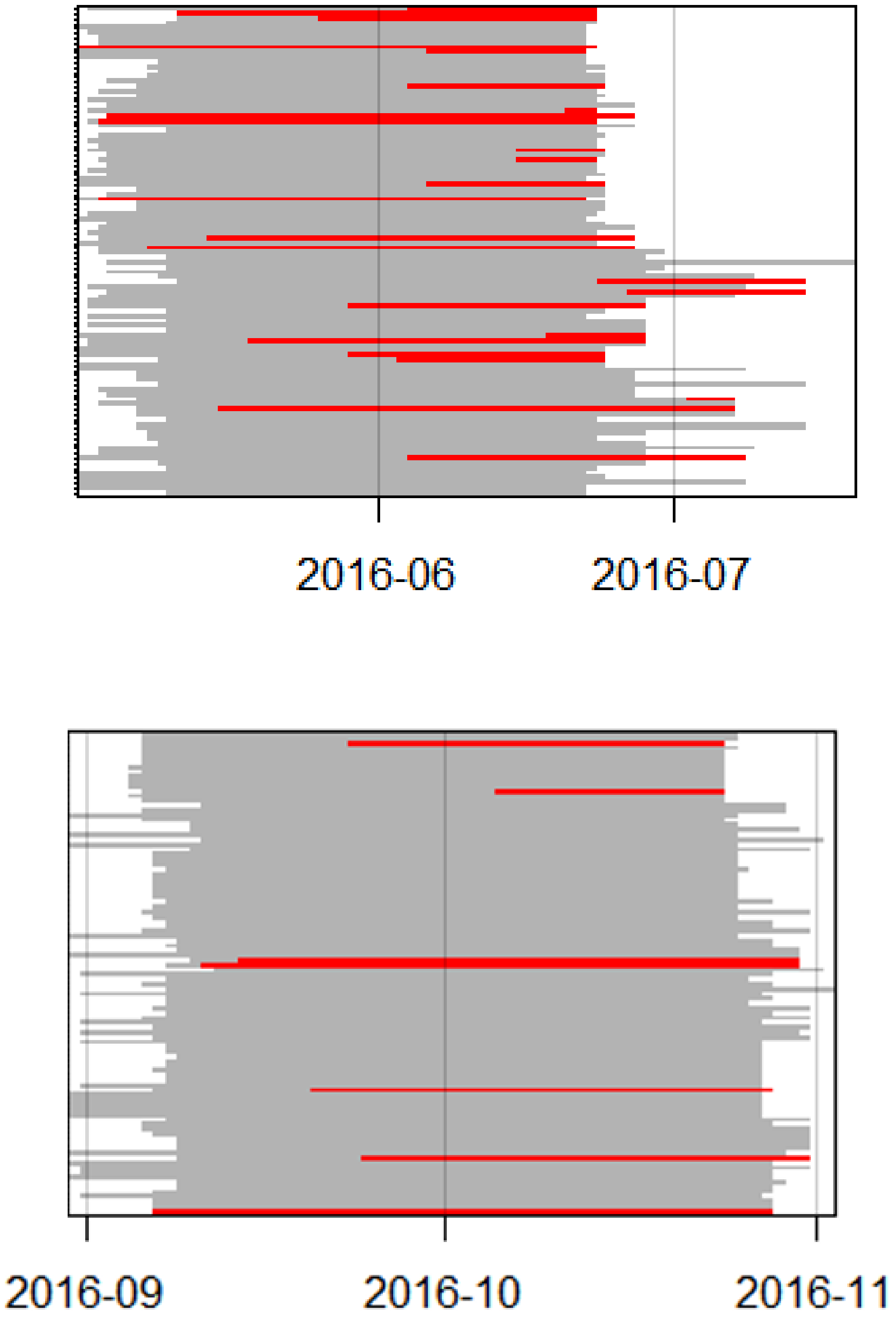
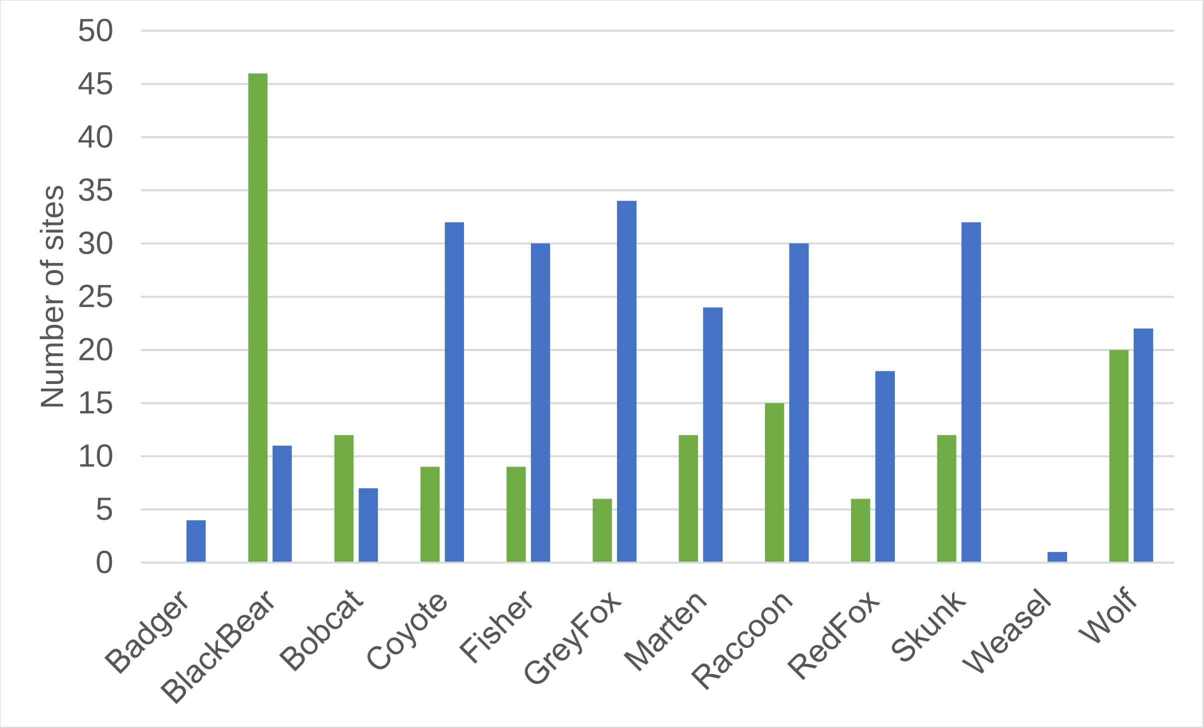


Figure 4. Operating time for each of the 100 cameras deployed in the spring (top) and fall (bottom) 2016, Itasca County, MN. Red segments represent times when cameras were not operable. At the time of the retrieval, 93 cameras were still operating in the fall, whereas only 75 were still operable in spring.



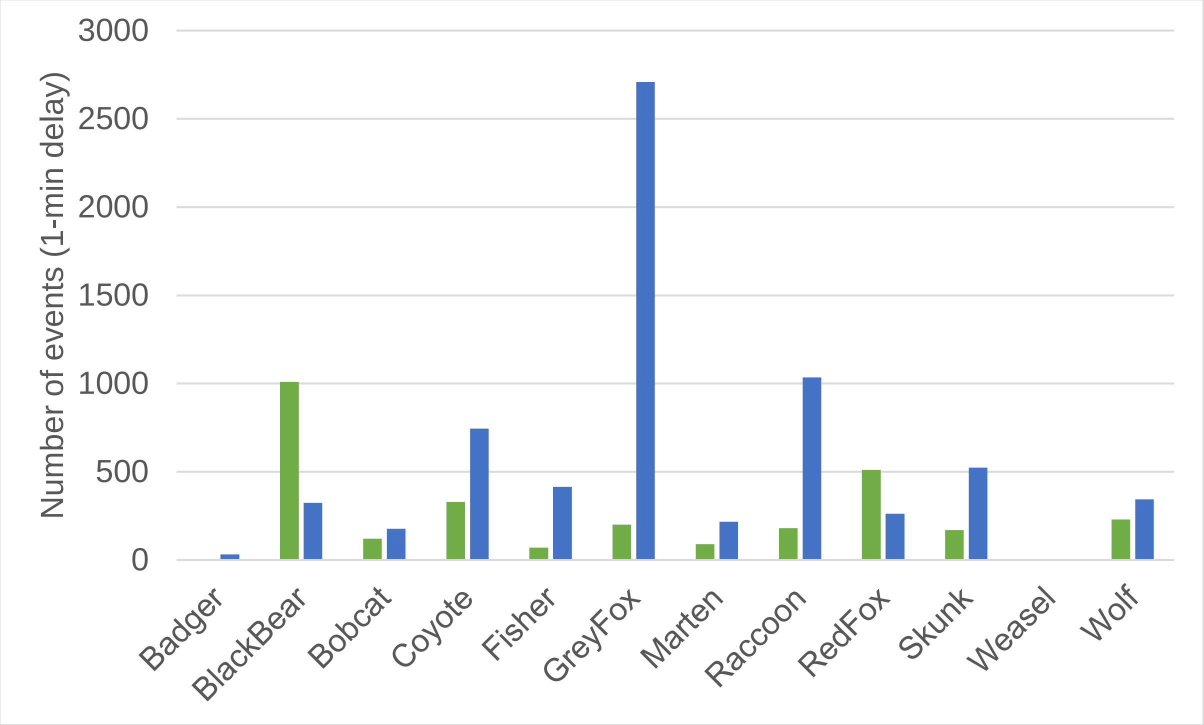


Figure 5. Number of events (bottom) and number of sites (top) at which each species was detected during spring (green bar) and fall (blue bar) 2016 survey, Itasca County, MN. An event was defined as a detection with at least 1 minute delay from the previous picture of the same species at the same site.

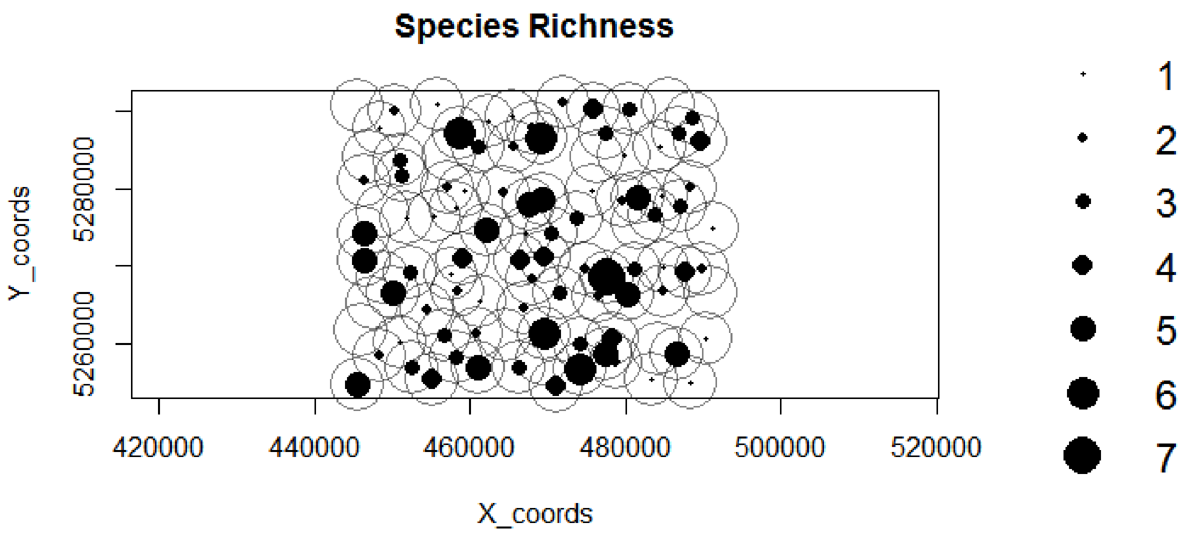


Figure 6. Species richness at each camera location during fall 2016, Itasca County, MN. Most of the cameras detected from 1 to 3 carnivore species.

Figure 7. Example of images collected during the spring 2016 survey, Itasca County, MN. From top-left to bottom-right: gray wolf, red fox, bobcat, bear with two cubs, fisher, raccoon, striped skunk, and coyote.

USING LIDAR DATA TO QUANTIFY FOREST STRUCTURAL HABITAT VARIABLES IMPORTANT TO FISHERS AND MARTENS

Michael Joyce[1], John Erb, Barry Sampson, and Ron Moen[2]

SUMMARY OF FINDINGS

Fishers (*Pekania pennanti*), martens (*Martes americana*), and many other wildlife species rely on three-dimensional structural habitat characteristics to provide essential resources. Spatially-continuous data on fine-scale structural habitat features are generally not available across large landscapes because passive remote sensing systems are not capable of measuring three-dimensional characteristics and because it is financially and logistically challenging to collect field-data continuously across the landscape. Light detection and ranging (LiDAR) is an active remote sensing technology capable of providing accurate, high-resolution data on three-dimensional vegetation structure across large spatial extents. Many past studies have demonstrated that LiDAR data can be used to map coarse- and fine-scale habitat characteristics at the scale of individual trees, field plots, or forest stands. However, most research has focused on forestry applications, and relatively few studies have focused on modeling structural variables that serve as basic wildlife habitat indicators.

We were interested in using LiDAR to supplement field data collected as part of a long-term project on fisher and marten ecology in Minnesota. Our objectives were to evaluate the potential of LiDAR technology to quantify both coarse- and fine-scale forest habitat metrics and to evaluate the effect of pulse density on prediction accuracy. We acquired high-density LiDAR data (8 pulses/m^2) for a portion of our marten study area and selected 200 random locations within that portion to collect detailed vegetation measurements. Random sites were selected using a LiDAR-informed stratified random sampling design. We measured vegetation on 189 of the 200 plots during summer 2015 and 2016; the remaining plots could not be sampled due to wind disturbances that altered forest structure after LIDAR data collection. Statistical analyses are ongoing, and we defer reporting results until final analyses are completed.

INTRODUCTION

To create and implement effective habitat management plans, wildlife managers depend on reliable knowledge of species-specific habitat requirements, accurate information on the current abundance and distribution of suitable habitat features, and an understanding of how management actions influence habitat suitability over a range of spatio-temporal scales. In many situations, having accurate information on abundance and distribution of habitat characteristics is necessary for understanding species-specific habitat requirements and evaluating how management actions influence habitat use. Forest wildlife species vary in their dependence on specific habitat characteristics. For some species, habitat requirements may be adequately

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described using coarse-resolution data such as forest cover type, stand age or successional stage, or proximity to permanent water or other specific landscape features. For these species, broad-scale forest inventory data and GIS layers derived from passive remote sensing technologies (e.g., satellite imagery, aerial photographs) are often adequate to map and monitor changes in habitat quality. However, other wildlife species, including fishers, martens, and many forest songbirds, respond to three-dimensional, structural habitat features at fine spatial scales. Spatially-continuous data on fine-scale structural features generally are not available because passive remote-sensing systems are not capable of measuring three-dimensional characteristics and because it is financially and logistically challenging to collect fine-scale, field-based measurements continuously across large areas. Instead, habitat models for these species typically incorporate information gathered from detailed field-sampling at sites used by the species of interest, often for specific purposes (e.g., foraging, nesting, or denning sites). While site-level habitat models created from field data provide informative and mechanistic insights into a species' habitat requirements, they are often difficult to apply to larger scales at which forest management decisions are generally made. Regardless of whether a species relies on coarse- or fine-scale characteristics, having data on forest characteristics at continuous spatial scales is critical for sound habitat management and assessment.

Light detection and ranging (LiDAR) is an active remote sensing technology capable of providing accurate, high-resolution (<1 to >20 laser pulses/m^2) data on three-dimensional physiographic and vegetative structure over large spatial extents (e.g., entire study areas or wildlife management units up to statewide coverage; Merrick et al. 2013, Vierling et al. 2008). LiDAR data are collected from a scanner that emits frequent, short-duration laser pulses and records the radiation signal returning to the sensor. As the emitted laser pulse is intercepted by an object or surface (e.g., vegetation, building, terrain), a portion of the laser energy is reflected and returned to the sensor. Discrete-return LiDAR systems record the spatial coordinates where the laser pulse intercepted an object or surface, resulting in a three-dimensional "cloud" of interception points or "returns". Modern discrete-return LiDAR systems are capable of recording ≥4 returns per laser pulse (Vierling et al. 2008).

High pulse density, multiple-return LiDAR data provide the detail necessary to accurately map a variety of forest structural attributes including both fine-scale attributes (e.g., canopy height [Means et al. 2000], canopy cover [Lefsky et al. 2002], shrub-density [Martinuzzi et al. 2009]) and coarse-scale attributes (e.g., forest successional stage [Falkowski et al. 2009]) continuously and with high precision across the landscape. Because of these capabilities, LiDAR is increasingly used to analyze forest structure and is becoming an integral part of operational forest management (White et al. 2013). LiDAR can be used to measure biophysical variables at the level of individual trees, forest inventory plots, and forest stands (Falkowski et al. 2006, White et al. 2013). Forest inventory metrics that have been successfully predicted at the plot and stand level using LiDAR include canopy height (Hawbaker et al. 2009, Thomas et al. 2006), canopy density or volume (Lefsky et al. 2002, Martinuzzi et al. 2009), basal area (Means et al. 2000, Woods et al. 2011), average diameter at breast height (Hawbaker et al. 2009, Jakubowski et al. 2013), tree density (Treitz et al. 2012), and forest biomass (Thomas et al. 2006, Treitz et al. 2012, Woods et al. 2011). LiDAR data can be used to make direct estimates for some attributes such as canopy cover, canopy height, and canopy volume (Graf et al. 2009, Lefsky et al. 2002, Merrick et al. 2013). However, many structural metrics require accurate field-plot data that can be used to build predictive models from LiDAR-derived explanatory variables. Overall, studies have focused on forestry-specific metrics and there has been less work focused on predicting structural attributes important to wildlife (but see Goetz et al. 2010, Graf et al. 2009, Hagar et al. 2014, Martinuzzi et al. 2009).

The potential for LiDAR to improve wildlife research and management has been recognized for some time. LiDAR data can be used to improve wildlife-habitat modeling in 2 different ways (Merrick et al. 2013, Vierling et al. 2008). First, it provides a tool that can be used with telemetry data or known species distributions to better understand resource selection. Forest attributes can be measured at fine spatial scales with LiDAR, allowing researchers to assess resource use at scales near those at which animals respond to structural attributes (Vierling et al. 2008). By providing spatially-continuous data, LiDAR data allows researchers to directly address how both landscape composition and configuration influence habitat selection. Furthermore, LiDAR can be used to investigate resource selection across a wide range of spatial scales including sites used for specific behaviors, individual home ranges, and entire wildlife management units or other regional units. Second, LiDAR can be used to predict habitat suitability or species distributions based on prior knowledge of habitat requirements or life-history characteristics. The ability to translate habitat models into spatially-explicit maps is particularly useful for wildlife management, for example, by providing accurate predictions of the distribution and abundance of suitable habitat or by allowing managers to monitor changes in habitat suitability through time with repeated LiDAR acquisitions.

Fishers and martens are 2 species that could benefit from LiDAR-based habitat modeling because they respond to both coarse- and fine-scale forest attributes (Joyce 2013, Raley et al. 2012, Thompson et al. 2012), habitat loss from human land use is thought to be a major threat to population persistence for both species (Proulx et al. 2004), and continuous data on fine-scale attributes required by fishers and martens are not currently available. At coarse scales, fishers and martens show strong selection for mature and old-growth forest conditions (Buskirk and Powell 1994), although both species have been documented using a variety of seral stages (Joyce 2013, Raley et al. 2012, Thompson et al. 2012). Fine-scale attributes, however, appear to drive fisher and marten habitat selection at multiple spatial scales. Both species depend on large-diameter cavity trees and other specific forest structures that serve as rest sites and reproductive dens (Joyce 2013, Raley et al. 2012, Thompson et al. 2012). Sites used for resting and denning typically have dense overhead cover, abundant coarse woody debris, and large-diameter trees (Aubry et al. 2013, Joyce 2013, Thompson et al. 2012). Coarse woody debris provides subnivean access (Corn and Raphael 1992) and is a critical component of marten winter foraging behavior in the boreal forest (Andruskiw et al. 2008). At landscape scales, shrub cover (Slauson et al. 2007) and canopy cover (Cushman et al. 2011, Shirk et al. 2014) are associated with home ranges selected by martens. Furthermore, canopy cover is one of the strongest and most consistent predictors of fisher habitat use across spatial scales (Raley et al. 2012).

Despite the amount of research focused on understanding fisher and marten habitat requirements, there are critical aspects of habitat ecology that are not well understood. For example, several studies have suggested that availability of suitable denning habitat could limit fisher and marten populations (e.g., Ruggiero et al. 1998), but few studies have actually investigated distribution of suitable denning habitat, in part because continuous fine-scale data are needed to apply den-site habitat models across the landscape but are generally not available. Furthermore, most studies have focused on landscape composition, but landscape configuration likely also drives habitat use (Sauder and Rachlow 2014), and landscape configuration is strongly influenced by ownership and management history (Cohen et al. 2002, Kennedy et al. 2012, Spies et al. 1994). Because of their dependence on structural features that have been accurately predicted using LiDAR, LiDAR data has the potential to provide novel insights into fisher and marten habitat ecology and improve habitat management for these species.

Many of the resources exist for LiDAR data to be incorporated into natural resource management in Minnesota. Minnesota is one of a growing number of states for which statewide LiDAR data have already been acquired. One important question that still needs to be addressed to use the statewide data or direct future LiDAR acquisitions is what pulse density is required to accurately quantify forest structural attributes at plot and stand levels. LiDAR acquisition costs increase with increasing pulse density (Jakubowski et al. 2013). Therefore, acquiring LiDAR data at the minimum pulse density necessary for accurate projections will enable researchers and managers to maximize gain from finite resources. Previous research has shown that many forest metrics can be accurately predicted at fairly low pulse densities and that higher pulse density does not necessarily improve model accuracy, but the effect of pulse density on model accuracy depends on the variable of interest (Thomas et al. 2006, Treitz et al. 2012, Jakubowski et al. 2013). In general, the structural variables measured in these studies are strongly biased toward forestry applications. Although some of the biophysical variables evaluated are important indicators of wildlife habitat, a better assessment of how pulse density affects wildlife-specific forest attributes (e.g., canopy structure, coarse woody debris, shrub cover) is necessary before LiDAR can be used in the same operational capacity for wildlife management as it is currently being used for forestry.

Our objective was to evaluate the potential of LiDAR technology to quantify both coarse- and fine-scale forest habitat variables and to create applied GIS tools that can be used in day-to-day decision-making by forest and wildlife managers. Additionally, we will evaluate the effect of pulse density on prediction accuracy. This project will provide new information and tools for applied habitat management for fishers and martens, and will also increase the value of data already collected in ongoing research on fisher and marten ecology. Combining LiDAR-derived estimates of forest structural attributes with location data from radiocollared fishers and martens will enable us to address important research questions aimed at improving management of these species in Minnesota.

STUDY AREA

Marten research has taken place in portions of east-central St. Louis and west-central Lake counties in northeastern Minnesota (Figure 1). The marten study area (~1250 km^2) is composed of a variety of forest types including upland mixed coniferous-deciduous forest, lowland conifer or bog, upland coniferous forest, and regenerating forest, as well as marshes, fens, shrublands, and anthropogenic cover types. We acquired high-density LiDAR data for a 65 km^2 portion within the larger marten study area during spring 2014 (Figure 1). The location of the high-density LiDAR acquisition was chosen because it included a large number of locations from radiocollared fishers and martens (i.e., rest sites, dens, and aerial telemetry locations), it encompassed ~100 ground-based vegetation survey sites measured previously as part of the larger fisher/marten research project, and it contained almost all of the forest types and successional stages available throughout the larger marten study area. Both the marten and embedded LiDAR study areas are predominantly public ownership including portions of the Superior National Forest, state, and county lands.

METHODS

There are 2 LiDAR datasets available that provide variable coverage of our study area (Table 1). Both datasets are discrete, multiple-return LiDAR data acquired from fixed wing aircraft during leaf-off conditions. The first dataset (hereafter, statewide data) was collected during spring 2011 as part of the Minnesota elevation mapping project (http://www.mngeo.state.mn.us/chouse/elevation/lidar.html) and provides complete coverage for Carlton, Cook, Lake, and St. Louis counties. The second dataset (hereafter, high-density data) was acquired in spring 2014 over a 25 square-mile portion of the marten study area. In general, specifications from both datasets (Table 1) match recommendations for forest inventory analysis (White et al. 2013). Those that do not (e.g., scan angle) are consistent with published studies that have successfully modeled forest structure using LiDAR (e.g., Treitz et al. 2012 used a scan angle of ±20˚).

Several pre-processing steps are necessary prior to vegetative analysis. Raw LiDAR return points must be classified as ground or non-ground (e.g., vegetation, water, buildings) returns and manual quality assurance/quality control (QA/QC) steps must be taken to verify data conform to desired specifications. Digital elevation models (DEMs) are then created from ground returns and converted to digital terrain models (DTMs). Pre-processing steps have been completed for statewide data. For the high-density LiDAR data, we are using LP360 (QCoherent Software, LLC) for LiDAR point classification and DEM construction.

We are using the area-based approach to create predictive models of forest structural attributes that relate to habitat quality for marten. The area-based approach combines field-plot and LiDAR data to create predictive statistical models that can be projected across an entire landscape (White et al. 2013). The area-based approach has 4 main steps: 1) collect and summarize field-plot data; 2) extract and summarize LiDAR data corresponding to field sampling locations; 3) create and evaluate predictive models; and 4) apply models across the area of interest.

We measured forest inventory plots at random sites distributed throughout the high-density LiDAR acquisition area. We used a stratified random sampling design to ensure field sampling covers a large range of the forest conditions present on our study area (Hawbaker et al. 2009, White et al. 2013). We calculated mean LiDAR return height (m above ground) and standard deviation of return height for each 20- x 20-m cell in the study area to represent the range of structural conditions present throughout the landscape (Figure 2). Each cell in forest condition represented a potential sample location. Sample locations were further stratified into upland and lowland soil types using ecological landtype classifications from the Superior National Forest's terrestrial ecological unit data to ensure sampling covered a variety of soil types. For each broad soil type category, the available sampling space defined by the 2 LiDAR metrics was divided into 8 quantiles for mean return height and 2-3 quantiles for the standard deviation of return height to form 23 sample strata per soil type (Hawbaker et al. 2009). We selected a total of 200 random locations to sample. The number of locations selected per stratum was proportional to the total number of available cells in each stratum throughout the entire study area.

At each randomly-selected location, we measured structural variables within a 400-m^2 (11.3-m radius) circular plot. Plot size was selected to match recommendations for LiDAR-based forest inventory modeling (Laes et al. 2011, White et al. 2013) and corresponds to a 20-m pixel for landscape-level application of predictive models. Structural attributes were selected based on their importance to marten habitat from published literature (e.g., Andruskiw et al. 2008, Allen 1982, Raphael and Jones 1997, Slauson et al. 2007) and previous research in Minnesota (Joyce 2013; Table 2). Sampling protocols were largely based on USDA Forest Inventory and Analysis program protocols to maintain consistency with previous data collected at rest sites and reproductive dens used by radiocollared marten in Minnesota (Joyce 2013). All field measurements were taken in full leaf-on condition, although canopy cover and understory density also were sampled during leaf-off condition for a subset of field plots. During field sampling, locations of field plots were recorded using both consumer-grade (Garmin eTrex 30) and mapping-grade GPS receivers (Geneq SXBlueII+GNSS). The mapping-grade receiver communicated with both GPS and GLONASS satellites and utilized a combination of space-based augmentation system (SBAS) and real-time differential correction to obtain precise locations without post-processing. When using the mapping-grade GPS, we collected points for

≥30 minutes at a rate of ~20 points/min. Preliminary data at geo-referenced survey markers suggested mapping-grade GPS locations collected this way provided sub-meter accuracy under full forest canopy. For the consumer-grade GPS, we used location averaging for ≥30 minutes.

LiDAR can be used to directly measure a subset of the forest attributes being measured at field plots (e.g., canopy height, canopy cover/closure, canopy structure metrics; Merrick et al. 2013, White et al. 2013). For remaining attributes, we will create predictive statistical models using LiDAR metrics as explanatory variables and attributes summarized from field plot data as response variables. We will use FUSION software (McGaughey 2013) to extract LiDAR point clouds corresponding to field plots and summarize statistical properties of individual point clouds based on return height, return intensity, or point density for use as explanatory variables in statistical modeling.

The type of statistical model we used depended on the structural characteristic. We used multiple linear regression for continuous variables (e.g., average diameter at breast height). We used Poisson or negative binomial GLM count models for count variables (e.g., tree density). Snags were not present at a large number of plots. Consequently, Poisson GLM count models and multiple linear regression could not account for inflated zeros, and use of these types of statistical models could produce biased estimates of snag characteristics (Russell 2015, Zuur and Ieno 2016). We used zero-altered (hurdle) models for snag density (zero-altered Poisson), snag volume (zero-altered gamma), and average snag diameter (zero-altered gamma). Zero-altered models have 2 components (Zuur and Ieno 2016). The first component accounts for presence/absence of snags, while the second component accounts for snag density, volume, or diameter if snags were present.

Despite differences in model type, we used the same statistical framework for all forest structural variables. There are 3 steps in the statistical framework: 1) model-fitting and model selection, 2) model evaluation using cross-validation, and 3) model re-calibration. First, for each response variable, we created a set of candidate models using individual predictor variables or combinations of non-collinear predictor variables. The number of predictor variables included in multi-variate models did not exceed sample-size-based recommendations to avoid over-fitting data (Babyak 2004, Guidice et al. 2012). Models were fit in Program R (R Development Core Team, 2013) using techniques and packages best-suited to the type of model being fit. Candidate models were compared using an information-theoretic approach to select the best-supported model(s) from the candidate set (Burnham and Anderson 2002). Candidate models were chosen based on expected relationships between response variable and individual predictor variables. Second, we evaluated how well best-supported models predicted new data using a 5-fold cross-validation procedure. We evaluated each cross-validation set using root mean squared error (RMSE), R^2, and bias. Finally, we used a bootstrapping procedure to re-calibrate model coefficients in an effort to reduce the effect of over-fitting and therefore improve prediction accuracy (Harrell 2001, Giudice et al. 2012, Fieberg and Johnson 2015).

To evaluate the effect of LiDAR pulse density on accuracy of predictive models we will subsample LiDAR data to obtain 7 different pulse densities (8, 6, 4, 2, 1, 0.5, and 0.25 pulses/m^2) using FUSION software. Subsampling will be performed in a way that accurately simulates data acquired at specific pulse densities (i.e., we wish to thin the density of laser pulses rather than the number of returns per pulse). Predictive models will be created at each pulse density, and prediction accuracy will be plotted as a function of pulse density (Jakubowski et al. 2013). Prediction accuracy will be assessed using R^2, RMSE, and bias. From these plots we will determine the minimum pulse density necessary to create accurate predictive models (turning point, *sensu* Jakubowski et al. 2013) as well as the pulse density corresponding to the most accurate predictive model (best accuracy *sensu* Jakubowski et al. 2013). Results from this analysis will determine which forest attributes can be predicted throughout the entire marten study area using statewide LiDAR data (0.45 pulses/m^2).

RESULTS AND DISCUSSION

Pre-processing steps (QA/QC, point classification, DEM creation and conversion) have been completed for the statewide LiDAR data. High-density LiDAR data were collected during spring 2014 and delivered from the vendor during fall 2014. We have completed QA/QC on the high-density data and classified returns for large portions of the dataset. We are still refining point classification protocols. DEMs will be created and converted to DTMs once we complete point classification. Additional information about point classification and DEM construction is not provided here because methodology is still being refined.

Our 200 randomly-selected field plots included 115 plots in upland soil types and 85 plots in lowland soil types. During summer 2015, we measured 100 forest inventory plots. Data from these plots have been entered and checked for errors. We measured 89 additional plots during summer 2016, and completed data entry for all plots. We were not able to measure all 100 remaining plots in 2016 because wind storms altered some of the pre-selected plots before we could measure them. The final set of 189 field plots includes 110 plots in upland soil types and 79 plots in lowland soil types. We have started preliminary statistical analyses, but we defer results until all statistical analyses are completed.

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Table 1. Specifications for statewide (2011-12) and high-density (2014; portion of St. Louis County) LiDAR datasets collected in Minnesota.

Specifications	Statewide	High-resolution
Acquisition date(s)	Spring 2011 & Spring 2012	Spring 2014
Vendor	Wolpert, Inc.	AeroMetric, Inc.
Laser system(s)	ALS60, ALS70, and Optech GEMINI	ALS70
Altitude	2000-2300 m	1050 m
Flight speed	240 - 278 km/h	278 km/h
Scan angle	± 20˚	± 20˚
Side Ooverlap	25%	50%
Nominal point spacing	≤ 1.5 m	≤ 0.35 m
Pulse density	0.45 pulses/m^2	8.0 pulses/m^2
Vertical accuracy	5.0 cm (RMSE)	6.7 cm (RMSE)
Horizontal accuracy	1.16 m (95% confidence)	100 cm

Table 2. Partial list of forest attributes that will be estimated using LiDAR data collected in Minnesota from 2011-14. Attributes were selected because of their biological significance to martens.

Forest attribute	Biological significance	Citation(s)[a]
Coarse woody debris density/volume	Prey habitat, facilitates prey capture, subnivean access, rest and den site characteristic	Andruskiw et al. (2008), Corn & Raphael (1992), Joyce (2013)
Tree diameter at breast height (dbh)	Indicator of stand age, related to arboreal denning and resting structures	Raphael & Jones (1997), Slauson & Zielinski (2009)
Basal area	Indicator of stand age, related to arboreal denning and resting structures	Payer & Harrison (2003,2004)
Canopy closure	Open canopy forests and non-forested habitat associated with predation risk and low prey availability	Slauson et al. (2007), Moriarty et al. (2015)
Canopy structure/heterogeneity	Associated with structural diversity of stands	Zielinski et al. (2006), Weir et al. (2012)
Stand height	Indicator of developmental stage	Bowman & Robitaille (1997)
Sapling density	Provides habitat for prey species (snowshoe hare) and may serve as escape cover	Carreker (1985), Slauson et al. (2007), Joyce (2013)
Shrub density	Provides habitat for prey species (snowshoe hare) and may serve as escape cover	Carreker (1985), Slauson et al. (2007)
Snag density/volume	Indicator of stand age and vertical complexity	Gilbert et al. (1997); Slauson & Zielinski (2009)
Horizontal cover	Related to sapling and shrub density; may serve as escape cover or provide habitat for prey species (snowshoe hares)	Carreker (1985), Slauson et al. (2007)

[a] Citation for biological significance of attribute to martens.

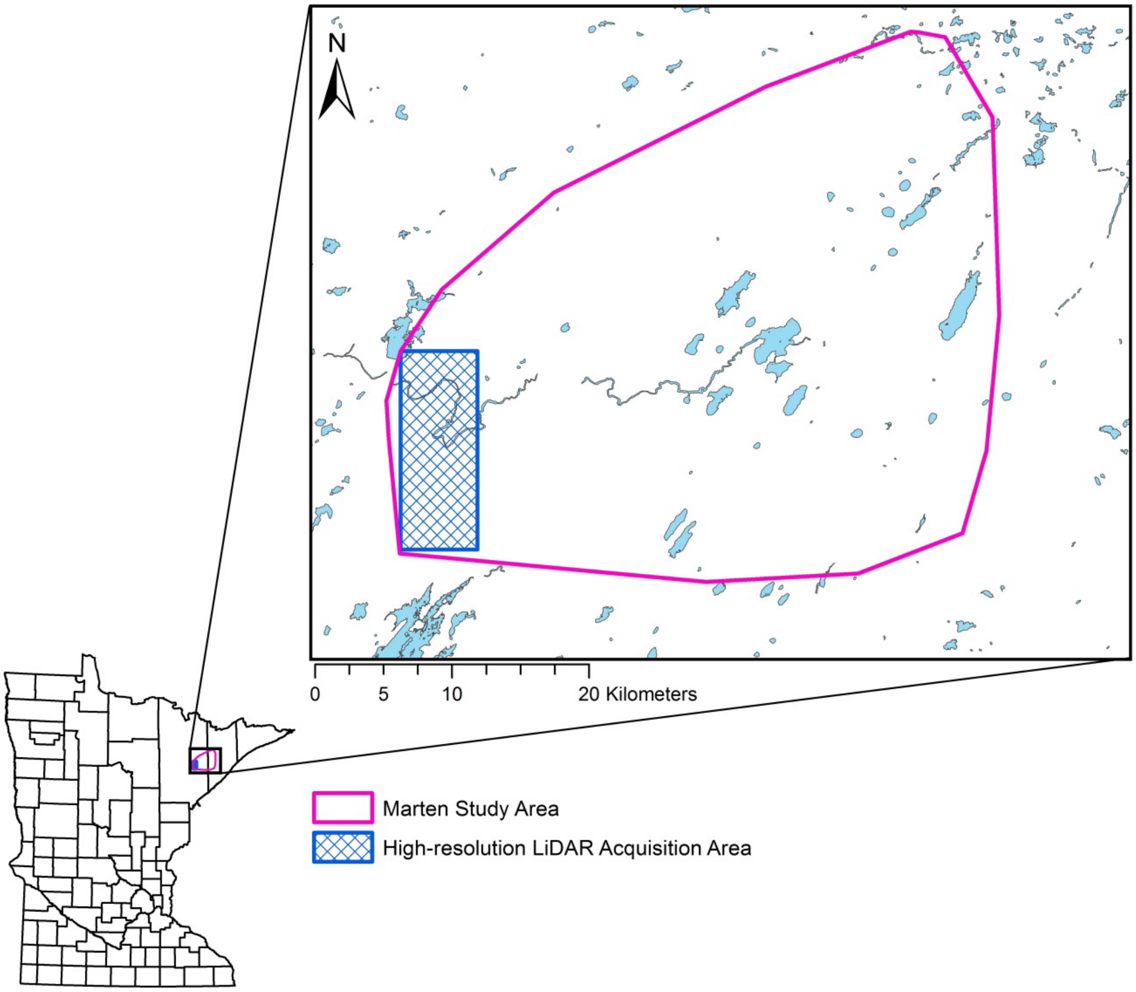

Figure 1. Map of primary marten study area in northeastern Minnesota with the location where high-density LiDAR data were acquired in 2014.

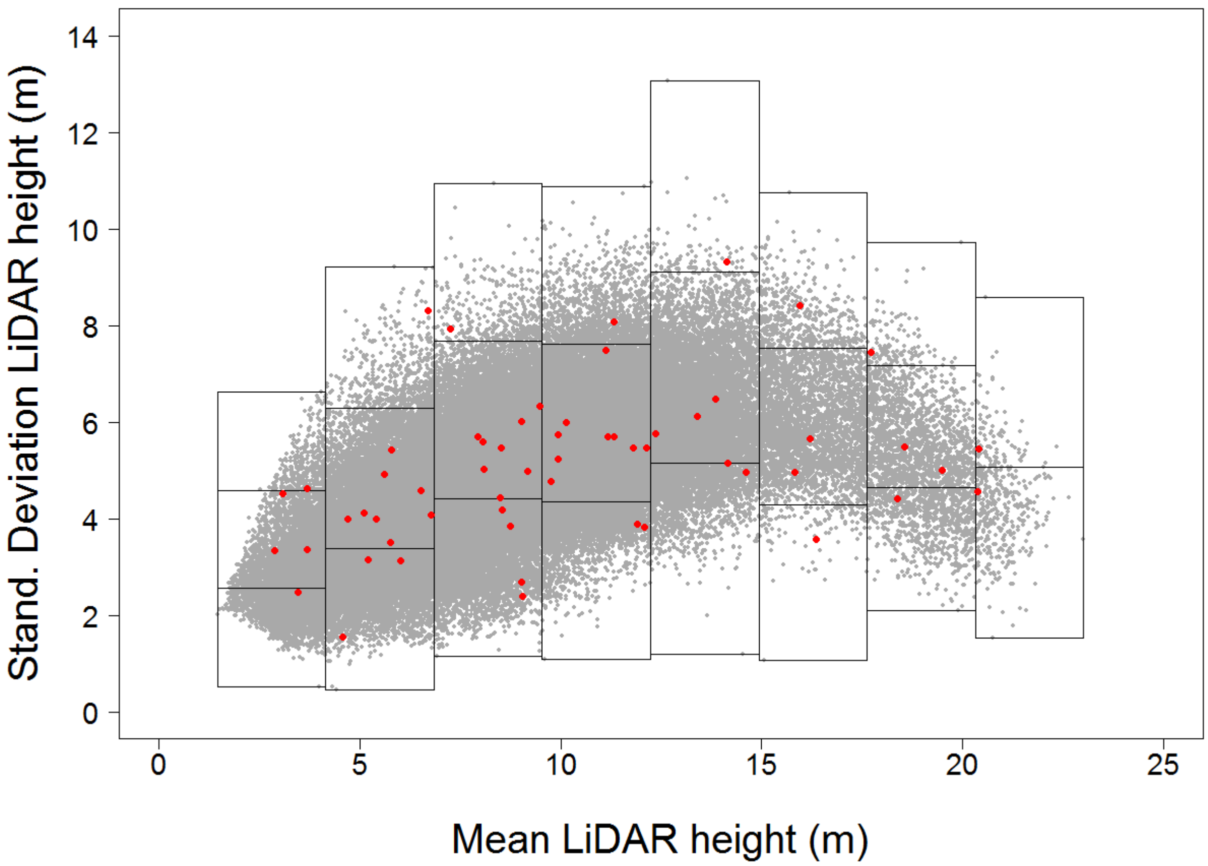

Figure 2. Sampling space for LiDAR-informed stratified random sampling design in a 25 mile^2 portion of St. Louis County, Minnesota. Structural variability within the study area is represented by mean and standard deviation in LiDAR return height for each 20 m pixel in the study area (gray circles). Black squares represent strata from which a random sample of plots was selected (red circles) and surveyed from 2014-16. Stratification was performed separately for areas with upland and lowland soil types.

PREDICTING THE EFFECTS OF GRASSLAND CONSERVATION RESERVE PROGRAM ENROLLMENTS AND EXPIRATIONS ON GREATER PRAIRIE-CHICKENS IN NORTHWESTERN MINNESOTA

Kalysta Adkins[1], Charlotte Roy, Robert Wright[2]

ABSTRACT[3]

The Conservation Reserve Program (CRP) has the potential to influence the abundance of greater prairie-chickens (*Tympanuchus cupido pinnatus),* a species of special concern in Minnesota, by altering the amount and configuration of grassland and wetland in agriculturally dominated landscapes. However, the CRP has experienced recent declines in enrollments in northwestern Minnesota, and these declines are expected to continue following the reduced enrollment cap in the 2014 Farm Bill, which funds the program through 2018. These cuts increase the need to prioritize CRP reenrollments or new enrollments that are likely to have the most impact on greater prairie-chicken populations. To predict changes in greater prairie-chicken abundance caused by expirations of CRP contracts and target CRP enrollments at both the landscape and lek scale, we used models relating lek density and the number of males at leks to CRP enrollments and the resulting landscape structure. We simulated different land cover scenarios of CRP contract expirations, and results indicated that the abundance of greater prairie-chickens would be negatively impacted. Simulations of targeted CRP contract enrollment suggested mixed effects on greater prairie-chicken abundance. Adding grassland cover that increased existing grassland contiguity had a positive impact, while additions that decreased contiguity had a negative impact. Landscapes with a large proportion of existing CRP grasslands and wetlands are most likely to continue to support high prairie-chicken abundance through reenrollment and enrollment of new contracts that are large and contiguous with existing grassland and wetland cover types. Our findings highlight the importance of maintaining existing CRP grasslands and wetlands in landscapes that currently have low levels of grassland and wetland cover.

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MULTISCALE ASSOCIATIONS BETWEEN GREATER PRAIRIE-CHICKENS, GRASSLAND CONSERVATION RESERVE PROGRAM ENROLLMENTS, AND LANDSCAPE COMPOSITION IN NORTHWESTERN MINNESOTA

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ABSTRACT[3]

Both the abundance of greater prairie-chickens (*Tympanuchus cupido pinnatus)* and the area enrolled in Conservation Reserve Program (CRP) have undergone recent declines in northwestern Minnesota. Although wildlife conservation is a stated objective of the CRP, the impact of CRP grassland on greater prairie-chicken populations has not been quantified. To address that information need, we evaluated the association between greater-prairie chicken population indices (i.e., lek density (leks/km^2) and number of males per lek) and CRP enrollments in the context of landscape structure and composition in northwestern Minnesota. We used data from the standardized annual prairie-chicken surveys coordinated by the Minnesota Department of Natural Resources and land cover data in 17 42-km^2 survey blocks during the period 2004-2016. We used a mixed-effect model and a layered approach in an information-theoretic framework at multiple spatial scales to identify covariates related to prairie-chicken abundance. At the landscape scale, the best-supported model for lek density included the amount of CRP grassland; state-, federal-, and The Nature Conservancy (TNC)-managed grasslands; CRP wetland; state-, federal-, and TNC-managed wetlands, "other" wetlands; the contiguity of grasslands; and the number of patches of grasslands and wetlands in each survey block each year. At the lek scale, the best-supported model to explain the number of males/lek included the amount of CRP grassland; state-, federal-, and TNC-managed grasslands; CRP wetland; state-, federal-, and TNC-managed wetlands; "other" wetlands; forests; developed areas; shrubs; and the contiguity of CRP grassland. These results suggest that increasing the quantity of grassland and wetland CRP contracts throughout the existing range of greater prairie-chickens in northwestern Minnesota and aggregating CRP grassland contracts in areas of known lek sites may increase greater prairie-chicken abundance.

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DEPARTMENT OF
NATURAL RESOURCES

CHRONIC WASTING DISEASE IN A MINNESOTA WILD DEER HERD: FIRST DETECTION IN FILLMORE COUNTY

Erik Hildebrand, Michelle Carstensen, Margaret Dexter, Chris Jennelle, Lou Cornicelli, and Patrick Hagen

SUMMARY OF FINDINGS

In fall 2016, the Minnesota Department of Natural Resources (MNDNR) sampled 2,966 hunter-harvested white-tailed deer (*Odocoileus virginianus*) for chronic wasting disease (CWD) in southeastern Minnesota. The surveillance effort focused on testing deer within deer permit areas (DPA) in the 300 series zone, in response to increased incidence of CWD in wild deer in both southwest Wisconsin and northeast Iowa. Three deer tested positive for the disease in Fillmore County (DPA 348) and MNDNR enacted its CWD Response Plan which called for an immediate ban on recreational deer feeding, a formal survey of the area CWD was found, creation of a disease management zone (DPA 603), and additional sampling efforts to better understand the prevalence and spatial extent of the outbreak. During a winter (January-March 2017) supplemental surveillance effort, an additional 1,179 samples were tested through 3 operational phases; a special late hunt, landowner shooting permits, and a contract with United States Department of Agriculture–Wildlife Services (USDA-WS) for targeted deer removals. As a result, 8 more CWD positive deer were found. Surveillance efforts for CWD will be intensified in southeastern MN in fall 2017 and also expanded into 2 other areas of the state (Crow Wing and Meeker Counties) where the disease was recently discovered in captive cervid farms.

INTRODUCTION

Chronic wasting disease is a transmissible spongiform encephalopathy (TSE) that affects elk *(Cervus canadensis)*, mule deer *(Odocoileus hemionus)*, white-tailed deer, and moose *(Alces alces)*. TSEs are infectious diseases that alter the morphology of the central nervous system, resulting in a “sponge-like” appearance of this tissue. The etiological agent of CWD is an infectious protein, called a prion. Incubation time of the disease can range from 1.5 to nearly 3 years, although infected animals have been shown to shed prions in their feces up to a year before showing signs of illness (Tamguney et al. 2009, Haley et al. 2011). Clinical signs are non-specific and may include a loss of body condition and weight, excessive salivation, ataxia, and behavioral changes. There is no known treatment or vaccine for the disease and it is always fatal. Experimental and circumstantial evidence suggest that transmission of the disease is primarily through direct contact with infected animals or their infective saliva or excrement (Mathiason et al. 2006, Safar et al. 2008). However, persistence of prions in the environment and resulting indirect transmission has been shown to occur (Miller et al. 2004, Johnson et al. 2007, and Maluquer de Motes et al. 2008).

The Center for Disease Control (CDC) and other public health agencies have concluded there is no known link between CWD and any neurological disease in humans (MaWhinney et al. 2006, Sandberg et al. 2010). However, both the CDC and the World Health Organization (WHO) recommend that no part of a known positive animal should be consumed by humans. Additionally, there is no evidence that CWD can be naturally transmitted to species other than deer, elk, or moose. However, new research conducted by the Canadian Food Inspection Agency has demonstrated that by orally administering muscle under experimental conditions from cervids (deer and elk) naturally infected with CWD, the disease can be transmitted to macaques. This finding has sparked renewed concerns about potential human health risks to eating CWD-contaminated venison (Czub, S. 2017, May).

In December 2016, 2 captive white-tailed deer from a mixed white-tail and mule deer herd in Crow Wing County tested positive for CWD through routine slaughter surveillance. Four other captive facilities in Minnesota received deer within the past 5 years from this CWD-infected herd, these exposed animals were euthanized and tested for the disease. One of these trace-out herds was located in Meeker County and the deer linked to the Crow Wing County farm tested positive for CWD; following whole herd depopulation, 30% of the deer on this farm were infected. The remaining 3 trace out facilities tested negative for CWD.

Currently, Minnesota has approximately 460 captive cervid facilities (Minnesota Board of Animal Health). As the current statewide population estimate of wild deer approaches one million, there is an element of inherent disease transmission risk between captive and wild cervids. Overall, risk is difficult to quantify because deer populations are unevenly distributed over the landscape ranging in densities from < 1-15 deer/km^2 (i.e., 1–40 deer/mi^2), facility fences vary in construction quality, and direct/indirect contact rates between captive and wild cervids are unknown. In addition, captive cervid facilities are sporadically distributed on the landscape and are independent of wild deer densities.

Since 2002, MNDNR has conducted CWD surveillance and sampled >50,000 wild deer. The first occurrence of this disease in wild deer was in 2011, when a wild deer was found with CWD near an infected captive elk facility in Pine Island. Aggressive surveillance efforts from 2011-2013 tested an additional >4,000 deer in the surrounding area, and failed to detect another case of CWD in the wild. Since that time, MNDNR has been closely monitoring disease spread in the neighboring states and conducting additional surveillance efforts to ensure early detection of the disease, if present in Minnesota.

METHODS

Hunter-harvested surveillance was conducted at deer registration stations during the first 3 weekends and first week (only a subset of stations) of the 2016 regular firearm hunting season in southeastern Minnesota. Selected stations were staffed with MNDNR personnel and students, trained in lymph node collection. Stations were selected based on deer volume and distribution throughout the surveillance zone to meet sampling goals. Eight taxidermists were trained and collected samples throughout the entire archery season from trophy bucks harvested within our surveillance DPAs. Hunters were asked to voluntarily submit medial retropharyngeal lymph node samples from deer ≥1.5 years of age to be tested for CWD. All samples were inventoried, entered into a database, and sent to Colorado State University (CSU) for enzyme-linked immunosorbent assay (ELISA) testing. Any presumptive positive deer from ELISA testing would be confirmed using immunohistochemistry (IHC) testing at CSU.

At the time when deer were sampled, hunter information was recorded, including the hunter’s name, a telephone number, MNDNR number, and location of harvest. Maps were provided to assist the hunters in identifying the location (Township, Range, and Section) of the harvest site. Cooperating hunters were given a cooperator’s patch and entered into a raffle to win one of two, .50 caliber muzzleloaders or a compound bow package donated by Minnesota Deer Hunters Association (MDHA) and The Bluffland Whitetails Association. Hunters were not notified of their testing result unless it was positive.

Following detection of the disease near Preston, the public was invited to receive information about the CWD response plan, and to have questions answered by MNDNR. On the evening of December 15, 2016, >700 local landowners, hunters, media personnel, and general public attended a meeting held at the Preston Elementary School to better understand MNDNR’s plans going forward regarding the response to CWD in their local community.

A disease management zone, called DPA 603, was created to encompass a 10-mile radius around 2 CWD-infected deer and used enforceable boundaries such as county highways and township roads (Figure 1). An aerial survey was conducted in DPA 603 to better understand overall deer densities, local concentrations of deer, and locations of artificial feeding sites. From December 14 – 21, a helicopter was used to complete the survey. Towards the end of the survey, confirmation of the 3rd CWD-positive deer required an expansion of the flight area to include a northern “bump out”, which encompassed an additional 10 mi radius around this case. The survey design included 104 plots in a 1317.5-km^2 (508.7-mi^2) area. Following the survey, a census was flown around each of the known positive deer. To prevent further disease transmission, MNDNR banned recreational feeding and use of attractants for deer in a 5-county area in southeastern Minnesota (Figure 2). During winter (Jan-March 2017) effort was made to collect additional samples from DPA 603 and the northern “bump out” to help understand disease prevalence, and geographic extent. This was obtained through 3 operational phases; a special late season hunt, landowner shooting permits, and a contract with the United States Department of Agriculture-Wildlife Services (USDA-WS) to remove additional deer using sharpshooting.

First, during the special late season hunt 31 December 2016 to 15 January 2017, any person who had possession of either an unused 2016 deer license or a special disease management license (available for a reduce cost of $2.50 and unlimited bag) could harvest deer only inside DPA 603. Harvested deer were brought to one of 4 registration check stations, where CWD sampling of all deer >1 year old was required, and all deer received a special carcass tag from MNDNR staff. Carcass movement restrictions required the carcass remains could not leave the DPA 603 until a test negative result was received. However, 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 surveillance boundaries immediately. A MNDNR-leased refrigerated semi-trailer was provided for hunters to use, along with a lined dumpster to dispose of carcass remains; both items provided hunters viable options after harvesting deer to keep carcasses inside the zone until a test negative result was reported. Hunters checked their results on the MNDNR website using either their MNDNR number or the special carcass tag number assigned to their deer.

The second operational phase offered special shooting permits to landowners in DPA 603 as well as the northern “bump out”, from 16 January 2017 to 12 February 2017. Landowners inside this area didn’t have to own a minimum amount of acreage to qualify for a shooting permit, but did have to abide by city and state ordinances for discharging firearms. There were no limit to the number or sex of deer that could be harvested from the owner’s property and they could designate as many shooters under their permit as desired. The use of high powered center-fire rifles under this permit was allowed and since this area was historically regulated as a shotgun-only zone during the firearm season, the ability to use rifles during this sampling effort was viewed as a unique opportunity by many landowners. The landowner was required to contact MNDNR staff within 24 hours of harvesting by calling a MNDNR CWD hotline and trained staff either traveled to the landowner’s site to collect samples or deer were brought into the Preston Forestry Office for sampling. Each carcass was given a unique identification tag, and landowners were directed not to transport carcasses outside the surveillance area until a test negative result was received. 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 surveillance boundaries immediately.

The third operational phase was a contract with USDA-WS to use sharpshooting at bait piles from mid-February through mid-March to obtain additional samples surrounding areas where infected deer were harvested. By this time in our efforts, we had identified 2 core areas of concern, a 31-km2 (12-mi2) area surrounding the first 2 positive deer and a 23-km2 (9-mi2) area around the third positive deer, and sharpshooting efforts were focused in these areas. USDA-WS obtained permissions from private landowners to access their properties, place bait if needed, and target deer during evening and overnight hours. Intact carcasses were transported to the Preston Forestry office where a processing facility was set up. Here the deer were eviscerated immediately upon delivery, and samples were collected including medial retropharyngeal lymph nodes, a central incisor for aging, muscle tissue for genetic signatures, and blood collected from the heart for arbovirus screening. Data was also collected which included deer harvest location, age class and sex, pregnancy status of females and number and sex of fetuses. A unique carcass tag was issued to each individual animal by MNDNR staff. Entrails were deposited in a lined dumpster and all carcasses were held in a MNDNR-leased refrigerated trailer at 33-38 °F until test-negative results were reported (typically within 4 business days). All test negative deer went to a recipient from a venison donation list that contained more than 400 people or were given back to the landowner from where the deer was harvested. Any CWD-positive deer carcasses were disposed of by alkaline digestion at the University of Minnesota, Veterinary Diagnostic Laboratory in St. Paul, MN.

While MNDNR staff were working inside DPA 603 through each operational phase of winter sampling, opportunistic deer such as vehicle-kills, and found dead or opportunistic sick deer that were reported by the public were also collected and sampled for disease

Across all of MN, MNDNR routinely samples any cervid exhibiting clinical symptoms of CWD infection (opportunistic surveillance). We have disseminated information to wildlife staff regarding clinical signs of infection for symptomatic deer. These staff were also provided with the necessary equipment and training for lymph node removal and data recording. The number of samples expected through opportunistic statewide surveillance is estimated to be less than 100 animals annually, since few reports of deer with clinical signs are received.

RESULTS AND DISCUSSION

A total of 2,966 samples were collected in southeastern Minnesota from hunter-harvested deer during fall 2016; 200 of these were collected through participating taxidermists. Three deer tested positive for CWD, 2 of these deer were sampled at check stations and 1 deer was sampled by a taxidermist. All 3 deer were located in a relatively small geographic area near Preston, MN (Figure 3). Our CWD Response Plan was enacted shortly after the initial detection of disease was confirmed in December 2016.

The aerial survey estimated the deer population in DPA 603 and the northern "bump out" to be 11,656 deer, equating to an estimated deer density of 61 deer/km2 (23.6 deer/mi2) (Figure 4). Deer densities were highest within a 31-km2 (12-mi2) area surrounding the first 2 positive deer and within a 23-km2 (9-mi2) area around the third positive deer, with an average of 90 deer/km2 (35 deer/mi2).

A total of 626 deer were sampled for CWD during the special late hunt, with 3 testing positive for disease. A total of 411 permits were issued to landowners during the landowner shooting permit phase and 269 deer were sampled for CWD. Only 133 (32%) of the landowners that received shooting permits removed at least 1 deer, and of those, 71 (53%) landowners took only 1 deer and 10 (7%) landowners took 5 or more deer. Two deer tested positive for CWD through this operational phase. Finally, during the sharpshooting contract with USDA-WS, 238 additional deer were removed and tested; 2 were found positive for CWD.

Through this combined winter surveillance effort, a total of 1,179 deer (1,142 adults, 37 fawns) were sampled in our CWD surveillance area; 8 deer tested positive for the disease (Figure 5). Sampling included deer taken through the special late hunt (n = 626), landowner shooting permits (n = 269), contract with USDA-WS deer removal (n = 238), vehicle-kills (n = 30), found dead deer (n = 13), and opportunistic sick deer (n = 3) (Figure 6).

In total, 246 deer were issued to recipients that were on the venison donation list. The MNDNR-leased refrigerated trailer was utilized for the duration of the winter surveillance effort and 521 deer were held; 188 deer during the special late hunt, 91 deer in the landowner shooting permit phase, and 242 deer during the contract with USDA-WS.

We estimated the fall 2016 surveillance effort cost $364,000. The estimated total cost of the winter sampling effort was $557,800. This can be broken down into staff salary ($235,800), fleet ($35,200), travel expenses ($60,700), equipment leases or rentals ($14,100), USDA-WS contract ($144,000), and diagnostic testing ($19,200) By operation phase, we spent $136,000, $162,800, and $259,000 for the special hunt, landowner shooting permits, and sharpshooting, respectively.

From July 2016 to June 2017, MNDNR collected a total of 60 samples through opportunistic surveillance efforts. This included samples from 2 escaped captive deer, 1 possible escaped captive mule deer, 1 elk that was mistakenly shot by a deer hunter outside of MN's elk range, head and carcass remains from 1 elk that was found dumped in a gravel pit outside of elk range, 8 vehicle-killed deer, and 46 free-ranging deer with clinical signs; all samples were negative for CWD.

Future Surveillance Plans

CWD surveillance will take place inside DPA 603 for all deer harvested in fall 2017. Mandatory sampling of adult (≥ 1.5 years of age) deer and restricted whole-carcass movements inside DPA 603 will continue to be in effect. The MNDNR plans to sample 7,200 hunter-harvested deer for CWD during the opening weekend of firearm season through a mandatory sampling framework for hunters in DPAs: 343, 345, 346, 347, 348, 349, 155, 171, 172, 242, 246, 247, 248, 249, 218, 219, 229, 277, 283, and 285. This effort is in response to the recent detection of CWD in 2 captive cervid farms (Crow Wing and Meeker counties) and in wild deer (Fillmore County). Targeted CWD surveillance of deer exhibiting clinical signs of illness will continue statewide.

ACKNOWLEDGMENTS

We would like to thank all the MNDNR Wildlife and Enforcement staff, who volunteered to assist with this disease outbreak surveillance project. 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 tackling our GIS mapping needs and Pete Takash for his efforts in communications and internet-related work. We would also like to thank Andrew Herberg, Kelsie LaSharr, David Pauly, and Robby Rathbun for their help through this effort. Additional thanks to Brian Haroldson, Ryan Tebo, and DNR Pilot Brad Maas for their aerial deer survey work. We very much appreciate the support of the USDA-Wildlife Services disease biologist Tim White and his entire crew.

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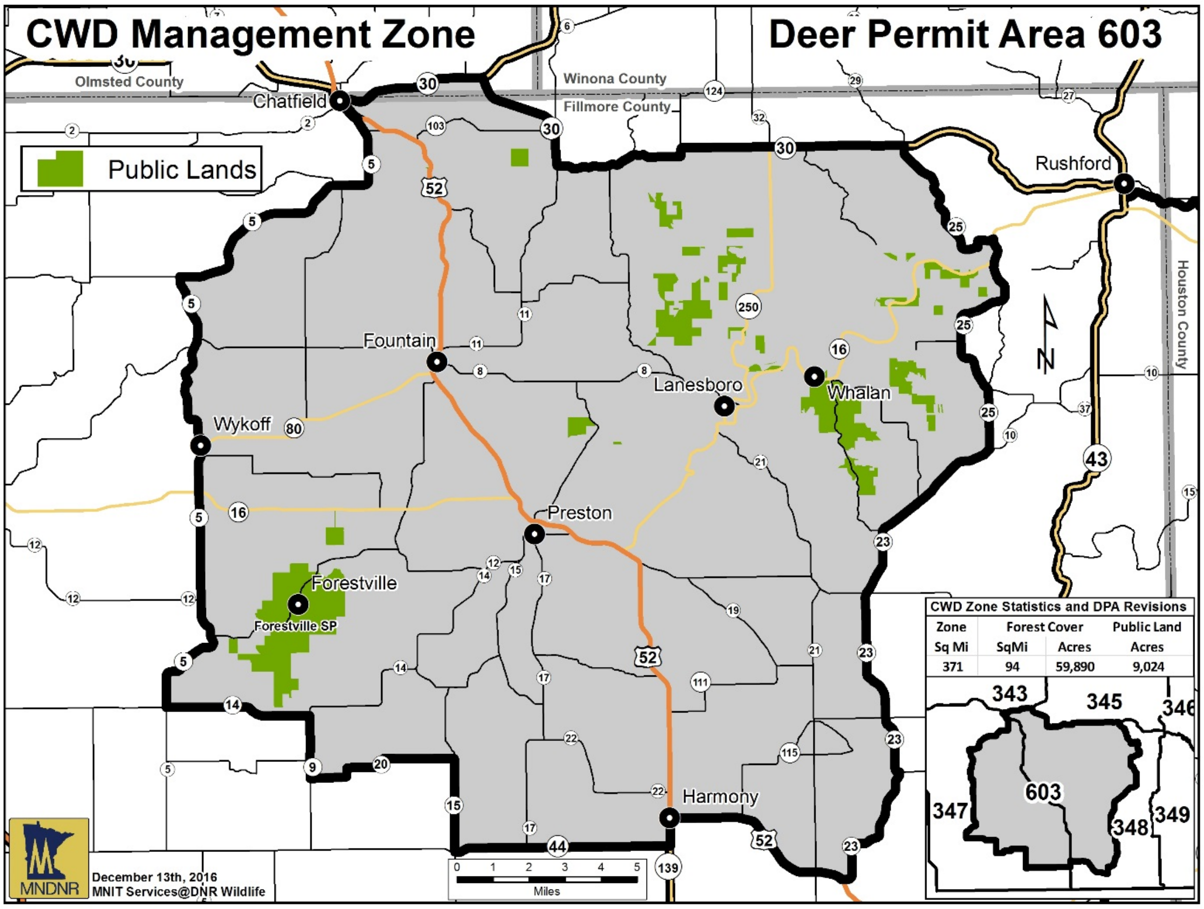


Figure 1. Chronic wasting disease (CWD) management zone, deer permit area (DPA) 603 boundaries in southeast Minnesota winter, 2017.

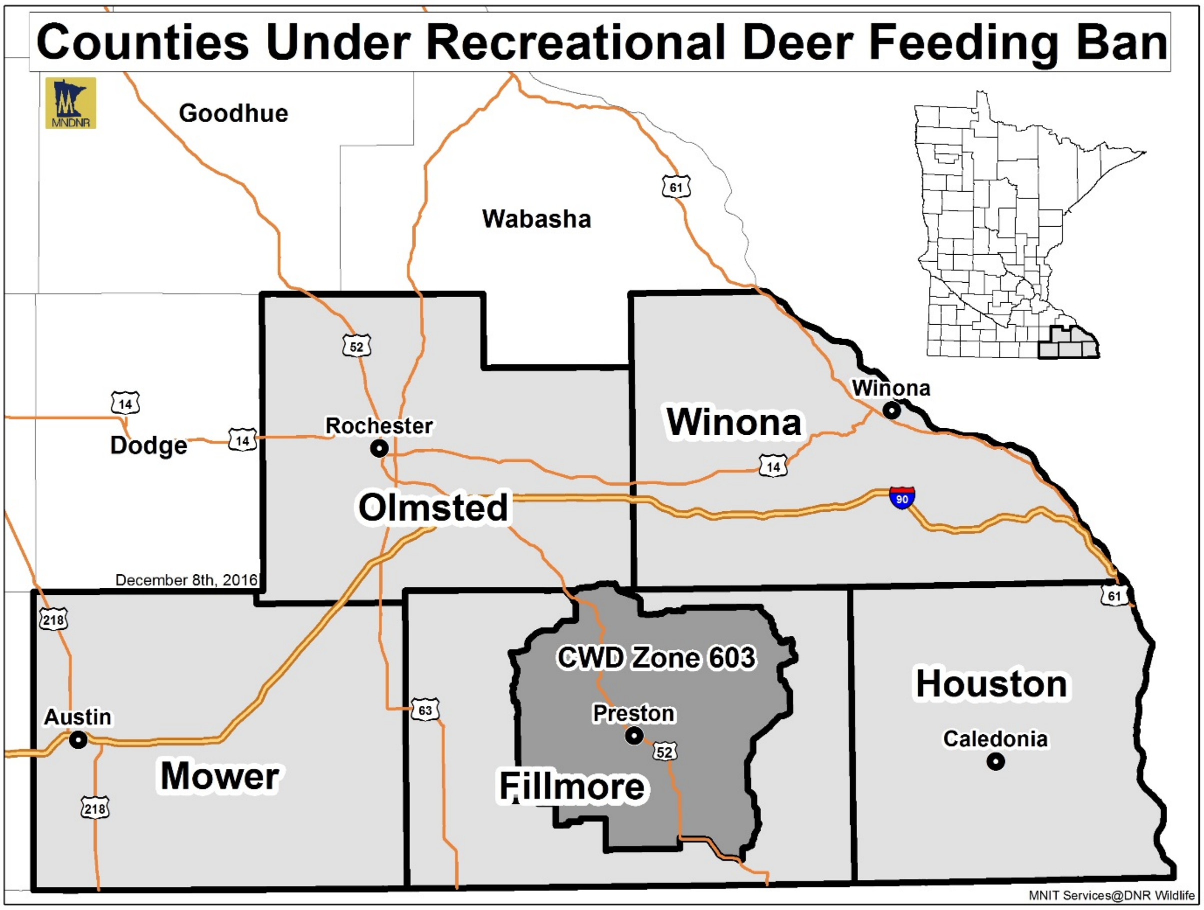


Figure 2. Five-county area in southeastern Minnesota where recreational feeding of wild white-tailed deer was banned in December 31, 2016, following the discovery of chronic wasting disease in Fillmore County.

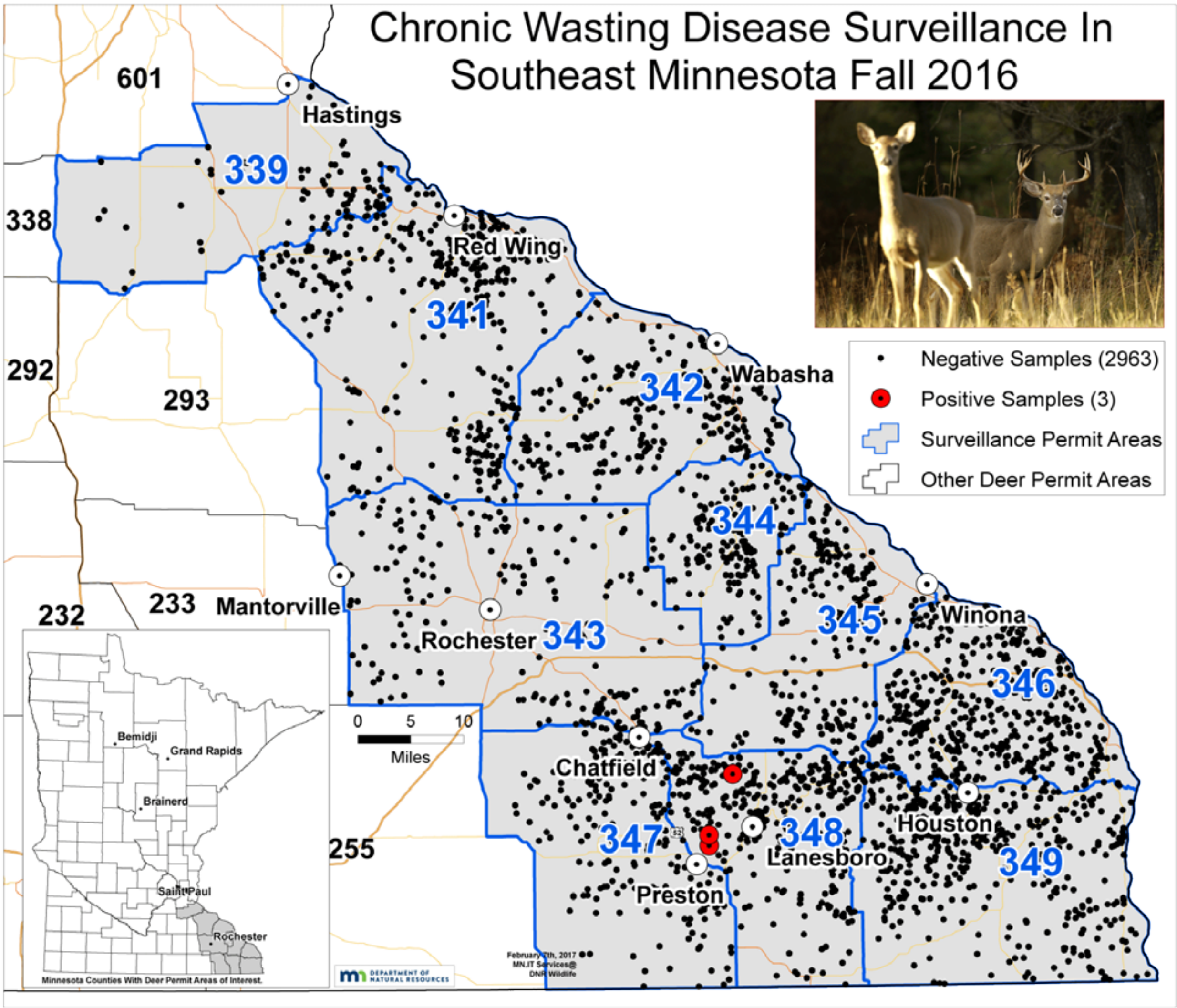


Figure 3. Sampling distribution for all hunter-harvested white-tailed deer (n=2,966) tested for chronic wasting disease (CWD) in southeastern Minnesota, fall 2016. Three tested positive for CWD near Preston, Minnesota.

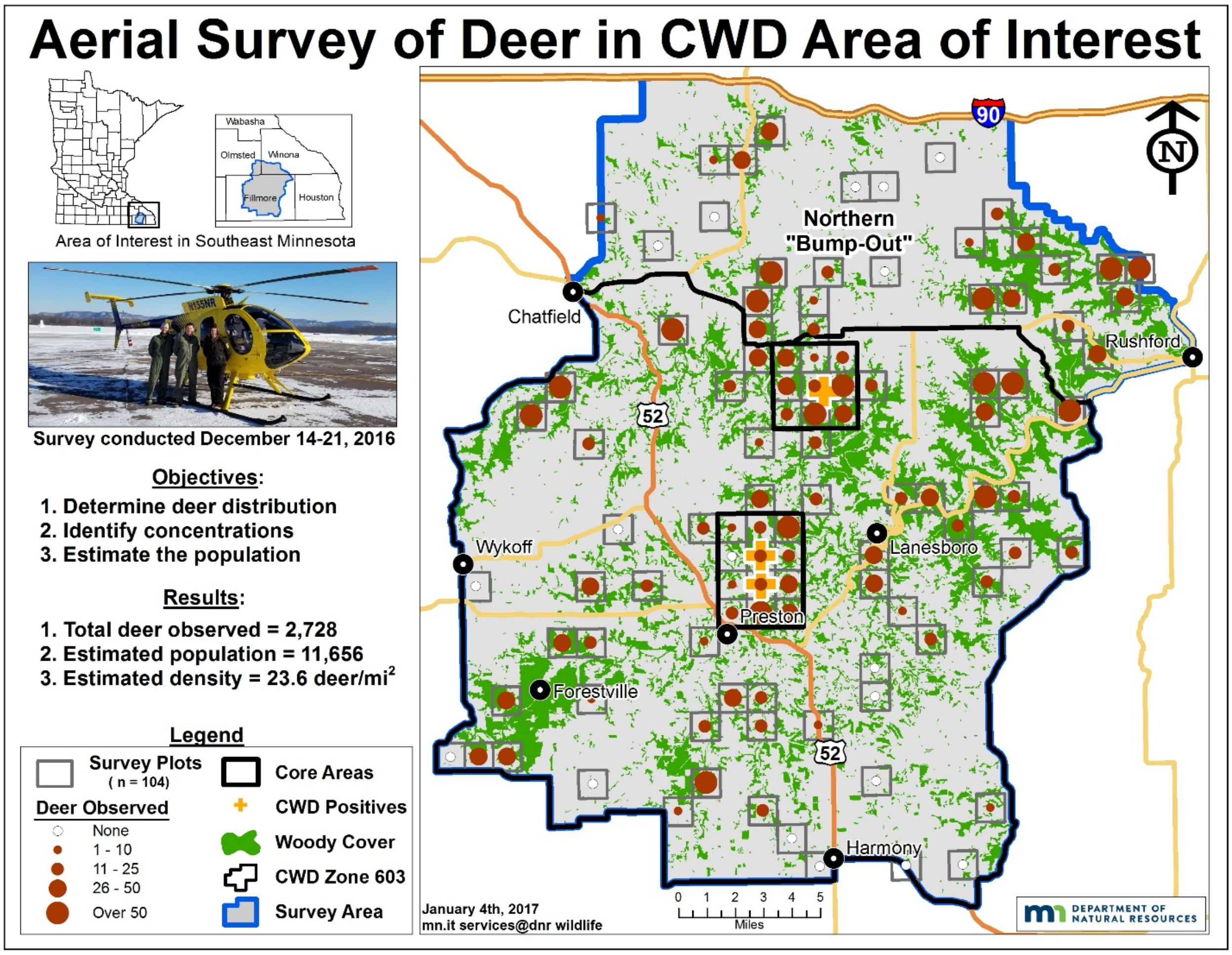


Figure 4. Helicopter, aerial survey results for 1317.5-km^2 (508.7-mi^2) area surrounding the location of the white-tailed deer that tested positive for chronic wasting disease (CWD), southeastern Minnesota, December 2016.

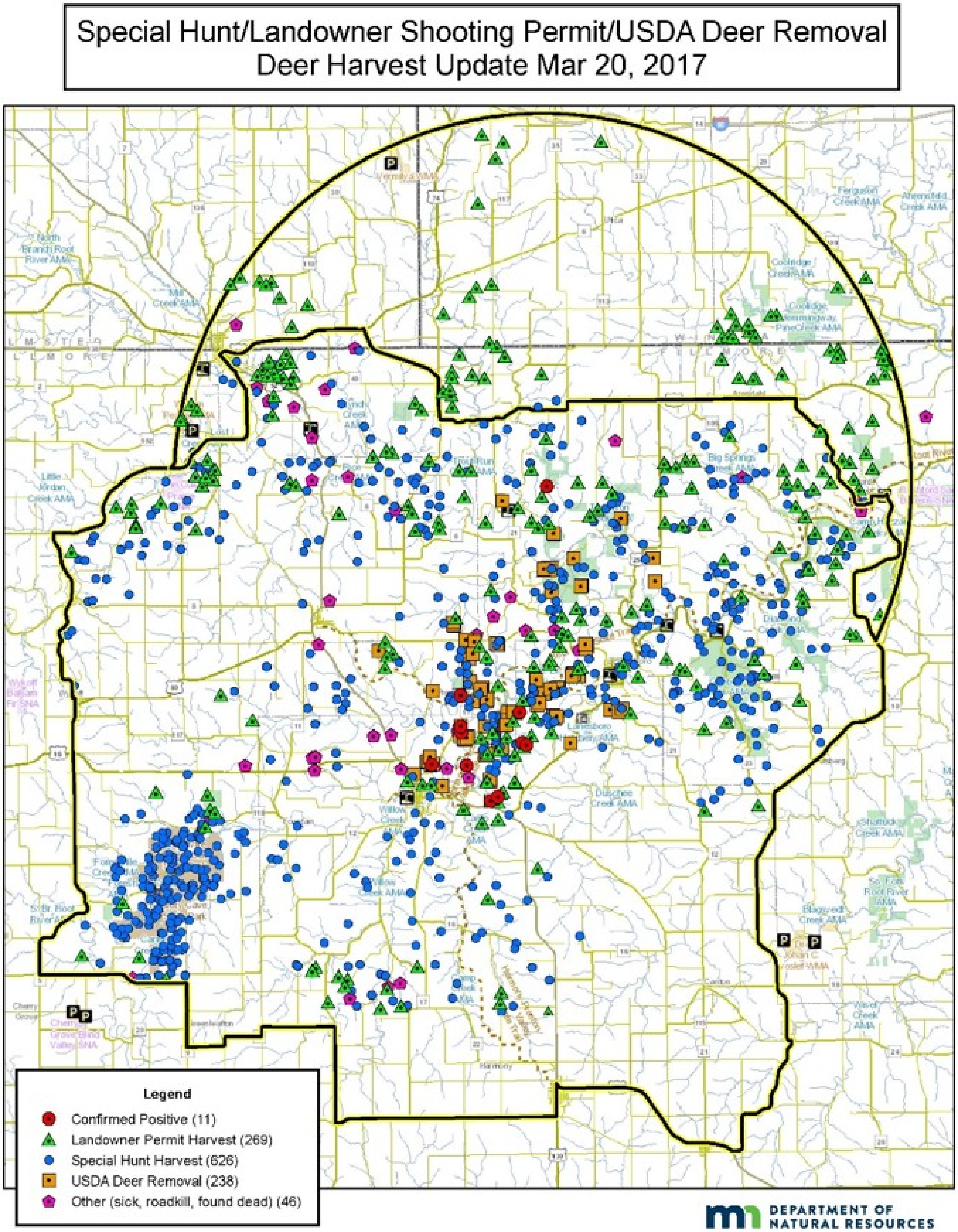
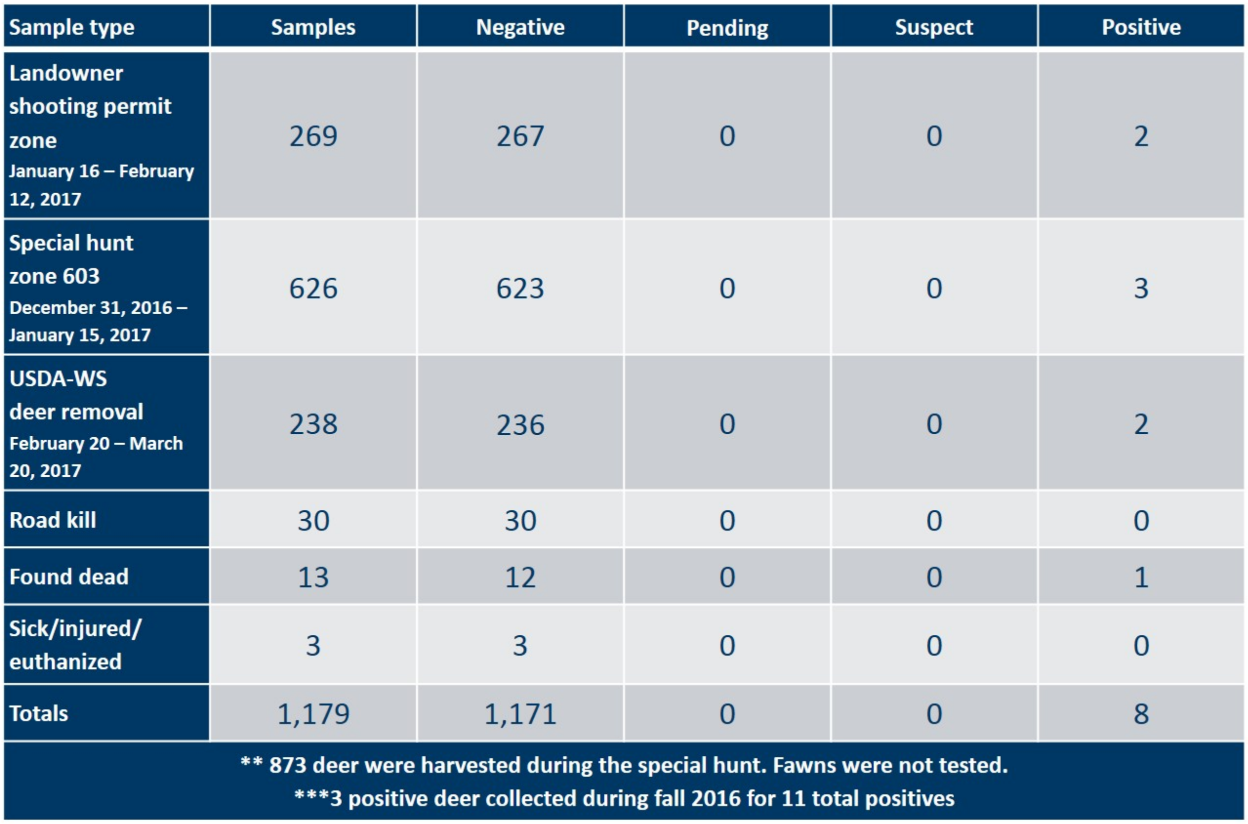


Figure 5. Sampling distribution of deer (*n*=1,179) sampled for chronic wasting disease (CWD) in southeast Minnesota's CWD surveillance area winter, 2017.



Sample type	Samples	Negative	Pending	Suspect	Positive
Landowner shooting permit zone **January 16 – February 12, 2017**	269	267	0	0	2
Special hunt zone 603 **December 31, 2016 – January 15, 2017**	626	623	0	0	3
USDA-WS deer removal **February 20 – March 20, 2017**	238	236	0	0	2
Road kill	30	30	0	0	0
Found dead	13	12	0	0	1
Sick/injured/ euthanized	3	3	0	0	0
Totals	1,179	1,171	0	0	8

**** 873 deer were harvested during the special hunt. Fawns were not tested.**

*****3 positive deer collected during fall 2016 for 11 total positives**

Figure 6. Breakdown by method of total white-tailed deer tested for chronic wasting disease (CWD) by MNDNR in the CWD surveillance area during winter, 2017.

APPLICATION OF A BAYESIAN WEIGHTED SURVEILLANCE APPROACH FOR CHRONIC WASTING DISEASE IN WHITE-TAILED DEER[1]

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ABSTRACT

Surveillance is critical for understanding the emergence and epidemiology of infectious diseases, and weighted surveillance takes advantage of heterogeneity in host disease risk to increase the efficiency of sampling efforts. We apply a Bayesian approach to estimate weights for 16 surveillance classes of white-tailed deer in Wisconsin, USA, relative to yearling hunter-harvested male deer. We use these weights to design a surveillance program for detecting CWD in white-tailed deer at Shenandoah National Park (SHEN) in Virginia, USA. Generally, apparent infection hazard increased with age and was greater in males. Clinical suspect deer were the highest risk class with weight estimates of 33.33 and 9.09, for community reported and hunter reported suspect deer, respectively, while fawns were the lowest risk class with an estimated weight of 0.001.

We used Wisconsin derived surveillance class weights to determine sampling effort required to detect a CWD-positive case in SHEN if prevalence in male yearlings ≥0.025. The sampling effort required to detect CWD at the 2.5% level in our reference class, male yearling deer, was 37–91 adult deer, depending on the ratio of bucks to does in the surveillance stream. We collected rectal biopsies from 49 and 21 adult female and male deer, respectively, and 10 additional samples from vehicle-killed deer. All samples tested negative and demonstrated with 95% probability that CWD prevalence in the reference population (yearling males) was between 0.0 to 3.6%. Our approach allows managers to estimate relative surveillance weights for different host classes and quantify limits of disease detection in real time, resulting in financial, resource, and personnel savings for agencies tasked with performing wildlife disease surveillance. Additionally, it provides a rigorous means of estimating disease prevalence limits when the disease/pathogen is not detected, and can be applied to other ecological questions of interest where data is available on heterogeneous probability of risk or occurrence.

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DETERMINING CAUSE-SPECIFIC MORTALITY OF ADULT MOOSE IN NORTHEAST MINNESOTA, FEBRUARY 2013 – JULY 2017

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SUMMARY OF FINDINGS

The primary goal of this study is to improve our understanding of the causes of non-hunting mortality in northeastern Minnesota’s declining moose (*Alces alces*) population. Our goal is to respond to potential mortalities within 24 hours of death, prior to decomposition of tissues, and determine proximate cause of death and contributing factors. From 2013–2015, we captured and radio collared a total of 173 adult moose (123 females, 50 males). Mean age at capture was 6.1 (±0.3) years of age; range was 1 to 16 years. A total of 57 collared moose (40 females, 17 males) have died, excluding 12 capture-related mortalities that will be censored from subsequent survival analyses. Annual mortality rates were 19%, 12%, 15%, and 14% in 2013, 2014, 2015, and 2016, respectively. The mortality rate from January–July 2017 was 9%. Overall proximate causes of death included: wolf predation (n=18, 32%), parasitic infections (n=17, 30%), bacterial infections (n=12, 21%), accidents (n=2, 3%), hunter-harvest (n=1, 2%), calving complication (dystocia) (n=1, 2%), and undetermined health issues (n=6, 10%). At least 40% of the moose killed by wolves had other serious health issues that may have predisposed them to predation. *Parelaphostrongylus tenuis* was confirmed in 23% of all moose mortalities as either the direct cause (n=7, 12%) or a contributing factor (n=6, 11%) in their deaths. Whole carcasses were retrieved for 22 (39%) of mortalities, with field necropsies performed on the remaining 35 (61%) moose. Response times from initial mortality notification (e.g., text message or email) to a team in the field at the death site were ≤24 hours in 38 cases (67%), between 24 and 48 hours in 11 cases (19%), and >48 hours in 8 cases (14%). There are currently 27 moose remaining in the study with active collars that are still being monitored for survival. Unfortunately, collar failure rates have been high (causes unknown, assumed to be battery or transmission failures), with 21 collars recovered from live moose via remote blow-off mechanisms and another 53 collars unaccounted for; thus, we are not certain of their status (live or dead).

INTRODUCTION

Until recently, 2 geographically distinct moose (*Alces alces*) populations occurred in Minnesota (MN), one in the northwestern (NW) and the other in the northeastern (NE) part of the state. Since the mid-1980s the NW population has decreased from an estimated 4,000 to less than 100 moose, and since 2006 the NE population has declined 58% from an estimated 8,840 to 3,710 moose (DelGiudice 2017). However, there is some evidence that the moose population in the NE may be stabilizing over the last 5 years (2012-2017) at approximately 4,000 animals. Mean annual mortality rates of adults have been similarly high (21%) in both regions (Murray et al. 2006, Lenarz et al. 2009). Parasites, including liver flukes (*Fascioloides magna*) and brainworm (*Parelaphostrongylus tenius*) and other non-specific health-related issues have been documented in the majority of moose deaths through these past research efforts (Wünschmann et al. 2015). Climate change has also been implicated as an underlying factor in both population declines.

There were inverse relationships between warming ambient temperatures and decreasing survival of adult moose (Murray et al. 2006; Lenarz et al. 2010). Trends in temperature and precipitation patterns are likely to increase in intensity over the next century. If moose are unable to sufficiently thermoregulate above certain ambient temperature thresholds, we might expect to see increased body temperatures and energy expenditures required to stay cool, which over time could have negative consequences for body condition, reproduction, and survival. Currently, no data exist to support the direct adverse effects of ambient temperature on the physiology, survival, or reproduction of free-ranging moose.

This study will determine cause-specific mortality by deploying satellite-linked Global Positioning System (GPS) collars on moose in NE MN and by preparing an extensive network of responders highly trained in conducting field necropsies. Moose mortalities will be thoroughly investigated within 24 hours of death to identify the proximate cause of mortality and to examine the influence of potential contributing factors. Once causes of death and major influential factors are identified, appropriate management actions may be taken to address the population decline. Our main objectives are to 1) determine causes of non-hunting mortality (i.e., identify specific disease and parasite agents) and assess the role nutrition plays as a contributing factor; and 2) investigate how ambient temperatures relate to moose survival in NE MN by applying an unprecedented field approach and comprehensive data collection methods.

Recently, a minimally invasive telemetry system for ruminants, called a mortality implant transmitter (MIT), has been developed to allow nearly continuous monitoring of body temperature with a battery lifetime of approximately 2 years. Using these MITs and GPS collars on adult moose in this study will allow us to correlate ambient temperature with their physiology, behavior (habitat use and activity), and fitness (survival and reproduction). This study will be the first to examine these relationships in a way that includes monitoring body temperature. The results of this study will be critical to an improved understanding of if, when, and how moose are able to successfully modulate their internal body temperature. Such an understanding should prove valuable in the formulation of future population and habitat management strategies and activities.

METHODS

Moose (*n*=173; 123 females, 50 males) were captured within the 3,732.8 km^2 study area located between 47°12’N and 47°95’N latitude and 90°33’W and 91°72’W in NE MN (Figure 1) from 2013 to 2015, as described previously (Butler et al. 2013; Carstensen et al. 2014, 2015, 2016). All moose were fitted with GPS-Iridium satellite collars (Vectronic Aerospace GmbH; Berlin, Germany). Mortality implant transmitters (Vectronic Aerospace GmbH) were placed orally into a subset of the captured moose and provided immediate notification of mortality and recorded internal body temperature. External temperature loggers (Hobo TidbitV2; Onset Corporation, Bourne, MA) were attached to the GPS collars and were programmed to collect ambient temperature every 60 minutes. Additional ambient temperature loggers (black globes and white funnels) were placed in 7 open habitat sites throughout the study area. Data from the temperature loggers will be used along with data from 12 National Oceanic and Atmospheric Association and Remote Automatic Weather Stations in NE MN to determine the best ambient temperature predictor for moose with MITs in this study.

Moose mortality response teams have 8 primary team leaders that have undergone extensive necropsy training, and they are supported by about 20 secondary and tertiary team members (including MNDNR, tribal, academic, US Forest Service, and other personnel) available upon request. Every effort is made to respond to a moose mortality event with 24 hours of notification and 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. If a moose was found to be alive, but obviously ill, it was euthanized (via gunshot to the neck). If carcass extraction was not possible, a thorough and complete field necropsy was performed, guided by an established protocol. Samples were submitted to the UMN VDL for diagnostic evaluation (Carstensen et al. 2014, 2015, 2016).

Moose age was determined by cementum annuli at time of capture and we used one-way analysis of variance to compare age among years. A two-sample T-test was used to compare the mean age of moose killed by predators to those that died of health-related causes. Dead moose were categorized by age as young (≤3 years), prime (4–8 years), and old (≥9 years) and chi-square analyses was used to compare age cohorts by predator and health-related causes of death. Annual (January-December) survival rates were estimated using Kaplan-Meier to allow for staggered entry design.

RESULTS AND DISCUSSION

Annual Survival and Cause-Specific Mortality

From 2013–2015 a total of 173 adult moose (123 females, 50 males) were captured and radio collared. Mean age at capture was 6.1 (±0.3, n=163) years for all moose; range was 1 to 16 years. Age of moose at capture was similar [F-$stat$=1.65, p=0.19] among years (6.0 years in 2013, n=101; 5.8 years in 2014, n=32; and 7.2 years in 2015, n=30). Annual (January–December) survival rate was 81%, 88%, 85% and 86% in 2013 through 2016, respectively; 91% of moose have survived from January–July 2016 (Figure 2). A total of 57 collared moose (40 females, 17 males) have died since this study began; which excludes 12 capture-related mortalities that are censored from subsequent survival analyses. Overall proximate causes of death included: wolf predation (n=18, 32%), parasitic infections (n=17, 30%), bacterial infections (n=12, 21%), accidents (n=2, 3%), hunter-harvest (n=1, 2%), calving complication (dystocia) (n=1, 2%), and undetermined health issues (n=6, 10%; Figure 3). Health-related causes were attributed to 68% of total deaths, with the remaining 32% being predator-related.

Eight (44%) of the wolf-killed moose had significant health conditions that likely predisposed them to predation, including encephalitis and meningitis in the brain, *P. tenuis* infections, winter tick (*Dermacentor albipictus*) infestations, calving, and pneumonia in the lungs (Figure 4a). Unfortunately, diagnostics were limited in 10 of the wolf-killed moose due to the degree of carcass consumption prior to the mortality team's arrival to the scene. It is possible that health issues may have compromised some of these moose as well.

Parasitic infections were the second leading cause of moose deaths (Figure 4b). *P. tenuis* directly led to the death of 7 moose in this study; however, this parasite was also implicated in 5 wolf-caused deaths and 1 bacterial infection. Overall 23% of the moose that died during this study have been impacted by *P. tenuis* and this is likely an underestimate, as not all dead moose could be evaluated for this parasite. Winter tick infestations were primarily seen in spring 2013 (attributed to 3 moose deaths), as the severe and prolonged winters in 2012-13 and 2013-14 likely reduced tick survival. However, the past 3 winters have been extremely mild and it's likely that winter tick loads have recently increased on moose. In spring 2016, one moose in the study died from winter ticks; however, significant tick infestations were observed in other moose as well. We had expected that moose surviving into spring 2017 would experience a significant winter tick burden and this would result in an increase in tick-related mortalities; however, none of the collared moose died from winter tick burdens in 2017 but the sample size has markedly declined to only 41 animals left to monitor at the beginning of this year. Most moose in this study had livers that were damaged by liver flukes (*F. magna*), the severity of which varied from mild cases to severe infections that directly caused the death of 3 moose. Similarly, the majority of moose in this study had hydatid cysts in the lungs or liver, caused by *Echinococcosis granulosis*, but only 2 moose had severe enough infections with this parasite to cause mortality. We also observed one moose with an extensive cysticercus (*Taenia krabbei*) infection throughout the body, including the heart, which resulted in death due to reduced cardiac function.

Bacterial infections were the third leading cause of moose deaths (Figure 4c). Four moose were attacked by a wolf or wolves and survived the initial encounter, but the wounds became infected and led to their death days to several weeks later. Prior to this study, scant evidence in the literature points to secondary bacterial infections caused by a predator attack as a major cause of moose mortality. Other trauma, including one case of conspecific fighting of antlered males, resulted in puncture wounds that provided a route for bacteria to enter the body and cause systemic infection and septicemia. The exact circumstances that led to some of these trauma-induced injuries were unknown.

The remainder of moose deaths were caused by accidents (1 vehicle collision and 1 fall through the ice), hunting (1 moose was legally harvested by a tribal member), calving complications or dystocia (1 moose had twin calves stuck in the birth canal while being expelled simultaneously), and undetermined health-related deaths (6 moose).

There are currently 27 moose remaining in the study with active collars. Unfortunately, collar failure rates have been high (causes unknown, assumed to be battery or transmission failures), with 21 collars recovered from live moose via remote blow-off mechanisms and another 53 collars unaccounted for; thus, we are not certain of their status (live or dead). Three moose had their collars slip off their necks, presumably due to an excessively loose fit, and were recovered in the field.

Timing of Mortalities

Timing of these mortalities suggest that most deaths occur in spring (44%, March–May); however, moose died in all seasons (winter 17%, summer 23%, and fall 16%; Figure 5). Health-related mortalities occurred during all months of the study; however, there were no wolf-related deaths in October through January (Figure 6).

Mean age of moose (*n*=55, excludes 2 moose with age results pending) at death was 8.4 years (±0.5 year); range was 1 to 15 years old. Mean age of moose that died from health-related causes (*n*=35; excluding 2 moose with accidental deaths and 1 moose harvested by hunters) was 8.1 years (±0.6 year), similar [*T-stat*=2.0, *p*=0.7] to those (*n*=18) that died of wolf-related causes (8.6 ±1.0 years). Interestingly, both health and predator-related causes of death impacted nearly every age cohort in this study (Figure 7), yet there was some evidence (X^2 *stat*= 45.0, *p*=0.08) to support that wolves were more selective for the young (≤3 years of age) or old (≥9 years of age) cohorts and more prime-aged moose were dying of health-related issues.

Mortality Response Times

Whole carcasses were retrieved for 22 (39%) mortalities, with field necropsies performed on the remaining 35 (61%) moose. Response times from initial mortality notification (e.g., text message or email) to a team in the field at the death site were ≤24 hours in 38 cases (67%), between 24 and 48 hours in 11 cases (19%), and >48 hours in 8 cases (14%). Delays in mortality responses >24 hours have been due to collar failures and wolves actively feeding on the moose carcass and preventing the collar from sending a mortality alert.

Mortality Implant Transmitters

We successfully deployed 63 MITs in moose during this study. Deployment failures occurred in 20 moose, where the MIT was not fully swallowed and regurgitated (*n*=19) or the bolus failed to reach the rumen (*n*=1). A revised and improved MIT deployment technique was implemented during the final year of capture of this project, and improved success rates from 73% (43 of 59 attempts, 2013-2014) to 85% (20 of 23 attempts, 2015) (Minicucci et al. 2017). To date, 20 moose with working MITs have died in the study and their body temperature data was collected; however, 4 of these moose were capture-related mortalities and their data will be censored from further analyses. Another 11 MIT datasets have been recovered from remotely blown collars (n=9) and slipped collars (n=2); however, 17 moose with MITs have collars that have malfunctioned and their survival status is unknown; it's unlikely these data will be ever be recovered. Currently, 15 of the 27 moose remaining in the study with functioning collars have MITs.

MNDNR collaborated with the Alaska Department of Game and Fish to conduct a MIT calibration project at the Moose Research Center in Kenai, Alaska from 2014–2015. The MIT was shown to be a highly accurate measurement of internal body temperature in moose (Herberg et. al 2016). After removing water intake-induced low temperatures, MITs recorded internal body temperatures only 0.03 °C (95% CI -0.57-0.55) lower than vaginal implant transmitters (VITs) and were therefore considered highly accurate. We have begun some preliminary analyses using MIT data recovered to date. MIT data from 25 wild moose (15 females, 10 males) in this study were recovered either at death (*n*=16), from collars that slipped or were remotely released (*n*=6), or from recollaring events (*n*=3). MIT values were obtained for an average of 400 days for these moose (range 57 to 941 days). Out of these 25 animals, 23 had between 0.19% and 11.25% of internal temperatures considered above normal (i.e. ≥ 39.2°C). The percent MIT temperatures that were above normal varied seasonally, ranging from 0.63-25.07% (μ=8.18%), 0.04-13.46% (μ=1.92%), 0-2.41% (μ=0.64%), and 0-2.39% (μ=0.15%) in summer, fall, spring, and winter, respectively. There may be behavioral tradeoffs moose have to make to seek dense cover to "cool-off", especially in the summer, to the detriment of time spent in high quality forage habitat. Further analyses, focusing on the summer months, will incorporate the environmental conditions (habitat & ambient temperatures) moose experienced, as well as movement patterns and activity levels they exhibited, in the time periods before and after periods of abnormally high internal body temperatures, in an effort to unveil some of these tradeoffs.

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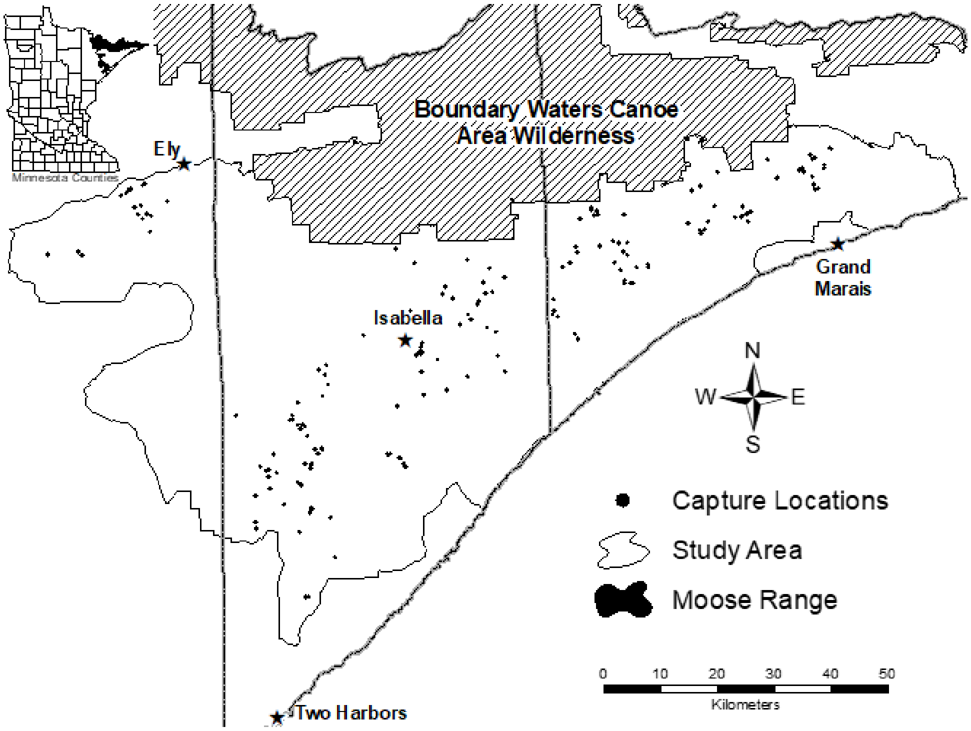

Figure 1. Study area in northeast Minnesota where 179 moose (included 6 recaptures) have been captured and radio-collared (2013–2015) to study cause-specific mortality.

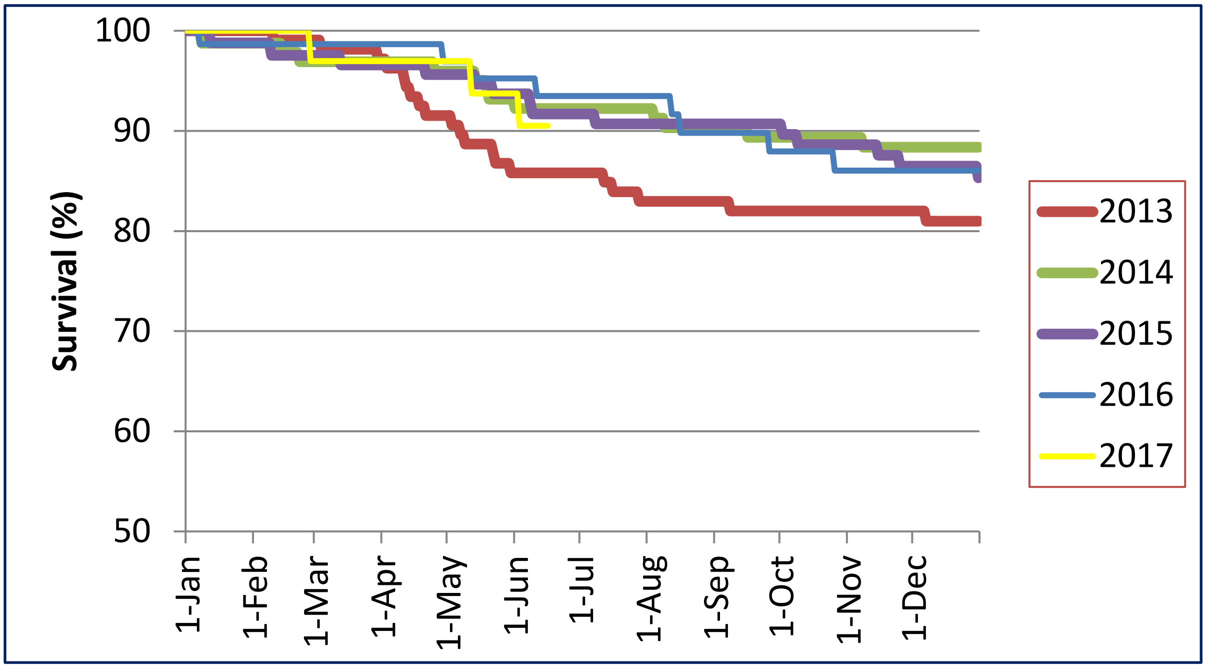

Figure 2. Annual survival of radio-collared, adult moose (*n*=173) captured from 2013-2017, northeast Minnesota.

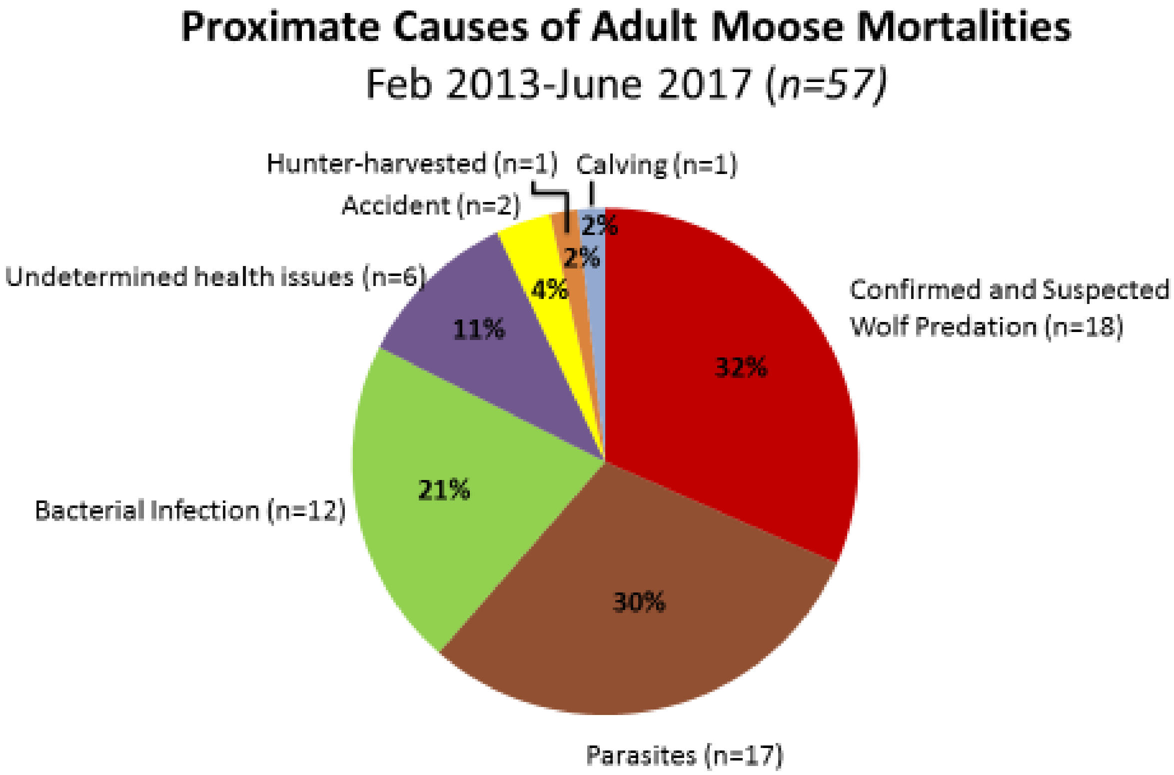

Figure 3. Cause-specific mortality of radio-collared, adult moose (*n*=57) from February 2013-July 2017, northeast Minnesota.

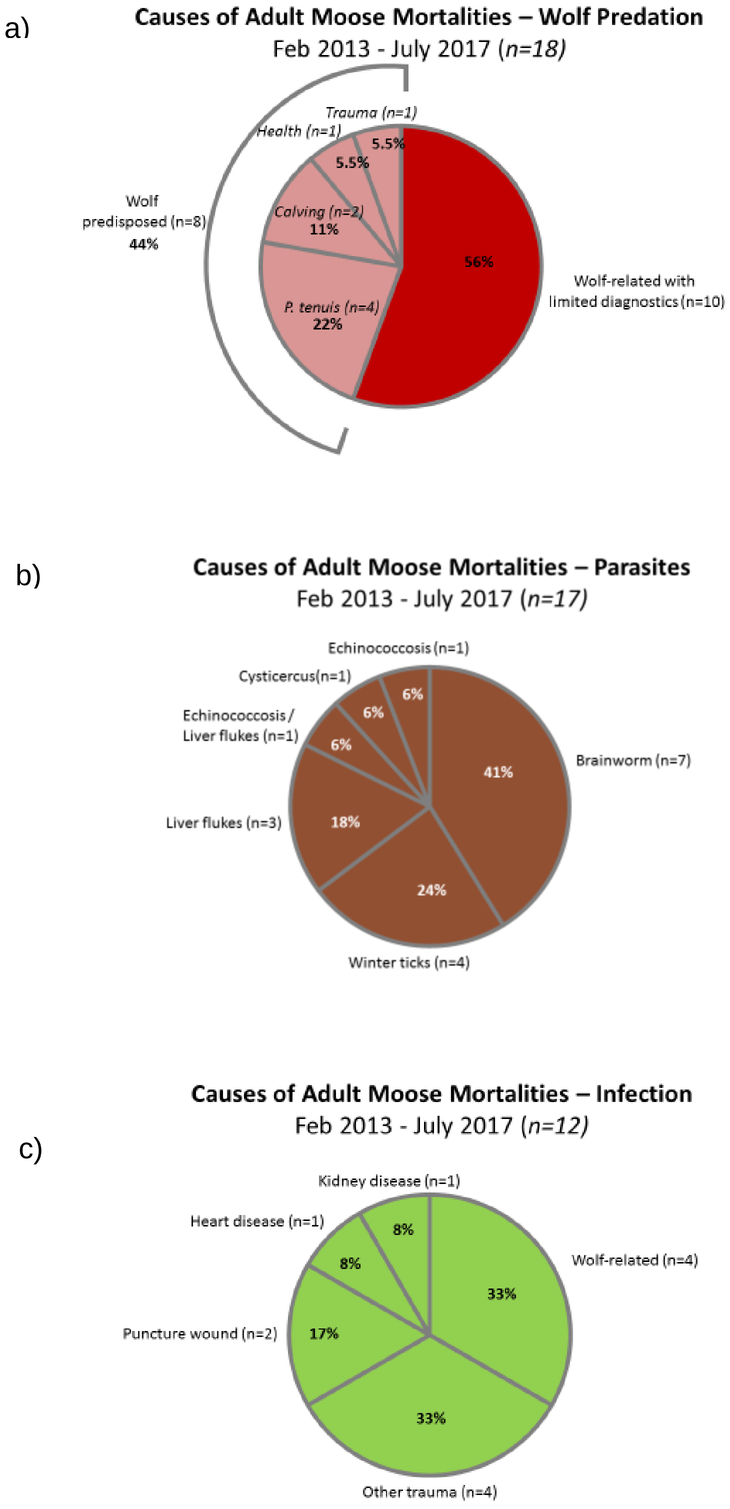

Figure 4. Breakdown of adult moose mortalities caused by wolf predation (a), parasites (b), and bacterial infections (c), Feb 2013-July 2017, northeast Minnesota.

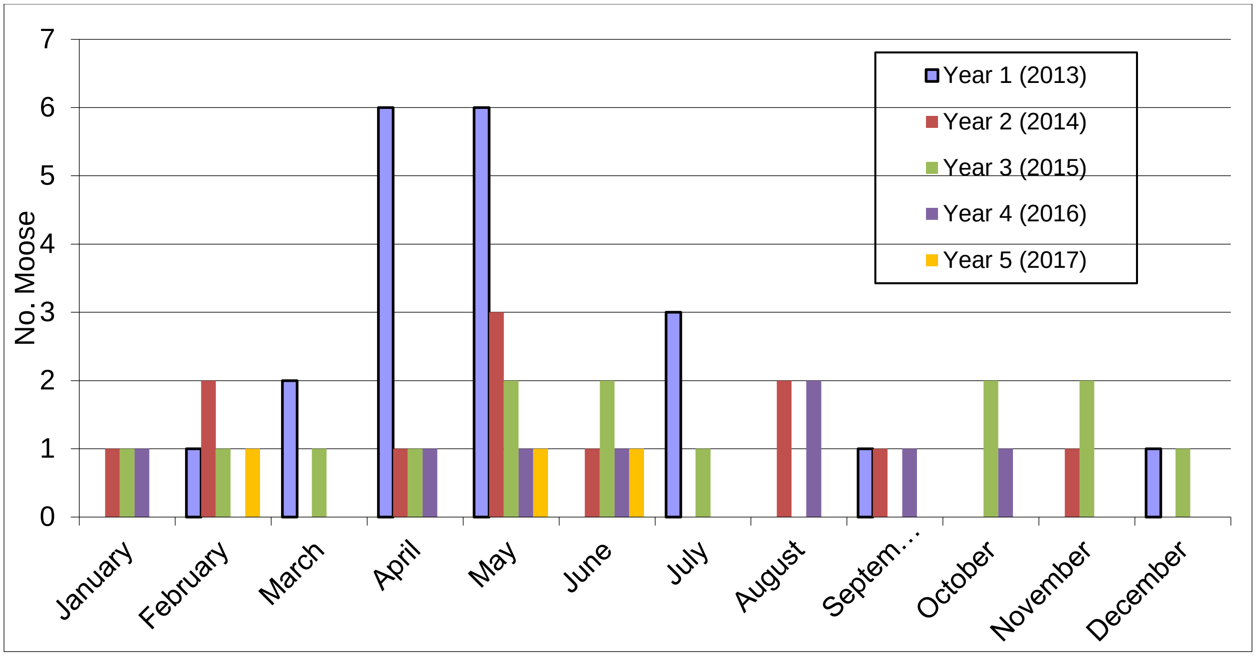

Figure 5. Timing of mortalities for radio-collared, adult moose (*n*=57) from January 2013-July 2017, northeast Minnesota.

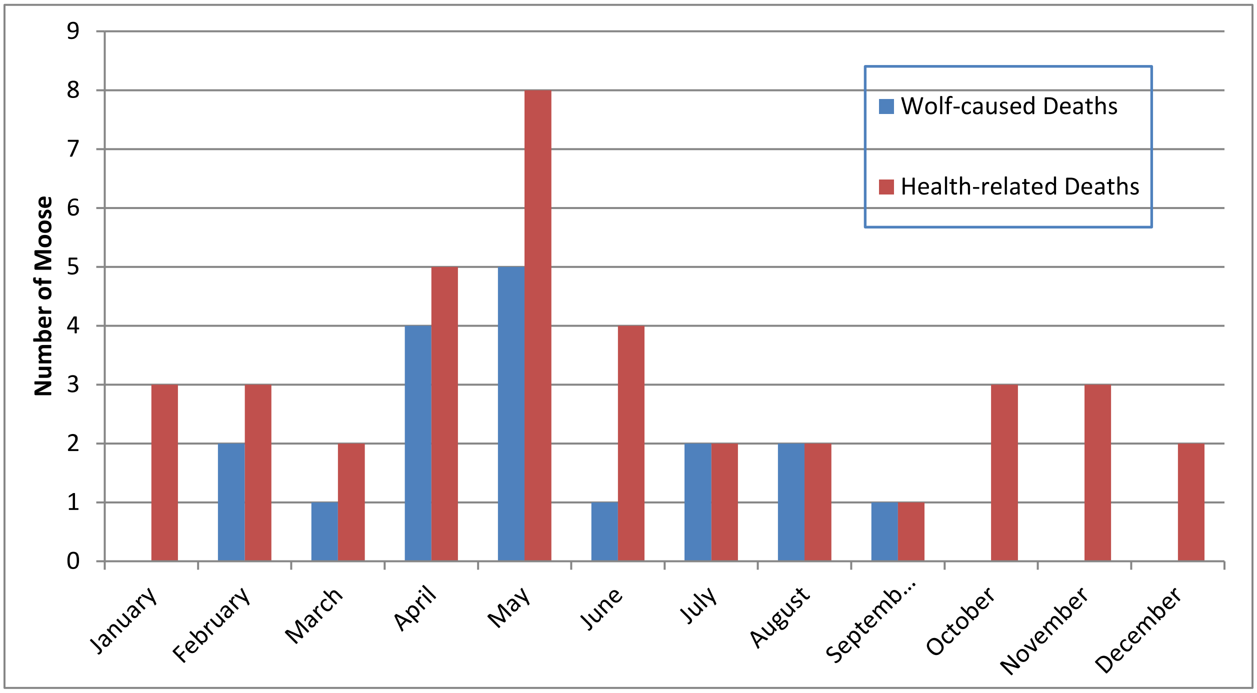

Figure 6. Timing of wolf-caused (*n*=18) and health-related (*n*=38) moose mortalities, 2013-2017, northeast Minnesota.

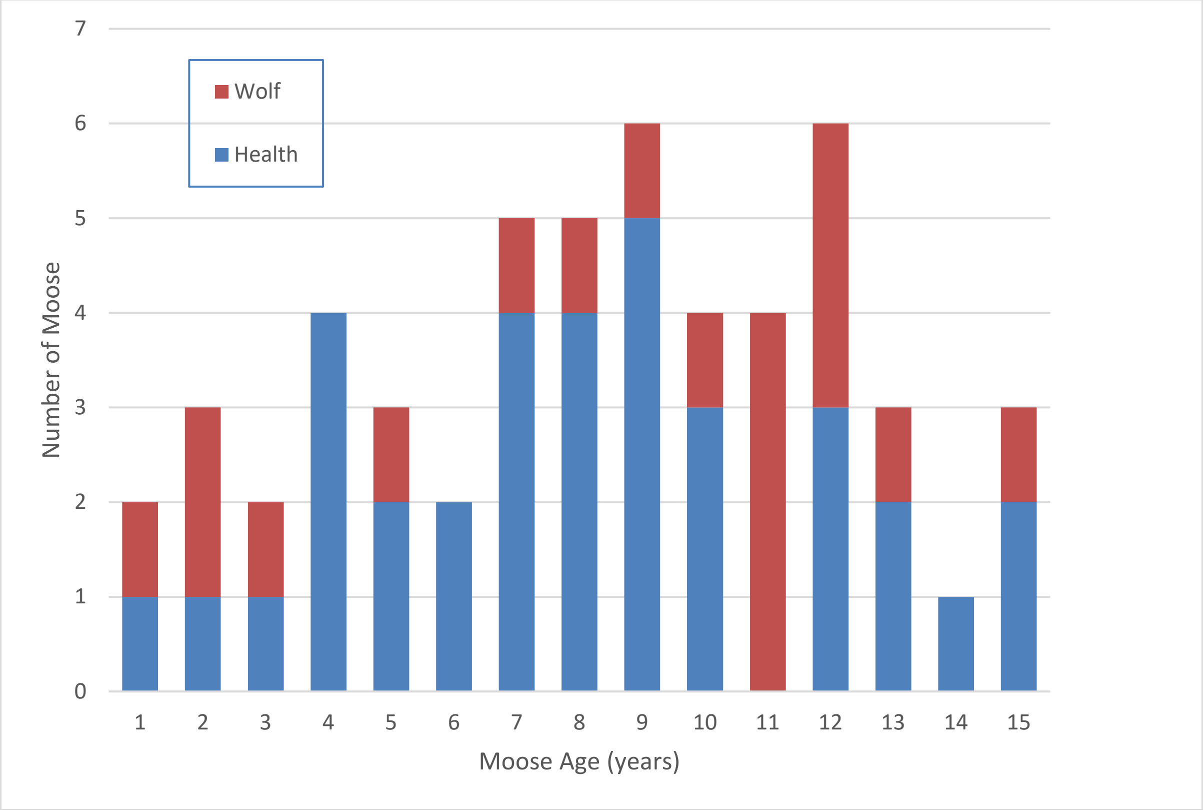

Figure 7. Known ages of radio-collared, adult moose (*n*=53) that died from health-related (blue) or wolf-related (red) causes (2013-2017), northeast Minnesota, 2013-2017.

DEPARTMENT OF
NATURAL RESOURCES

ARE MOOSE GETTING WARM, AND HOW DO THEY RESPOND BEHAVIORALLY? VALIDATION OF AN APPROACH FOR REMOTELY MONITORING MOOSE BEHAVIORS

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SUMMARY OF FINDINGS

As part of an ongoing cause-specific mortality study of adult moose (Alces alces) in northeast Minnesota, and to better understand behavioral and physiological responses of moose to increasing ambient temperature, the Minnesota Department of Natural Resources (MNDNR) deployed Global Positioning System (GPS) collars on a total of 173 individuals from 2013-2015. This provided information on broad-scale animal movements and habitat use, but also fine-scale activity patterns using data from dual-axis activity sensors contained in the collars. In the portion of the research we present here, we wanted to test the efficacy of using dual-axis activity sensors for remotely predicting behavioral states of moose. Utilizing 10 captive female moose (>2 years old) at the Moose Research Center in Kenai, Alaska, fit with the same GPS collars as wild moose in Minnesota, we collected a total of 384 hours of behavioral observations during 4, 2-week windows distributed across seasons to evaluate if we can predict behavioral states using fine-scale activity data. Our results demonstrate that combining biotelemetry devices with modern statistical approaches allows researchers to examine the physiological and behavioral responses of moose to increasing ambient temperatures and changing landscapes, and at finer temporal and spatial scales than previously possible. Ultimately, results from this research will be applied to the data we obtained from Minnesota moose to better understand moose behavioral responses to increasing body temperatures.

INTRODUCTION

Moose are experiencing lower survival rates at the southern edge of their range compared to core geographic range (Dodge et al. 2004; Murray et al. 2006; Maskey 2008; Lenarz et al. 2010). In Minnesota, moose in the northwestern portion of the state are all but extirpated (Murray et al. 2006), and the northeastern population has declined from an estimated 8,840 in 2006 to 3,710 in 2016 – a reduction of 55% (DelGuidice 2017). Although the ultimate driver of the northeast population decline remains unknown, recent research has demonstrated that the majority of moose mortalities can be attributed to health-related causes (Murray et al. 2006; Carstensen et al. 2014). Moose are known to be physiologically sensitive to heat (Renecker and Hudson 1986; Renecker and Hudson 1989; McCann et al. 2013) and to alter the habitat types they use when ambient temperature increases (Schwab and Pitt 1991; van Beest et al. 2012; Street et al. 2015; Street et al. 2016) by selecting for habitats that act as thermal refuges (Dussault et al. 2004); i.e., to the potential detriment of spending less time in optimal foraging habitat (Street et al. 2016). Understanding not only where moose are in the landscape, but what they are doing in different areas of their range (e.g., resting, moving, foraging), is a critical step towards developing forest

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management practices that may benefit moose (e.g., enhance cover but also improves forage quality).

Between 2013-2015, the MNDNR deployed GPS collars equipped with dual-axis activity sensors 173 free-ranging adult moose in an effort to gain a better understanding of moose behavioral and physiological responses to ambient temperatures and habitat, among others (Carstensen et al. 2014). The activity sensors, besides recording an animal's geographic location, detect and record changes in neck movements as a measure of fine-scale activity (Ungar 2005); this provides a unique opportunity to understand and remotely predict behavioral states in free-ranging animals (Löttker et al. 2009, Ungar et al. 2010, Roberts et al. 2016). Few studies, however, have taken advantage of this technology in part due to a poor understanding of how activity sensor measurements correlate with specific behaviors. Understanding fine- and broad-scale spatial and temporal patterns in activity and habitat use has direct applications for the management and conservation of imperiled species (Gervasi et al. 2006).

The goal of this study was to develop an approach, based on observations of animals in a captive setting, to predict the proportion of time moose spend in different behavioral states over a given time period using activity sensor data. The ability to predict behavioral states from GPS-collars equipped with dual-axis activity sensors may offer insights into how moose behavior changes in response to its environment, and has direct applications to the GPS- and activity-collar data collected as part of the ongoing project of moose in Minnesota. Our specific objectives included:

1) Determine if dual-axis activity sensors can accurately classify behavioral states in moose,

2) Develop a predictive model that can be used to remotely infer behavioral states, and

3) Examine the potential for using remotely predicted behavioral states to investigate behavioral responses of moose to time of day, and changes in habitat and ambient temperatures.

STUDY AREA

The study was conducted at the Kenai Moose Research Center (MRC) located on the Kenai Peninsula, Alaska (Figure 1). The MRC, a 970-hectare captive facility operated by the Alaska Department of Fish and Game, was built in the 1960s to study moose's responses to their environment (Hundertmark et al. 2000). All moose in this study were maintained in outdoor enclosures approximately 240 ha in size and encompassed a mix of habitat types (Thompson et al. 2017). The moose were maintained at densities of 4-8 adult females per enclosure (30-60 ha/moose), depending on the time of year and conspecific aggression. Moose naturally foraged within the enclosures and were only supplemented during times of low nutritional condition, handling (e.g., weighing), or during specific studies. Moose had access to water from lakes and wetlands throughout the enclosures. To supplement water intake during the warm season (late spring to early fall) cattle troughs were available in enclosures with fewer wetlands. While predation risks were low, encounters with brown bears (*Ursus arctos*), black bears (*Ursus americanus*), and wolves (*Canis lupus*) occur sporadically within the enclosures (Dan Thompson, pers comm.).

METHODS

Moose Handling

A total of 10 moose were immobilized during 4 routine immobilization periods at the MRC (December, April, June, and September) following the procedure outlined by Thompson et al. (2017). Each of the 10 moose was fitted with a uniquely marked (i.e., color taped) Vectronic GPS collars (GPS Plus Iridium; Vectronic Aerospace GmbH; Berlin Germany) to facilitate individual recognition by the observers. The GPS collars recorded location data on board in 30-minute intervals and all data were downloaded after removal. Each GPS collar was equipped with a dual axis acceleration sensor, generating acceleration values on both a horizontal (X-value) and vertical (Y-value) plane which were summarized over 5-min intervals (refer to Herberg et al. 2017 for details). In addition to geographic location and activity, the GPS-collars also recorded ambient temperature in 5-min intervals.

Moose Observations

To determine how the activity values relate to moose behavior, we conducted behavioral observations on 8 captive moose during 6-hour long intervals. Each animal was observed twice in four 2-week long observations period in January, April, July, and October (i.e., totaling 48 hours of observations per moose over the entire study and 384 observation hours for all animals). The 6-hour observation intervals were spread randomly throughout the day from 600 to 2200 hours, and observations were made during all weather conditions. During an observation window the observer would stand at a distance ≤10 m from a given moose, and record the exact time when a new behavior occurred using Recon/Juno data loggers (Trimble Navigation Limited Trimble, Sunnyvale, California) to the nearest second in a procedure similar to Moen et al. (1996). The behaviors that were recorded included: foraging low (snout below the bottom of stomach), foraging medium (snout above the bottom of the stomach and below the top of the shoulders), foraging high (snout above the top of the shoulders), resting, ruminating, drinking/eating snow, walking, standing, running, shaking, grooming, and interacting (i.e., boxing). A 6-hour observation interval might contain missing data in instances where visual contact was lost with the observed moose (i.e. spooked and ran). If this happened, the moose being observed was relocated using VHF telemetry and missing observations were removed from subsequent analyses. All animal handling procedures were approved by the Animal Care and Use Committee, Alaska Department of Fish and Game, and Division of Wildlife Conservation (protocol No. 09–29).

Weather and Temporal Covariates

We obtained weather conditions from the National Oceanic Atmosphere Administration (NOAA) Climate Reference Network (CRN) weather station located at the MRC (Alaska, USA, 66.7251, -150.4493; https://www.ncdc.noaa.gov/crn/qcdatasets.html). Weather data were collected in 5-minute intervals and linearly interpolated to match the exact time stamps of GPS locations and activity data, respectively. The angle of the sun was calculated to further test how it might influence moose behavior; values were < 0 when the sun was below the horizon and > 0 when the sun was above the horizon. Seasons were assigned as follows: winter (1 November-31 March), spring (1 April-30 May), summer (1 June-31 August), and fall (1 September-31 October). Solar angles changed with each season, with larger negative values occurring during winter (i.e., less daylight) and greater positive values during summer (i.e., more daylight).

Statistical Analyses

Time stamps of NOAA temperature measurements, behavioral observation, GPS locations, and activity sensor data were not always the same; consequently we linearly interpolated temperature measurements and GPS locations between consecutive time stamps to match activity sensor time stamps. Behaviors were classified into the following 3 categories due to the overlap in X- and Y-activity values of many behaviors as well as the large number of 5-minute intervals consisting of >1 behavior: resting, foraging, and moving (Table 1).

We first calculated the proportion of time spent in each behavior category for every 5-minute activity interval by summing up the total time spent in each behavior category and dividing up by the total interval time (~5 minutes). All behavioral proportions within a 5-minute activity interval summed to 1. We also incorporated step length into some of our models, as it was shown to allow for a better distinction between resting behaviors with increasing head movements and low exertion forage/traveling behaviors (Gervasi et al. 2006). Because the temporal resolution differed between GPS locations and activity sensor data, we linearly interpolated GPS locations between consecutive 30-minute GPS locations to match the time stamps of the 5-min long behavior intervals, therefore assuming linear movements between GPS locations.

We used compositional Dirichlet regression models to quantify the relationship between the proportion of time spent resting ($R_{i,j}$), foraging ($F_{i,j}$) and moving ($M_{i,j}$) for each moose (i-th) within each 5-minute interval (j-th) as a function of X- ($X_{i,j}$) and Y-values ($Y_{i,j}$) as well as step length (Maier 2014).

We examined the effects of ambient temperature, time of day (i.e., solar angle), and habitat on the proportion of time spent resting, foraging, and moving using our best predictive model that included step length. To relate habitat use to changes in behavioral states, we first determined habitat use by spatially intersecting GPS locations with a classified imagery of habitat in ArcMAP 10.2 (ESRI 2013). Habitats were derived by the Alaska Fish and Game from a combination of satellite imagery and ground verification. Habitat types consisted of the following: Aspen, birch, water, bog, black spruce, mixed, grass, grass/black spruce, mixed closed, black spruce/birch, grass/black spruce/birch. To examine the seasonal effects of habitat in relation to changes in behavioral state and time of day, behavioral predictions were binned into 5% solar angles for each habitat within each season. Means and 95% confidence intervals were calculated for each bin using a bootstrap with 1000 iterations.

RESULTS

Captive Observations

We classified behaviors during direct observations for 4,608 5-minute intervals from 8 moose spread across 4 seasons, and retained 3,501 5-minute intervals (291.75 hours) after removing observations when the collar failed or if we lost visual contact with an animal. Moose rested more during summer observation periods (Table 2), with 67% of the time moose were observed at rest, 25% as foraging, and 8% as moving. During spring, the observed moose spent more time foraging relative to the other seasons; we classified 40% of the time moose were observed foraging, 54% as resting, and 6% as moving (Table 2). The proportion of time spent moving was similar for all seasons and ranged from 6% to 8%. Of these 3,501 5-minute intervals, 1,559 consisted entirely of resting behaviors, 106 foraging behaviors, while none consisted of only moving behaviors. The majority of 5-minute intervals (*n* = 1,836) consisted of more than one target behavior category (resting, foraging, and/or moving; hereafter referred to as mixed intervals) (Table 3).

Mean X and Y values were lowest during pure resting intervals ($\bar{x}_X$ = 1.71 ± 6.21 [SD]; $\bar{x}_Y$ = 0.84 ± 5.09) and highest for mixed intervals (X-value $\bar{x}$ 39.13 ± 23.75; Y-value $\bar{x}$ 28.90 ± 24.18; Figure 2; Table 3). Average X- and Y-activity values for all behavioral categorizations (resting, foraging, and mixed) varied significantly across seasons (ANOVA$_{X:}$ $F_3 = 22.13$, $p < 0.001$; ANOVA$_Y$: $F_3 = 35.53$, $p < 0.001$). X and Y values were consistently higher for all behavioral categories during spring and summer compared to fall and winter, with the highest values observed during summer

(Table 4). A post-hoc Tukey test showed that accelerometer data were significantly different among all seasons (adjusted p-value < 0.001) with the exception of winter and fall season X-values (adjusted p = 0.78). These results justified the need to build different models for spring, summer, and combined fall/winter seasons.

Captive Models

Models were built utilizing a total of 2,449 5-minute intervals from 8 moose spread across the combined fall/winter (n = 1199), spring (n = 578) and summer (n = 672) seasons. Models were evaluated using 1052 5-minute intervals withheld from model building from the same 8 moose spread across fall/winter (n = 515), spring (n = 249), and summer (n = 288). The best model for all 3 seasons predicted the proportion of time spent resting, foraging, and moving as a function of X and Y values as well as step length (refer to Herberg et al. 2017 for details). The lowest Root Mean Squared Error (RMSE) was observed for the winter/fall model (RMSE: 0.1640), followed by summer (0.1871) and spring (0.2045). Small differences in RMSE between seasonal models using activity values and step length as predictors and those using only activity values as predictors suggests that X and Y values alone can provide good predictions of proportions of behaviors for studies utilizing larger time gaps between GPS locations. However, models without step length were found to consistently over predict proportion of time spent resting during observed foraging bouts, especially during summer.

Captive Moose Predictions

During spring, summer, and fall, MRC moose were more likely to increase the proportion of time they spent resting during the middle of the day (greatest angle of the sun) and the middle of the night (lowest angle of the sun) and were more likely to be foraging and moving during crepuscular periods (Figure 3). This pattern differed for winter; moose activity (foraging and moving) peaked during crepuscular times as well as the middle of the night (Figure 3). Along with the sun's position, we observed changes in behavior in association with variation in ambient temperature. During all seasons, with the exception of winter, we observed a positive association between the mean proportions of time spent resting and higher ambient temperature (Figure 4). This association varied by season, with increases in rest occurring at higher temperatures during spring (>18°C) than summer (>16°C) and fall (10°C). We observed a slight increase in moving behavior at temperatures >25°C during summer. Moose were more active in aspen and birch stands during the summer season; as solar angle increased moose utilizing both black spruce stands and bogs displayed the highest proportions of resting behaviors (Figure 5). Moose utilizing black spruce stands were less active during all solar angles during spring compared to those utilizing bogs, aspen and birch stands. Activity patterns did not vary much between habitat type and time of day during the winter and fall seasons (Figure 5).

DISCUSSION

We established that dual-axis activity sensors programed to record activity values in 5-minute intervals can be used to predict the proportion of time spent resting, foraging, and moving in either captive or free-ranging moose. While previous studies have utilized behavioral observations of captive animals to validate collar activity sensors, most have chosen to use time intervals consisting of only purely active or inactive behaviors to build predictive models (Ungar et al. 2005; Löttker et al. 2009). Studies that did utilize time intervals encompassing more than one behavioral state typically converted intervals to the mode behavior observed within that time period (e.g., Moen et al. 1996), which often resulted in substantial increases in error when predicting intervals of mixed behaviors (Moen et al. 1996, Löttker et al. 2009). Nearly all of our observed active 5-minute time intervals contained a mix of active behaviors (foraging, walking, running, interacting, drinking) and inactive behaviors (standing, vigilance). These observations were consistent with findings in captive roe deer (*Capreolus capreolus*), where nearly all observed active intervals contained inactive behaviors (Gottardi et al. 2010) – this confirms the need for a modeling approach (e.g., the Dirichlet modeling technique we used) that incorporates a natural mix of behaviors in ruminants.

The significant differences we observed in activity values between all 4 seasons suggest there is a need to develop season-specific (i.e., spring, summer, fall/winter combined) predictive models in this system. Several factors may affect, alone or in concert, the seasonal differences we observed. Moose's body condition may affect GPS collar fit, with the loosest fit occurring during spring and transitioning to the tightest fit during late fall/early winter (Dan Thompson, pers comm.), although we did not observe significant differences in X and Y values resulting from variation in individual moose collar fit and behavior, potentially due to the low number of observation hours performed (results not shown) in a side project designed to test how the collar fit affects activity counts. In general, however, looser collar fit combined with increased foraging activity during spring and summer could explain the higher activity values observed during both resting and active 5-minute behavioral states during those seasons compared to winter and fall. Moen et al. (1996) found increased activity counts during summer due to a combination of increases in browsing, head movement needed to strip leaves, and head movement from insect harassment. Significant differences were found in both phases of a second collar-fit experiment developed in a lab; increased rotation ("movement") and looser collar fit on the collar machine resulted in increased activity values. Significant differences were found between behavioral type using trained horses, with standing behaviors exhibiting the lowest activity values and trotting exhibiting the highest values (McGraw et al. In prep). The results of this experiment corroborate with the finding of Moen et al. (1996).

Captive MRC moose appear to modify their behavior in response to changes in ambient temperature, solar angle, and habitat type. Moose are known to be physiologically sensitive to heat (Renecker and Hudson 1986, Renecker and Hudson 1989; McCann et al. 2013). Renecker and Hudson (1986, 1990) found that temperatures greater than -5°C in the winter and 14-20°C during the warm season (late spring to early fall) were associated with increased metabolic, heart, and respiratory rates, reduced food intake, and reduced body weight. McCann et al. (2013) found similar thresholds for late spring to early fall (17-24°C). These were based on 2 and 4 captive moose respectively, and highlight the difficulty of assessing thermal thresholds outside of a captive setting. Temperature-dependent changes in behavior were the least pronounced during winter for MRC moose. Street et al. (2015) found slight increases in activity values at moderate temperatures during winter. These findings corroborate with the slight increase in activity we observed as ambient temperatures increased toward 0°C. Well adapted to tolerate cold temperatures, moose are limited by both forage quantity and quality during the winter; this could explain why we observed relatively constant activity levels across much of the ambient temperature gradient during this season (Schwartz et al. 2007). During both the summer and fall we saw marked decreases in the proportion of active behaviors as temperatures increased. The mean proportion of active behaviors decreased considerably at temperatures exceeding 15°C and 5°C during summer and fall respectively, suggesting that moose during these seasons are faced with the tradeoff between resting more frequently to reduce thermal stress and seeking quality food sources and foraging. Forced to rest during times of increased ambient temperatures, moose forfeit feeding opportunities and this deficit has been shown to reduce weight and overall body condition (Renecker and Hudson 1992).

Our results suggest, that when experiencing warm temperatures during late spring (i.e., May), moose may choose to take advantage of increased forage quality and abundance at the cost of potential thermal stress. As spring advances, rapid plant growth occurs and nutritional quality peaks. This time period also corresponds with peak energetic demands on gestating and lactating female moose (Schwartz et al. 2007); Gasaway and Coady (1974) indeed found that the metabolizable energy requirement by the end of the gestation period is 6-fold compared to March. Parturition initiates an even more energy-demanding phase, 2- to 3-fold that of gestation. Energy needs therefore peak during the early summer and gradually decline as the young are weaned (Schwartz et al. 2007).

Behavioral responses of moose to thermal conditions are consistent throughout much of North American moose range. Moose occupying the boreal forest of Québec utilized conifer forest as a thermal refuge more frequently when ambient temperatures were high (Dussault et al. 2004). Likewise, moose in British Columbia were found to select for mature forest when temperatures exceeded critical limits (Schwab and Pitt 1991). We observed similar patterns at the MRC, where moose utilizing conifer stands during both spring and summer rested more than those utilizing aspen and birch stands, especially during the middle of the day when the sun and ambient temperatures were peaking. Additionally, moose in Alberta were found to bed in wet meadows during summer to reduce both respiration rates and energy expenditure (Renecker and Hudson 1990). At the MRC, we found that the captive moose that were using bogs during summer displayed high proportions of resting behaviors, which indicates that they may be using bogs as thermal refuges Future efforts should focus on incorporating more data from the Carstensen et al. (2014) study to investigate fine-scale behavioral patterns of moose in northeastern Minnesota in response to habitat and ambient temperature.

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Table 1. Description of the 3 behavior categories that we used to evaluate the performance of dual-axis accelerometer values for predicting moose behavior. Observations were made on 8 GPS-collared captive female moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015.

Behavior	Classification	Description
Laying	Resting	Bedded position with legs generally tucked, head could be up or down.
Ruminating	Resting	Predominantly bedded position, with or without head movement. Infrequently while standing.
Standing	Resting	Upright quadruped position, with or without head movement (i.e. vigilance).
Drinking	Foraging	Consumption of water during the warm season (spring, summer, fall). Could be standing or lying.
Snow intake	Foraging	Consumption of snow during the winter season. Could be standing or lying.
Foraging low	Foraging	Consumption of foraged plants, with mouth below the bottom of the stomach while standing or while lying.
Foraging medium	Foraging	Consumption of foraged plants with the mouth above the bottom of the stomach but below the shoulder hump.
Foraging high	Foraging	Consumption of foraged plants with the mouth above the top of the shoulder hump.
Walking	Moving	Slow methodical movement, forward or backward.
Running	Moving	Accelerated movement, forward.
Shaking	Moving	Accelerated up-down/side-to-side head and body movement while walking, foraging, standing, or lying.
Grooming	Moving	Self-grooming with hind hooves and/or rubbing against trees.
Interaction	Moving	Social interaction with other moose, with forelegs leaving the ground (i.e. boxing).

Table 2. Proportion of time 8 captive adult moose (>2 years of age) were observed in each behavioral state during 3,501 5-minute intervals at the Moose Research Center, Kenai Peninsula, Alaska distributed across four, user-defined seasons during 2015.

Season	Number of 5-minute intervals	Rest	Forage	Moving
Winter (1 Nov-31 Mar)	872	0.56	0.38	0.07
Spring (1 Apr-30 May)	827	0.54	0.40	0.06
Summer (1 June-31 Aug)	960	0.67	0.25	0.08
Fall (1 Sep-31 Oct)	842	0.60	0.34	0.06

Table 3. Mean (±SD) X- and Y-activity values of 5-minute intervals of pure behaviors such as resting or foraging, mixed behaviors, or for all 3,501 observation intervals obtained from observing 8 adult captive female moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015. None of the intervals we observed contained strictly moving behaviors.

Behavior	Number of 5-minute intervals	Mean activity X	Mean activity Y
Resting	1559	1.71 (±6.21)	0.84 (±5.09)
Foraging	106	38.94 (±13.31)	20.25 (±8.60)
Mixed	1836	39.13 (±23.75)	28.90 (±24.18)
All	3501	22.46 (±25.77)	16.15 (±22.59)

Table 4. Mean (±SD) X- and Y-activity values across four seasons of 5-minute intervals of pure behaviors such as resting or foraging, mixed behaviors, or for all 3,501 intervals obtained from observing 8 adult captive female moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015. None of the intervals we observed contained strictly moving behaviors.

Behavior	Season	Number of 5-minute intervals	Mean activity X	Mean activity Y
Resting	Winter	334	0.19 (±2.02)	0.13 (±1.44)
	Spring	316	1.23 (±5.93)	1.02 (±7.51)
	Summer	521	3.94 (±8.91)	1.64 (±6.20)
	Fall	389	0.42 (±22.44)	0.24 (±1.81)
Foraging	Winter	20	33.10 (±9.71)	15.60 (±5.92)
	Spring	44	38.84 (±7.62)	22.80 (±5.82)
	Summer	5	59.17 (±18.58)	41.00 (±10.56)
	Fall	29	39.17 (±17.90)	14.97 (±4.89)
Mixed	Winter	518	31.85 (±18.13)	22.95 (±18.18)
	Spring	467	37.27 (±21.30)	27.57 (±23.19
	Summer	434	55.17 (±28.97)	45.18 (±27.54)
	Fall	424	33.77 (±18.26)	21.01 (±19.82)

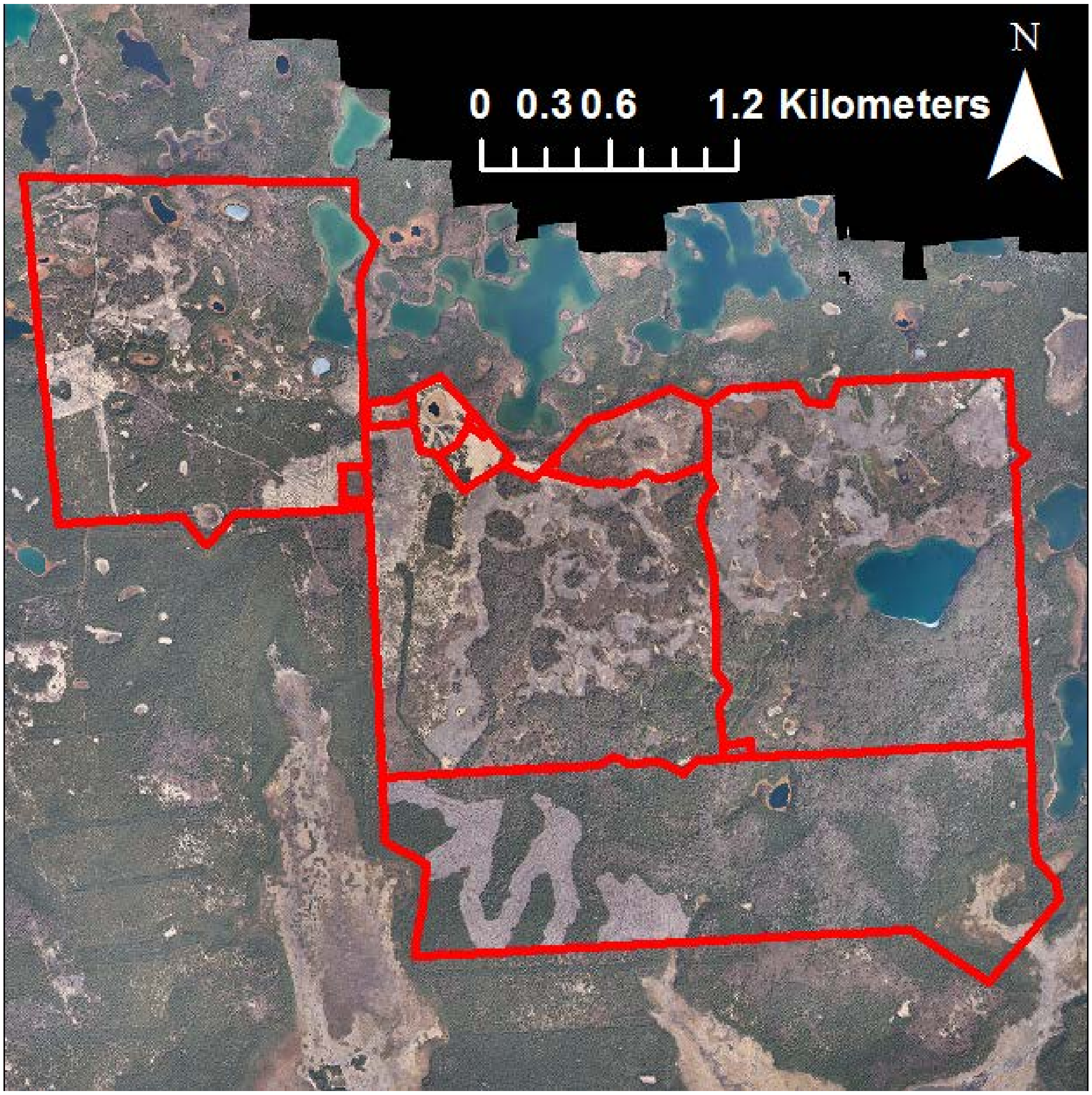

Figure 1. Map of the Moose Research Center in Game Management Unit 15A, Kenai Peninsula, Alaska.

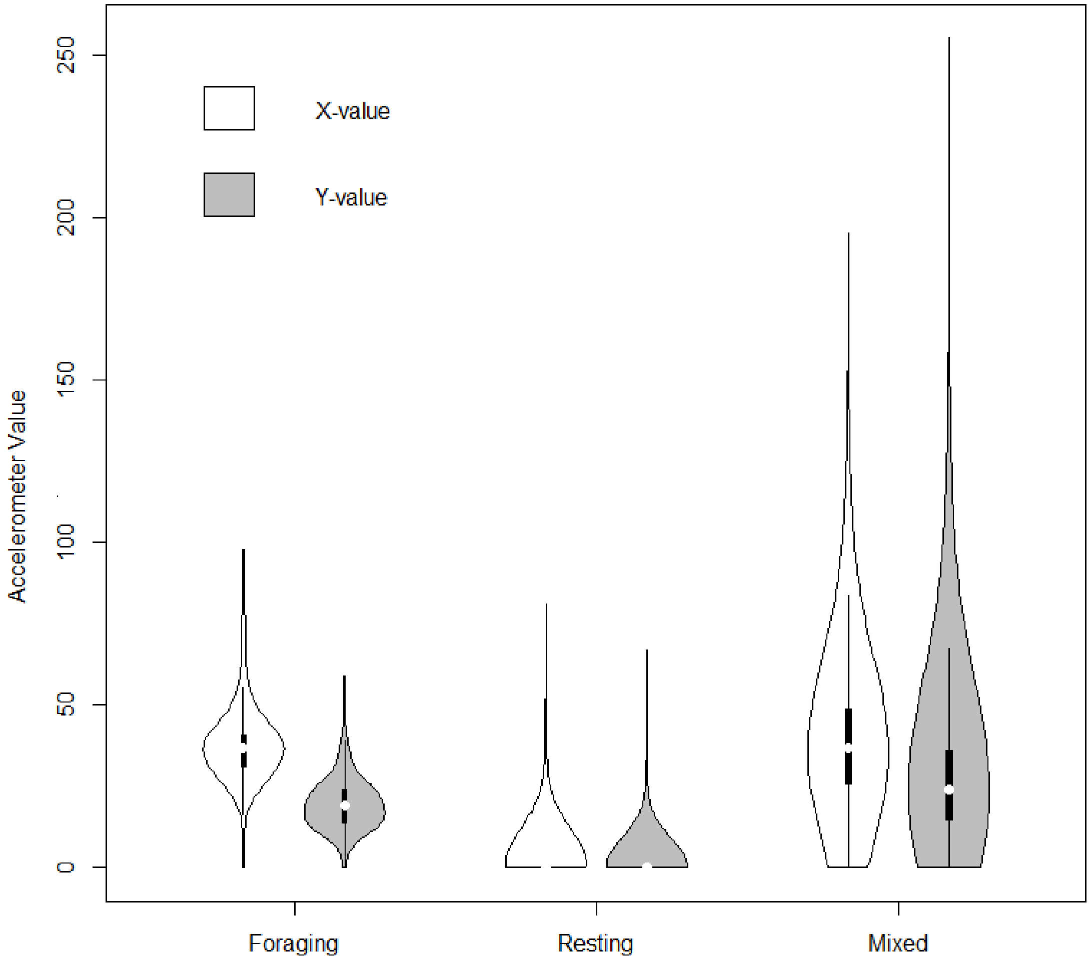

Figure 2. Visualization of X- and Y- accelerometer values from a total of 3,501 5-minute intervals across all seasons from 8 adult captive moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015. 1560 intervals contained only resting behaviors, 98 only foraging behaviors, but none contained only walking/resting behaviors. The majority (1843) were mixed and contained more than 1 behavior.

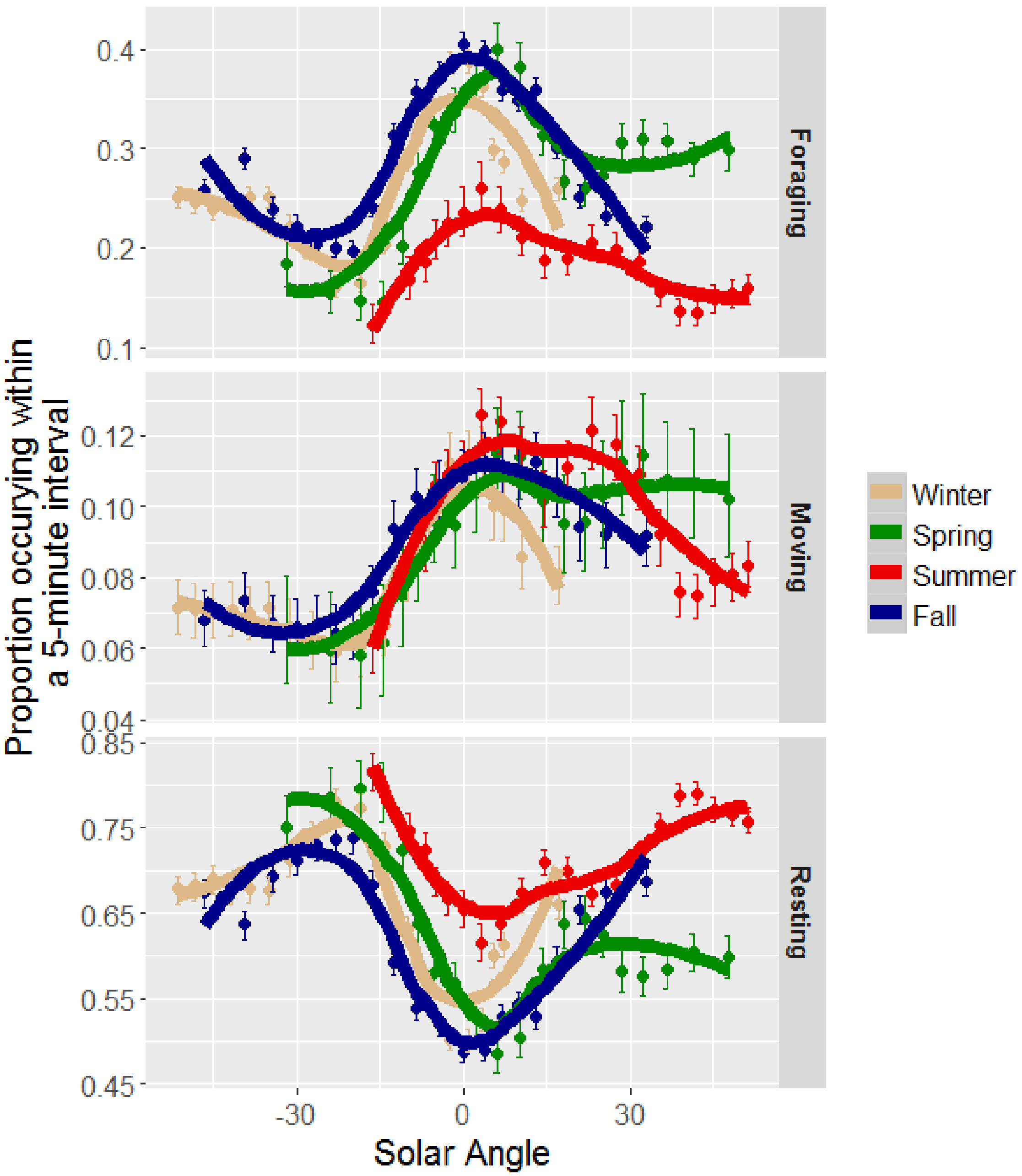

Figure 3. Mean predicted proportions of time spent foraging, moving, and resting within a 5-minute interval in response to changing solar angles for 8 captive moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015. Crepuscular times are centered on zero with lowest values corresponding to the middle of the night and the highest values to the middle of the day. Predictions were binned into 5% quantiles. Means and 95% confidence intervals were calculated using bootstrapping with 1000 iterations. Trends are depicted using locally weighted smoothing curves.

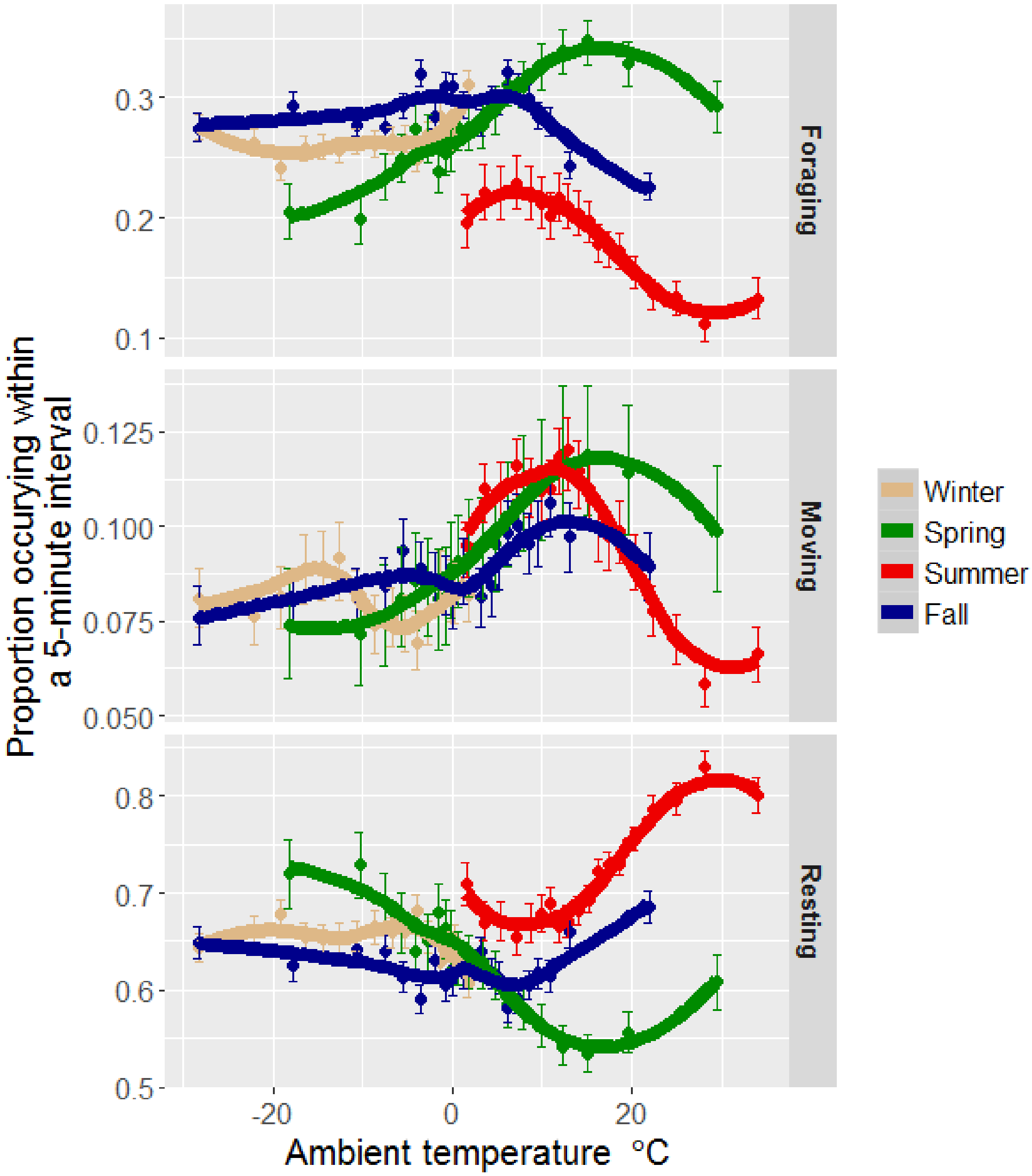

Figure 4. Mean predicted proportions of time spent foraging, moving and resting within a 5-minute interval in response to changing ambient temperature for 8 captive moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015. Predictions were binned into 5% quantiles. Means and 95% confidence intervals were calculated using bootstrapping with 1000 iterations. Trends are depicted using locally weighted smoothing curves.

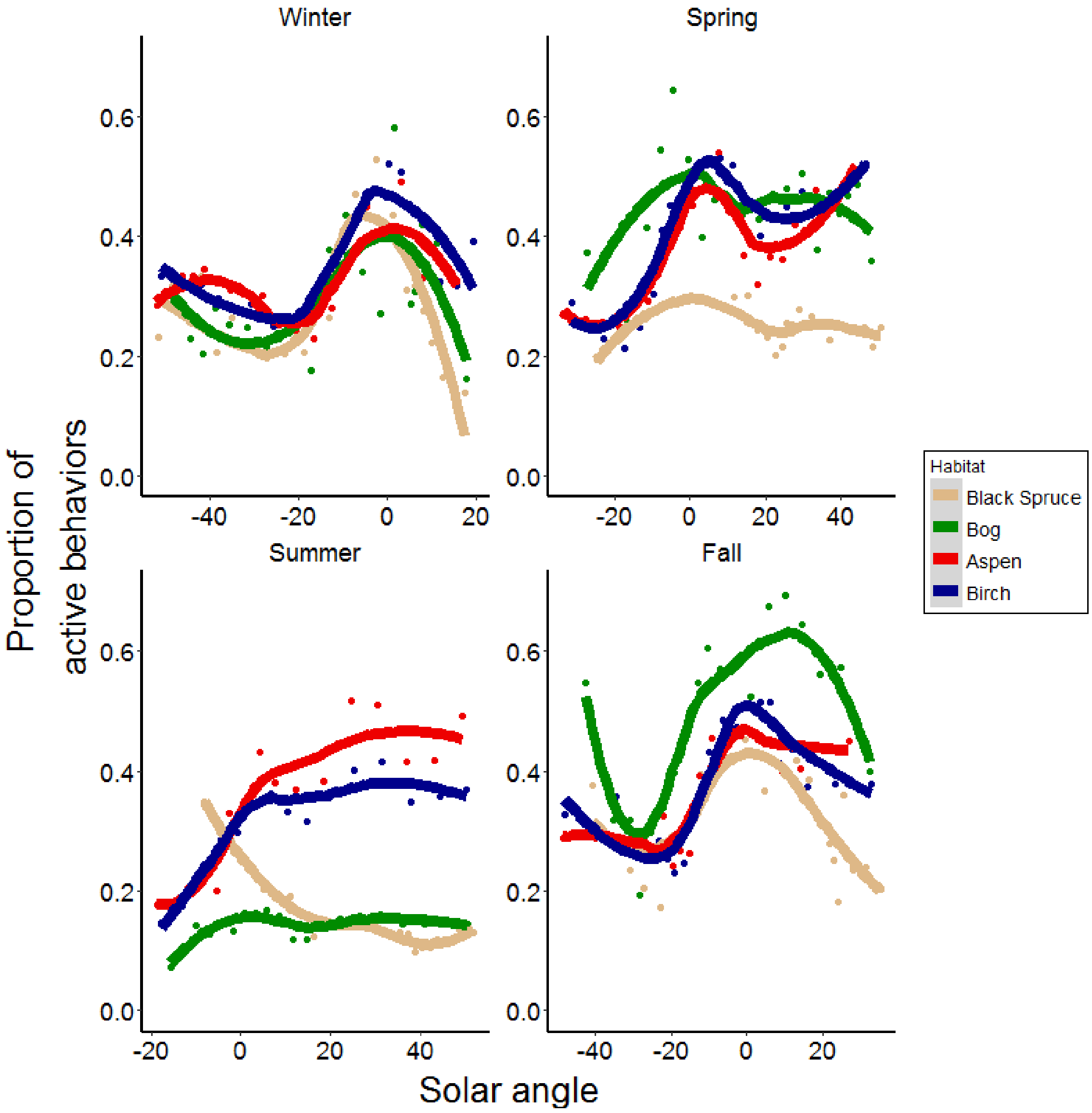

Figure 5. Mean combined predicted proportions of foraging and moving behavioral states (active) within a 5-minute intervals collected within each habitat in response to changing solar angles for 8 captive moose at the Moose Research Center, Kenai Peninsula, Alaska during 2015. Crepuscular times are centered on zero with lowest values corresponding to the middle of the night and the highest values to the middle of the day when the sun is at its highest point. Predictions were binned into 5% quantiles. Means and 95% confidence intervals were calculated using bootstrapping with 1000 iterations. Trends are depicted using locally weighted smoothing curves.

A TECHNIQUE FOR DEPLOYMENT OF RUMEN BOLUS TRANSMITTERS IN FREE-RANGING MOOSE (ALCES ALCES)[1]

Larissa Minicucci[2], Michelle Carstensen[3], John Crouse[4], Jon M. Arnemo[5,6], Aliva Evans[6]

ABSTRACT

Rumen boluses have been routinely used in domestic animals to prevent disease, administer medications, and provide identification. Recent uses for rumen boluses in wildlife, such as collection of physiological data, have made it necessary to adapt deployment techniques developed for livestock to free-ranging animal populations. Research aimed at determining causes of mortality in Minnesota free-ranging moose (*Alces alces*) included the use of a rumen bolus, called a mortality implant transmitter (MIT), to monitor heart activity and provide instant notification of death. In 59 attempts to deploy MITs from 2013-2014, 16 (27%) failures occurred when boluses were not swallowed. As a result, in 2014, captive moose (n = 10) in Alaska were utilized to evaluate new methods for MIT deployment. Measurement of distances from the mandible to the nose and from the mandible to the commissure of the lips in both skulls and captive live moose provided guidance for selection of an appropriate-sized bolus applicator. A Schulze mouth gag was used to aid insertion of the applicator and canola oil was used to lubricate the bolus to facilitate swallowing. Time to first swallow following sedative reversal was measured in both captive moose (Alaska) and free-ranging moose (Minnesota) and found to be less for captive moose (2.3 ± 0.2 min, n = 10) than free-ranging moose (4.4 ± 0.8 min, n = 20), and time to continuous swallowing was 8.1 ± 1.0 min for 18 free-ranging moose. Using the new technique, success rates for MIT deployment were 100% (10/10) for captive moose and 85% (20/23) for free-ranging moose in 2015. Thus, it is recommended that rumen bolus placement in free-ranging moose be attempted using appropriately sized equipment at least eight minutes after sedative reversal when using a xylazine/tolazoline drug combination.

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DEPARTMENT OF
NATURAL RESOURCES

SURVEILLANCE FOR HIGHLY PATHOGENIC AVIAN INFLUENZA IN MINNESOTA'S WILD BIRDS IN 2016-17

Chris Jennelle[1], Michelle Carstensen, Erik C. Hildebrand, Tim White[2], and Tom Cooper[3]

SUMMARY OF FINDINGS

Surveillance for highly pathogenic avian influenza (HPAI) virus in wild birds is a national priority in the United States. Outbreaks of HPAI continue to occur in domestic poultry, wild birds, and people in Asia and Europe, and there is concern that these viruses may be introduced into North America. The Minnesota Department of Natural Resources (MNDNR) has partnered with the United States Department of Agriculture's Wildlife Services (USDA-WS) since 2007 to conduct HPAI surveillance in wild birds, but it wasn't until 2015 that a highly pathogenic strain of H5N2 was detected in Minnesota. Since detection in a poultry facility in Pope County MN on February 27 2015, MNDNR extended our partnership with the United States Geological Survey's National Wildlife Health Center (USGS), the United States Fish and Wildlife Service (USFWS), and the University of Minnesota (UMN) to conduct surveillance for any HPAI virus subtypes in Minnesota wild birds. The H5N2 HPAI virus strain is a combination of the highly pathogenic Eurasian H5 and low pathogenic North American H2 subtypes. Since June 2015, there have been no detections of HPAI in MN poultry facilities or in wild birds sampled in MN. From May 2016 through March 2017, the MNDNR and partners collected swab samples from 1,065 dabbling ducks across six watersheds; AI viral material was detected in 17% of these samples. Only 2.2% and 0.3% of all samples contained detectable H5 and H7 viral material, respectively. The highest apparent prevalence of LPAI was in the St. Croix watershed at 44%, and the lowest was 10.4% in the Red watershed. Only three successful viral isolates were recoverable and included H2N3 (Mississippi Headwaters watershed), H10N7 (Upper Mississippi-Black Root watershed), and H4N9 (Upper Mississippi-Black Root watershed). All of these samples were collected as part of the 2016 USDA national surveillance efforts with oropharyngeal/tracheal and cloacal samples combined. No HPAI positive cases were found. In partnership with the UMN, MNDNR began a collaboration on a project to investigate avian influenza dynamics in ring-billed gulls (*Larus delawarensis*) across Minnesota; results are pending.

INTRODUCTION

Avian Influenza (AI) is a viral infection that occurs naturally in wild birds, especially waterfowl, gulls, and shorebirds. It is caused by type A influenza viruses that have 2 important surface antigens, hemagglutinin (H) and neuraminidase (N), that give rise to 144 possible virus subtypes. Influenza viruses vary widely in pathogenicity and ability to spread among birds. The emergence of an Asian strain of highly pathogenic avian influenza (HPAI) H5N1 virus in 1996, and subsequent spread of the virus in Asia, Africa, and Europe, killed thousands of wild birds and millions of domestic poultry. In 1997, HPAI H5N1 became zoonotic in Hong Kong and to-date has infected at least 859 humans around the world, resulting in 453 deaths (World Health

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Organization 2017). Furthermore, since 2013 HPAI H7N9 has been confirmed in 1,552 people (mostly in eastern Asia) with 596 deaths (Food and Agriculture Organization 2017). As of 6 June 2017, there were 51 ongoing outbreaks of HPAI in wild birds in Asia and Europe; these include strains of the subtypes H5N1, H5N2, H5N5, and H5N8 (OIE 2017). Furthermore, there were 667 ongoing outbreaks of HPAI subtypes H5N1, H5N2, H5N6, H5N8, H7N3, and H7N9 in domestic poultry operations around the world (World Organisation for Animal Health 2017). There have been no HPAI outbreaks in the US since March 2017, when an American strain of H7N9 was confirmed in domestic poultry facilities in Tennessee (USDA 2017). These results highlight that HPAI viruses continue to be active around the world and pose a threat to both wild birds and domestic poultry. The diversity of active highly pathogenic subtypes, coupled with the ability of avian influenza strains to mutate quickly underscores the pandemic risk from these viruses. As such, there is an urgent need to understand transmission dynamics, host-species susceptibility, and the role of the environment in AI dynamics.

Since the first Minnesota detection of HPAI H5N2 in March 2015, the MNDNR has collected over 7,500 samples for AI testing, which is the most of any state in the Mississippi flyway.

The migratory movements of waterfowl and other shorebirds and subsequent mixing of birds from Asia and North America in the northern latitude breeding grounds likely facilitated the mixing of low pathogenicity avian influenza (LPAI) and HPAI strains (Pasick et al. 2015). Such mixing resulted in discovery of 3 reassortant highly pathogenic strains including H5N1 (World Organisation for Animal Health 2014), H5N2 (World Organisation for Animal Health 2014, Pasick et al. 2015), and H5N8 (Ip et al. 2015) in British Columbia and the western United States in 2014.

In August and December of 2016, HPAI H5N2 was again detected in wild waterfowl in Alaska and Montana, respectively. In addition, there were several detections of HPAI H7N9 (distinct from Asian strain) that affected domestic poultry facilities in the Mississippi flyway. Since July 2015, there have been over 80,000 wild waterfowl tested in the continental US and only 4 positive HPAI detections have occurred. Our efforts to detect HPAI in wild birds, if present, include live-bird and hunter-harvest sampling of waterfowl and the continued monitoring of morbidity/mortality events. These efforts permit the estimation of temporal and spatial detection limits for AI on the Minnesota landscape, which leads to development of specific hypotheses that can help us understand AI dynamics in wild birds.

METHODS

We collected samples for AI testing from three sources: public- or agency-reported morbid or dead wild birds (i.e., morbidity and mortality events), live-captured and released ducks through banding programs, and hunter-harvested ducks. Dabbling ducks were primarily sampled, including mallard (*Anas platyrhynchos*), blue-winged teal (*A. discors*), American green-winged teal (*A. crecca*), American wigeon (*A. americana*), gadwall (*A. strepera*), American black duck (*A. rubripes*), northern pintail (*A. acuta*), northern shovelor (*A. clypeata*), wood duck (*Aix sponsa*), and ring-necked duck (*Aythya collaris*). Morbidity and mortality samples depended on opportunistic circumstances and public willingness to report or submit dead birds, and were collected statewide. Sampling live wild ducks and hunter-harvested ducks afforded more control over sampling design elements; both spatial and temporal dimensions were within our design control.

USDA National Plan Sampling

As part of the 2016 USDA National Surveillance Plan (USDA 2016a), which called for 1,040 oropharyngeal/tracheal and cloacal swab samples from dabbling ducks in MN, the MNDNR partnered with both USFWS and UMN to achieve the sample goal between summer and winter 2016. As part of an independent winter avian influenza study, Dr. Patrick Redig and his technicians assisted in our efforts to achieve our winter sampling goals. The samples collected were broken down by watershed (Minnesota, Mississippi Headwaters, Red River, St. Croix, Upper Mississippi – Black Root, and Western Lake Superior) and season (summer, fall, and winter). The source of samples was from live waterfowl or hunter-harvested waterfowl. We collected swab samples from the oropharyngeal cavity or trachea (depending on live or dead birds) and cloacal cavities of each bird in order to test for viral shedding. Both swab samples from a bird were placed in the same brain-heart infusion (BHI) media, and kept cool in a portable cooler with ice packs or a refrigerator. Samples were shipped overnight to the US Geological Survey National Wildlife Health Center (USGS) for avian influenza virus (AIV) testing using a real time reverse transcription polymerase chain reaction (rRT-PCR) matrix test, which tests for type-A influenza virus RNA. Material from positive matrix tests were further tested with an H5 and H7 assay. If either H5 or H7 assay were positive, the remaining sample material was sent to the National Veterinary Services Laboratories in Ames, IA for confirmation and strain-typing.

Morbidity and Mortality Sampling

Through outreach on the MNDNR and Minnesota Board of Animal Health websites and official press releases, we solicited the public and agency staff to report any wild birds exhibiting neurological symptoms consistent with AIV infection anywhere in the state. We investigated reports of dead ducks if circumstances of mortality were unclear and if individuals showed neurologic signs. We emphasized the need to report dead birds as soon as possible to ensure collection of viable tissue samples; generally we only collected samples from birds that were deceased for <24 hours. Depending on the resources available for staff (e.g., BHI media and swabs), we either collected whole carcasses (double-bagged and frozen) or swabs from the trachea and cloaca of dead birds. Both swab samples from a morbidity/mortality sample bird were placed in the same BHI media, and kept cool in a portable cooler with ice packs or a refrigerator. Whole carcasses were shipped overnight to the USGS National Wildlife Health Center or the University of Minnesota's Veterinary Diagnostic Laboratory for necropsy and AIV testing using real time reverse transcription polymerase chain reaction (rRT-PCR) test, which tests for AIV RNA. Swab samples were submitted to the US Department of Agriculture National Wildlife Disease Laboratory (USDA) in Fort Collins, CO. If samples tested AIV positive initially at any lab, they were forwarded to the National Veterinary Services Laboratories in Ames, IA for confirmation and strain-typing. We had no fixed sample goal for this surveillance effort due to the opportunistic nature of public discovery and reporting of sick or dead birds. We used these data as an auxiliary source of information in our surveillance efforts.

RESULTS AND DISCUSSION

From May 2016 through March 2017, the MNDNR in partnership with USDA-WS and the USFWS collected 1,065 oropharyngeal/tracheal and cloacal samples from dabbling ducks across 6 watersheds of Minnesota (Figure1). As part of the USDA national surveillance plan, Minnesota was asked to provide 1,040 samples towards their goals, and we slightly exceeded that number (Table 1). While about 17% of all samples were positive for LPAI, this aligns with expectations of type-A avian influenza prevalence in waterfowl (Webster et al. 1992). Only 3 LPAI subtypes were isolated from positive samples, and they included H2N3, H4N9, and H10N7. This result underscores the difficulty of acquiring enough viral material in swab samples to successfully identify AI subtypes. We did not detect HPAI in any samples. Of particular note is that apparent prevalence of H5 and H7 LPAI subtypes across all samples were 2.2% and 0.3% - these subtypes are typically what can become highly pathogenic.

From August 1 2016 through June 15 2017, we collected 14 morbidity and mortality samples from wild birds. Of these submissions (1 American crow, 4 common terns, 2 mallards, 2 trumpeter swans, 2 snow geese, 1 red-tailed hawk, 1 wild turkey, and 1 house finch), none tested positive for HPAI (Table 2). In June 2016, MNDNR partnered with UMN and USGS to collect and test oropharyngeal and cloacal samples (combined) from 200 common terns in a breeding colony at Interstate Island near Duluth, MN. Avian influenza was not detected in any of the samples.

Since the outbreak of HPAI began in Minnesota poultry in March 2015, through June 2017, the MNDNR and partners have collected and tested over 7,500 samples for HPAI, which included waterfowl feces (Jennelle et al. 2016), reported wild bird mortalities, hunter-harvested waterfowl, live waterfowl, and wild turkeys (Jennelle et al. 2017). To date, there has been only one confirmed HPAI H5N2 positive result in 2015, a likely spillover species - Coopers Hawk (predator of small birds) (Jennelle et al 2016). The positive hawk was only 12 miles from an infected poultry facility. The final report on the 2014-2015 HPAI outbreak in the US, the largest outbreak in the US, indicated that 7.4 million domestic turkeys and 43 million egg-layers/pullet chickens were impacted, costing nearly a billion dollars for the response, indemnity, and future preparedness actions (USDA 2016b). The report highlights poultry facility biosecurity as a major concern and likely contributor to the spread and broad impact of the outbreak (USDA 2016b).

Current Projects and Future Surveillance

The MNDNR is collaborating on a newly funded Legislative-Citizen Commission on Minnesota Resources (LCCMR) project led by Dr. Marie Culhane of UMN to investigate AI prevalence, exposure, and potential health effects on ring-billed and herring gulls across Minnesota. Other partners in this effort include the USFWS, USDA-WS, and MN Turkey Growers Association. The study began in fall 2016 and will continue through 2017, with field efforts led by M.S. student Todd Froberg who is supervised by Dr. Francie Cuthbert. Field sampling efforts have focused on capture and sampling of gulls at landfills, farm fields, and gull colonies across Minnesota. Thus far the team has collected oropharyngeal, cloacal, and blood samples from over 700 ring-billed gulls across MN. The results are pending, and data collection is expected to be completed by December 2017.

As part of the USDA national surveillance plan for 2017, Minnesota has a goal of 1,140 samples from dabbling ducks for avian influenza sampling and testing. Varying sample sizes will be requested from 5 to 6 watersheds across Minnesota spanning summer, fall, and winter seasons in 2017.

MNDNR sampling and testing of morbidity and mortality events is ongoing. We have adopted a risk-based approach to AIV surveillance in wild birds designed to respond to new detection events in a rapid and efficient manner. Three triggers will initiate intensive, and spatially and temporally designed AI surveillance efforts; if HPAI virus is detected in (1) wild, migratory birds in Minnesota through ongoing morbidity and mortality surveillance, (2) wild migratory birds in the Mississippi flyway, or (3) commercial or backyard poultry in Minnesota.

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Table 1. Avian influenza swab results (*n* = 1,065) from Minnesota participation in the 2016 USDA National plan*.

Watershed	n	Type-A	LPAI %	HPAI %
Minnesota	331	48	14.5	0
Mississippi Headwaters	272	52	19.1	0
Red	221	23	10.4	0
St. Croix	36	16	44.4	0
Upper Mississippi–Black Root	143	33	23.1	0
Western Lake Superior	62	7	11.3	0
TOTAL	1,065	179	16.8	0

*There were only 3 successful isolations of type-A influenza completed from these samples H2N3, H4N9, and H10N7

Table 2. Species and count of wild bird morbidity & mortality samples (n = 14) submitted by the Minnesota Department of Natural Resources for avian influenza testing from 01 August 2016 to 15 June 2017. No birds tested positive for HPAI.

Agency	Species sampled	n*
MNDNR	American crow (Corvus brachyrhynchos)	1
	Common Tern (Sterna hirundo)	4
	House finch (Haemorphous mexicanus)	1
	Mallard (Anas platyrhynchos)	2
	Red-tailed hawk (Buteo jamaicensis)	1
	Snow goose (Anser caerulescens)	2
	Trumpeter swan (Cygnus buccinator)	2
	Wild turkey (Meleagris gallopavo)	1
Total		14

*Note that multiple birds may have been submitted for a given location and time.

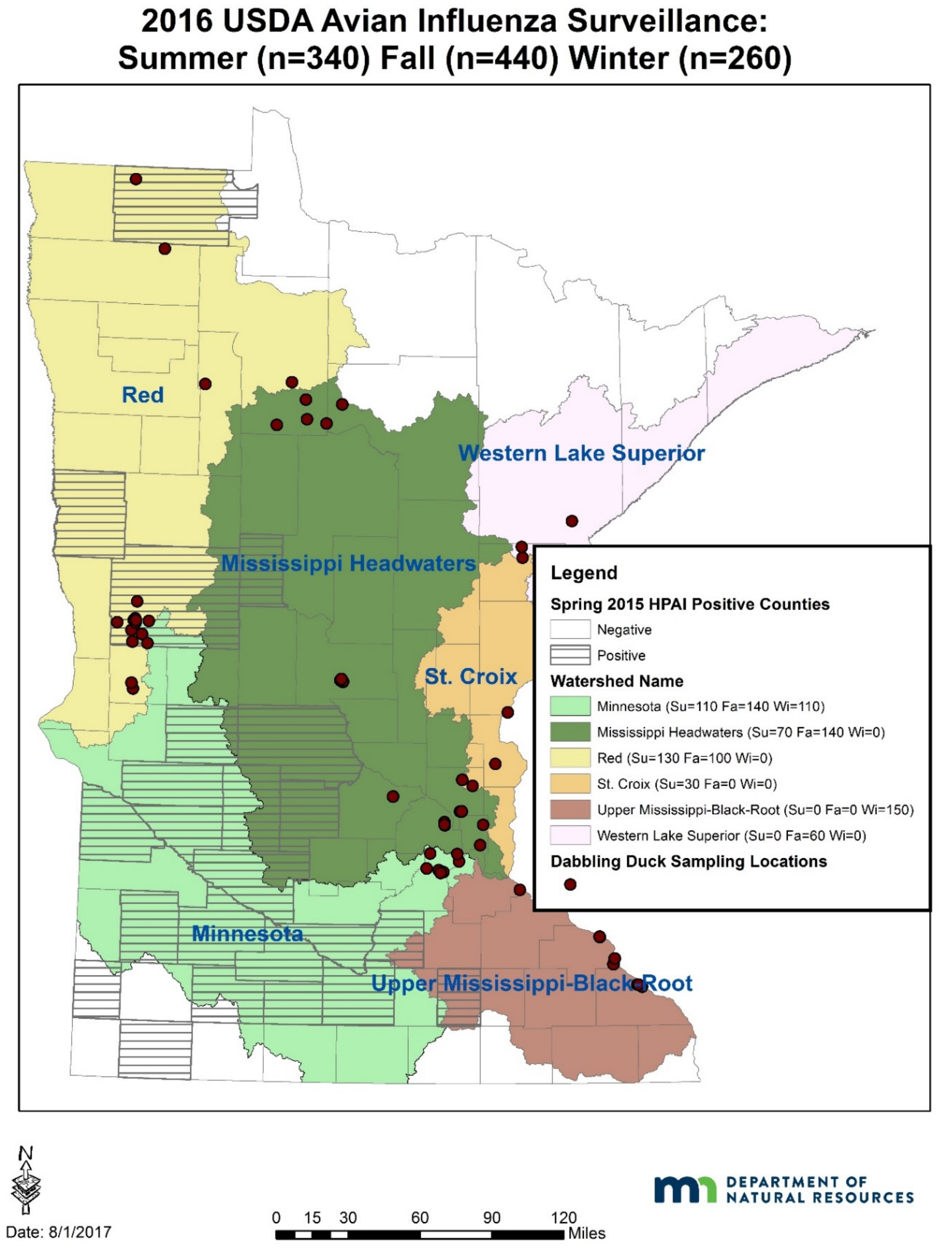

Figure 1. The United States Department of Agriculture (USDA) allocation of targeted Minnesota watersheds for avian influenza sampling (n=1,040) for summer, fall, and winter 2016. The 3 sample sizes noted beside watersheds in the legend are the quotas requested by USDA for summer, fall, and winter sampling, respectively. Note we exceeded our quota by 25 samples.

SURVEILLANCE FOR HIGHLY PATHOGENIC AVIAN INFLUENZA VIRUS IN WILD TURKEYS (*MELEAGRIS GALLOPAVO*) OF MINNESOTA, USA DURING 2015 OUTBREAKS IN DOMESTIC POULTRY[1]

Christopher S. Jennelle[2], Michelle Carstensen[2], Erik C. Hildebrand[2], Paul Wolf[3], Daniel A. Grear[4], Hon S. Ip[4], Louis Cornicelli[2]

ABSTRACT

An outbreak of a novel reassortant of highly pathogenic avian influenza A (H5N2) virus (HPAIV) decimated domestic turkeys (*Meleagris gallopavo*) from March through mid-June, 2015 in the state of Minnesota, USA. In response, as part of broader surveillance efforts in wild birds, we designed a pilot effort to sample and test hunter-harvestedwild turkeys *(Meleagris gallopavo*) for HPAIV in Minnesota counties with known infected poultry facilities. We also collected opportunistic samples from dead Wild Turkeys or live wild turkeys showing neurologic signs (morbidity and mortality samples) reported by the public or state agency personnel. Cloacal and tracheal samples were collected from each bird and screened for avian influenza virus (AIV) RNA by real-time reverse transcription PCR. From 15 April to 28 May 2015, we sampled 84 hunter-harvested male wild turkeys in 11 Minnesota counties. From 7 April 2015 through 11 April 2016, we sampled an additional 23 wild turkeys in 17 Minnesota counties. We did not detect type A influenza or HPAIV from any samples, and concluded, at the 95% confidence level, that apparent shedding prevalence in male wild turkeys in central Minnesota was between 0 and 2.9% over the sampling period. The susceptibility of wild turkeys to HPAIV is unclear, but regular harvest seasons make this wild gallinaceous bird readily available for future AIV testing.

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USING REPEATED MEASURES TO ESTIMATE CRITICAL THRESHOLDS AND CLASSIFY STATES IN SHALLOW LAKES

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SUMMARY OF FINDINGS

Shallow lakes can quickly transition between 2 alternative stable states: a clear state dominated by submerged aquatic vegetation, which provides critical habitat for waterfowl, and a turbid state characterized by extreme algal blooms, sparse submerged vascular plants, and poor habitat quality. Theoretical models suggest that critical nutrient thresholds differentiate highly resilient clear lakes, lakes that may switch between clear and turbid states following system perturbations (e.g., weather events, zooplankton community changes), and highly resilient turbid lakes. Lake managers need decision tools to help guide and prioritize future lake projects. We are developing models to identify combinations of factors responsible for lake deterioration, to assess management potential of individual lakes, and to help gauge the relative risk of state transitions for shallow lakes. We have developed an integrated modeling framework to (1) identify critical nutrient (TP) thresholds, (2) classify attracting lake states, and (3) estimate state-dependent relationships between TP and measures of algal abundance (Chla). Here, we provide a modified version of our model that utilizes repeated lake measurements. We plan to use these and other study products to develop an interactive decision support tool that will help managers identify lakes needing special protection, fine-tune management needs of individual lakes, and rank lakes as candidates for future lake management efforts.

INTRODUCTION

Shallow lakes generally conform to one of 2 alternative stable states: a clear state with primary production dominated by submerged aquatic vegetation (SAV) and a turbid state with phytoplankton dominating over SAV (Scheffer et al. 1993). Excessive nutrient inputs from current and historical land use, food web-mediated influences and sediment disturbance caused by planktivorous and benthivorous fish, and wind all drive transitions to, and affect the resilience of, turbid states (Scheffer 1998). Shallow lakes with high nutrient levels are prone to explosive, unhealthy phytoplankton “blooms,” especially when phosphorus (P) is readily available (Scheffer 1998). Submerged aquatic vegetation, which sustains the diverse invertebrate communities that provide important food sources for waterfowl, is reduced in this turbid, algae-dominated state (Hargeby et al. 1994). Parasites associated with amphibian malformations likely have higher prevalence in turbid lakes (Johnson and Chase 2004) and nitrogen may accumulate at higher rates (Zimmer et al. 2003). It is not surprising that key goals for shallow lake management are to prevent shifts from clear to turbid states, to induce shifts from turbid to clear states, and to maintain the natural resilience of clear-water shallow lakes.

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Complex ecological and physical mechanisms are responsible for maintaining the stability of each alternative state, such as competition between primary producers. When SAV declines, phytoplankton abundance typically increases, limiting light reaching the lake bottom and further restricting SAV in a positive-feedback loop (Scheffer et al. 1993). Additionally, when SAV is sparse, sediments are easily disturbed by benthivorous fish and waves. Suspended sediments further increase turbidity, and mobilized P stimulates even higher phytoplankton growth rates (Scheffer 1998). In contrast, in clear-state lakes, SAV remains widely distributed and helps maintain water clarity by stabilizing sediments and taking up nutrients (Søndergaard et al. 2003). Charophytes (*Chara*) often accompany clear-water conditions in Minnesota lakes and are believed to release algal toxins (Berger and Schagerl 2004) and provide refuge for zooplankton, which may further reduce the phytoplankton population and help stabilize clear-water conditions.

Shallow lakes are notoriously difficult to restore after shifting from clear to turbid states, with turbid conditions frequently returning within 5-10 years following lake management (Søndergaard et al. 2007, Hanson et al. 2017). Theoretical models are useful for understanding how nutrients influence whether lakes will tend toward turbid or clear water states in the long run. For example, Figure 1 shows a bifurcation diagram derived from a model describing shallow lake dynamics. At low nutrient levels (left of “tip down” threshold in Figure 1), lakes can only exist in the clear stable state. At high nutrient levels (right of the “tip up” threshold in Figure 1), lakes only exist in the turbid state. In between these 2 thresholds, the system exhibits hysteresis in which 2 different steady states are possible under the same nutrient conditions, depending on whether the initial turbidity levels lie above or below the unstable state in this region of bistability (dashed line in Figure 1).

The bifurcation diagram is also useful for understanding temporal dynamics and shifts between stable states. If a lake is in the clear state with high SAV (lower solid line) and nutrient input increases beyond the “tip up” bifurcation point, the lake will likely transition quickly to the turbid state with low SAV (upper solid line). Once SAV is lost, the internal loading of nutrients increases and becomes hard to control, and external nutrient loading must be substantially reduced to the lower “tip down” bifurcation point to reverse the state shift (Scheffer and Carpenter 2003). In practice, such drastic nutrient reduction may not be possible or may only be accomplished over long time periods. Alternatively, managers may attempt to induce a state shift by forcing the system across the unstable line, e.g., by decreasing the planktivore and benthivore populations with rotenone (if nutrients can at least be reduced to the region of bistability) (Jeppesen et al. 2009). These resulting transitions are typically short-lived, however, because perturbations to the system (e.g., fish colonization, destruction of submerged vegetation) can force the lake back to the turbid state. For instance, Lake Christina, a large shallow lake in Minnesota, has been rehabilitated with fish toxicants three times in recent decades in an effort to improve habitat quality for migrating waterfowl. In each case, improved water quality and clear-state characteristics followed lake management, but the lake persistently transitioned back to turbid conditions 5-10 years after treatment (Hanson and Butler 1994, Hansel-Welch et al. 2003, Hobbs et al. 2012). Clear water conditions in Danish and Dutch lakes have also been observed to start deteriorating five years following biomanipulation (Meijer et al. 1994). Similarly, Hanson et al. (2017) showed that 8 shallow lakes in Minnesota did not transition to stable clear-state conditions during a period 2-4 years after management. Returns to turbid conditions following biomanipulation suggest that some shallow lakes may have nutrient levels beyond the “tip up” threshold in Figure 1 where only the turbid state is possible, or that observed clear states may have little ecological resilience such that small perturbations easily push the lakes back into the basin of attraction of the turbid state. These patterns are also consistent with paleolimnological findings of Ramstack Hobbs et al. (2016) who suggested that some shallow Minnesota lakes never recovered after crossing from clear- to turbid-state ecological regimes.

Failed attempts to manage turbid lakes illustrate that managers need better tools to predict whether their efforts will maintain clear conditions in high quality lakes, whether clear lakes are approaching thresholds and thus are likely to transition to turbid conditions, or if management will succeed in improving highly deteriorated lakes. Theoretical models and empirical studies suggest that we need to more accurately predict implications of changing nutrient levels and biological community features on attracting states and the likelihood that lakes will flip to turbid states. Such information will help managers prevent undesirable state shifts in shallow lakes, identify lakes that are good candidates for rehabilitation, and inform future conservation strategies for both lakes and adjacent watershed areas.

As a first step toward addressing these information gaps, we have successfully developed an integrated modeling framework (Vitense et al. 2016) using Bayesian latent variable regression (BLR) models to: 1) classify attracting lake states (clear vs. turbid); 2) estimate deterministic steady state relationships between total phosphorus (TP) and chlorophyll *a* (Chla); and 3) identify critical TP thresholds that differentiate highly resilient clear lakes, lakes that can transition between clear and turbid states following perturbations, and highly resilient turbid lakes. However, our previously developed BLR model assumes each lake has been sampled only once, but it is common for researchers to have 1-3 years of data for a set of lakes.

Several possibilities exist for handling multiyear data for a population of lakes within our framework: 1) within-lake observations could be assumed to be independent after conditioning on TP and state, and data could simply be pooled (similar to Wang et al. (2014)); 2) the BLR model can be fit separately to each year of data and then summarized across years (similar to Zimmer et al. (2009)); 3) correlated errors for repeated lake measurements can be built into the BLR model; e.g., a multivariate normal distribution could be used to describe the distribution of Chla with state-dependent within-lake correlated errors specified via a non-diagonal variance-covariance matrix; 4) a hierarchical approach can be employed with lake-level regression coefficients assumed to be random variables arising from a population-level distribution; 5) state transitions could be built into the model to create a hidden Markov model for individual lake dynamics. The most appropriate approach will likely depend on the data and underlying research questions or intended use of the model.

We illustrate options 2 and 4 here – i.e., we summarize separate model fits to each year of data, and we also fit a model that includes random intercepts in the logistic regression between SAV and latent state. We discuss advantages and disadvantages of both approaches, but we ultimately find that threshold estimates and conclusions for our set of lakes in Minnesota are similar for both approaches.

METHODS

Data

The MDNR surveyed 130 lakes once in July during each of three consecutive years, 2009-2011. Measures of TP (µg/L), Chla concentration (µg/L), and SAV abundance (kg/sample) were obtained in each year. Nine lakes were sampled in only one or two years, and all lakes had maximum depths less than 5 m. Water samples for TP were collected at two stations in each lake-year and frozen until analysis with persulfate digestion and ascorbic acid colorimetry. Two samples for Chla were collected at the same time and place as TP by filtering water through GF/F filters. The filters were frozen until analysis for Chla by acetone extraction and flourometric analysis. The average Chla and TP values for each lake-year were used for analysis.

Submersed aquatic macrophytes were sampled with a weighted plant rake using methods modified from Deppe & Lathrop (1992). Plants were sampled at 15 stations in each lake by dragging the rake across 3 m of lake bottom and weighing plant biomass (wet weights) collected on the rake. The average plant biomass across the 15 stations for each lake-year was used for analysis.

Bayesian Latent Variable Regression (BLR) Model

Our BLR model describes relationships between the natural logarithms of TP and Chla with linear models with state-dependent intercepts, slopes, and normally distributed errors (Equations 1-3). Lake state (*S_i*) is estimated as a latent variable that follows a Bernoulli distribution (Equation 4). The probability that lake *i* is in the turbid state (denoted by S_i=1) depends on both its TP and SAV values (Equation 5). If the lake's TP level falls below the lower TP threshold (π_1 on the log scale), its probability of being turbid is 0; i.e., the lake is classified as clear. If the lake's TP level falls above the upper TP threshold (π_2 on the log scale), its probability of being turbid is 1; i.e., the lake is classified as turbid. If the lake's TP level falls between the thresholds, logistic regression is used to model its probability of being turbid as a function of SAV abundance.

$$\log(Chla_i) \sim N(\mu_i, \sigma_i^2) \tag{1}$$

$$\mu_i = a_0 + \tau S_i + b_0(1 - S_i)\log(TP_i) + b_1 S_i \log(TP_i) \tag{2}$$

$$\sigma_i = \sigma_0(1 - S_i) + \sigma_1 S_i \tag{3}$$

$$S_i \sim Bern(p_i),\ \mathrm{S}_i = \begin{cases} 0, & \text{if lake } i \text{ is clear} \\ 1, & \text{if lake } i \text{ is turbid} \end{cases} \tag{4}$$

$$p_i = P(\text{turbid}) = \begin{cases} 0, & \text{if } \log(TP_i) < \pi_1 \\ \operatorname{logit}^{-1}(\gamma_0 + \gamma_1 \times SAV_i), & \text{if } \pi_1 \le \log(TP_i) \le \pi_2 \\ 1, & \log(TP_i) > \pi_2 \end{cases} \tag{5}$$

We chose priors that ensured the slopes describing the relationships between Chla and TP were positive and that the probability of a lake being turbid decreased as its abundance of SAV increased. All other priors were weakly informative:

$$\begin{gathered} a_0 \sim \mathcal{N}(0,10^2); \quad \tau \sim \mathcal{N}(0,3.16^2); \quad b_0,\ b_1 \sim Unif(0,10);\ \ \sigma_0,\ \sigma_1 \sim Unif(0,20) \\ \gamma_0 \sim \mathcal{N}(0,10^2);\ -\gamma_1 \sim \ln\mathcal{N}(0.5,1);\ \pi_1,\ \pi_2 \sim Unif(0,6.5) \end{gathered} \tag{6}$$

Finally, we included a constraint to force the line connecting the turbid line at $\ln(TP_i) = \pi_1$ to the clear line at $\ln(TP_i) = \pi_2$ to have a negative or flat slope to reflect the "S"-shape of Figure 1:

$$(\tau + b_1\pi_1 - b_0\pi_2)/(\pi_1 - \pi_2) \le 0 \tag{7}$$

Fitting and Summarizing Separate Model Fits

We fit the BLR model above to each of the three years of Minnesota shallow lake data separately. We ran the models in JAGS (Plummer 2003) using the R package 'R2jags' (Su and Yajima 2015). For each year, we ran three chains for 10,000,000 iterations with a burn-in of 2,000,000 and thinning rate of 2,400. We examined convergence using trace plots and the Gelman-Rubin convergence statistic (Gelman and Rubin 1992). Within each year, we classified a lake as turbid (clear) if over half of the sampled states from the Markov chain Monte Carlo (MCMC) chains were turbid (clear) for that lake. We estimated regression coefficients and TP thresholds using medians and modes of the posterior distributions, respectively, and computed 95% credible intervals for the regression coefficients and TP thresholds for each year.

We summarized the fits across the 3 different years in a heat map of the state classifications in all 3 years, and we used median TP threshold estimates across the 3 fits as overall TP threshold estimates.

Random Parameter Extension to BLR Model

Our BLR model is flexible, and random parameters can be incorporated to account for correlation among repeated measurements or to allow certain relationships to vary among lakes. We highlight a model that includes random intercepts in the logistic regression describing how the probability a lake is turbid changes with SAV (Equation 12). This model formulation reflects that the lakes' inherent chance of being turbid in the bistable region may vary because of factors not accounted for here (e.g., zooplankton community characteristics), but changes in SAV abundance are assumed to have the same effect on the probability of being turbid for all lakes (i.e., γ_1 in Equation 12 is a fixed effect). The random logistic intercept model we fit is formulated as follows, where *i, j* denotes the *j*th observation from lake *i*:

$$\log(Chla_{i,j}) \sim N(\mu_{i,j}, {\sigma_{i,j}}^2) \tag{8}$$

$$\mu_{i,j} = a_0 + \tau S_{i,j} + b_0(1 - S_{i,j})\log(TP_{i,j}) + b_1 S_{i,j}\log(TP_{i,j}) \tag{9}$$

$$\sigma_{i,j} = \sigma_0(1 - S_{i,j}) + \sigma_1 S_{i,j} \tag{10}$$

$$S_{i,j} \sim Bern(p_{i,j}),\ \mathrm{S}_{i,j} = \begin{cases} 0, & \text{if observation } j \text{ for lake } i \text{ is clear} \\ 1, & \text{if observation } j \text{ for lake } i \text{ is turbid} \end{cases} \tag{11}$$

$$p_{i,j} = P(\text{turbid}) = \begin{cases} 0, & \text{if } \log(TP_{i,j}) < \pi_1 \\ \operatorname{logit}^{-1}(\gamma_{0,i} + \gamma_1 \times SAV_{i,j}), & \text{if } \pi_1 \le \log(TP_{i,j}) \le \pi_2 \\ 1, & \log(TP_{i,j}) > \pi_2 \end{cases} \tag{12}$$

$$(\tau + b_1\pi_1 - b_0\pi_2)/(\pi_1 - \pi_2) \le 0 \tag{13}$$

$$\begin{aligned} &a_0 \sim \mathcal{N}(0, 10^2); && \tau \sim \mathcal{N}(0, 3.16^2); && b_0,\ b_1 \sim Unif(0, 10) \\ &\sigma_0,\ \sigma_1 \sim Unif(0, 20); && \pi_1,\ \pi_2 \sim Unif(0, 6.5); && -\gamma_1 \sim \ln\mathcal{N}(0.5, 1) \\ &\gamma_{0,i} \sim \mathcal{N}(\mu_{\gamma_0}, \sigma^2_{\gamma_0}); && \mu_{\gamma_0} \sim \mathcal{N}(0, 10^2); && \sigma_{\gamma_0} \sim Unif(0, 10); \end{aligned} \tag{14}$$

We ran the random logistic intercept model in JAGS, examining the model for convergence. We computed parameter estimates and state classifications using the same approach outlined above. Chains were run for 1,000,000 iterations with a burn-in of 200,000 and thinning rate of 240.

RESULTS

The BLR model produced reasonable fits to each of the 3 years of shallow lake data treated separately and together (Figure 2). The fitted models resemble bifurcation diagrams with no evidence for lack of convergence.

The separate fits to the 3 years of data depict sampling variability between years. The heat map of state classifications across the 3 years (Figure 3) suggests that an approximate Chla threshold of 20 μg/L separates clear and turbid lakes in the bistable region. Indeed, roughly equal proportions of clear and turbid lake-years in the bistable region are divided by a Chla threshold of 19.9 μg/L. Across all lake-years, 53.7% of lake-years fall below the median lower TP threshold (49.97 μg/L), 43.7% fall in the bistable region, and 2.6% fall above the median upper TP threshold (366.36 μg/L). Across the 3 fits, 64.6% of lakes were classified as clear in all available years, 15.4% were turbid in all available years, and 20% transitioned at least once.

TP threshold posterior distributions and credible intervals are narrower for the random logistic intercept model compared to the separate fits (Table 1, Figure 2). We note that the upper TP threshold estimate for the random logistic intercept model is reduced to ~355 μg/L from ~437 μg/L if one influential observation from year 2009 is removed. For the random logistic model, the estimated unstable line ranges over Chla levels 15.0-19.9 μg/L. Additionally, roughly equal proportions of clear and turbid lake-years in the bistable region are divided by a Chla value of 19.8 μg/L, which is similar to the Chla threshold estimated using the 3 separate fits. For TP threshold estimates from the random logistic intercept model, 50.3% of lake-years fall below the lower threshold estimate (44.29 μg/L), 47.9% fall in the bistable region, and 1.9% fall above the upper threshold estimate (437.03 μg/L). Additionally, 65.4% of lakes were clear in all available years, 13.8% were turbid in all available years, and 20.8% transitioned at least once for classifications from the random logistic intercept model. These proportions are similar to those using classifications and median threshold estimates from the separate fits to the 3 different years.

DISCUSSION

Our Bayesian latent variable model (BLR) provides a formal modeling framework that can be adapted to allow for additional data features, such as repeated measures or more extensive time series. We illustrated both how separate model fits across different years can be summarized, as well as how multi-year data can be aggregated and the model extended to incorporate random parameters. The hierarchical modeling approach has the advantage of providing a single model fit for multi-year data and narrower TP threshold posterior distributions compared to the separate yearly model fits. However, summarizing across separate yearly fits reduces the influence of outlying data points and elucidates sampling variability between years. Future researchers may decide whether and which random parameters are appropriate for inclusion given their data and study systems.

Critical threshold estimates were similar, regardless of which approach was used. Chla levels of 19.8-19.9 μg/L separated clear and turbid lakes in the estimated bistable region into roughly equal proportions of clear lakes falling below and turbid lakes falling above these values for both approaches. The lower TP threshold estimates were also similar. The upper threshold estimate of the random logistic BLR model is similar to the median estimate of the separate yearly fits if one influential observation from 2009 is removed when fitting the hierarchical model; otherwise, the upper threshold for the hierarchical model is similar to the estimate in year 2009.

These threshold estimates provide important information to help managers make decisions about whether and how to treat different shallow lakes, and also help to define realistic expectations when attempting to rehabilitate a lake. Shallow lakes with TP levels below the lower estimated critical TP threshold may be deemed high priority clear lakes, with efforts focused on protecting adjacent watersheds or other features contributing to their pristine conditions. Indeed, the majority of the lakes in our study are highly stable clear lakes. On the other end, lakes with TP levels that are frequently above the upper TP threshold can be considered lower priority turbid lakes. The internal P loads can be so great in these lakes because of historically high nutrient inputs that the lakes will persistently return to turbid water conditions following management actions (Hobbs et al. 2012, Hanson et al. 2017, Ramstack Hobbs et al. 2016). Fortunately, a very low proportion of lakes in our study fall in this category. Finally, lakes that tend to fall in between the two thresholds are those for which active management is likely to be most practical, and our results suggest that 44-48% of the lake-years in our study fell in the bistable region. Lakes that tend to exist in this bistable region are highly dynamic, and managers may force these lakes from the turbid to clear stable state through actions such as biomanipulation of fish stocks or water level drawdowns. Additionally, the relative resilience of different lakes in the bistable region can be used to help prioritize lakes for management. For example, lakes can be placed on an estimated bifurcation diagram, and resilience can be estimated by each lake's proximity to TP thresholds or as the estimated distance between a lake's steady state and the unstable line or critical Chla threshold.

Finally, the BLR framework can be modified to include state transitions in which the trajectories of lakes crossing thresholds are directly modeled, which would likely allow for better identification of critical nutrient thresholds. Future research will be focused on model extensions for lake transitions to understand key factors driving changes to lake nutrient levels and top-down influences (e.g., fish and invertebrates) that drive regime shifts. These models will help to further refine predictions regarding which lakes are most likely to undergo successful rehabilitations and help to prioritize lakes for management.

ACKNOWLEDGMENTS

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Table 1. Estimated total phosphorus (TP) thresholds with 95% credible intervals from the fit of Bayesian latent variable regression models to 3 different years (2009-2011) of shallow lake data collected in July in Minnesota, USA. These thresholds determine which lake states are possible for a specific value of TP (only the clear state is possible when TP is less than the lower threshold, only the turbid state is possible when TP is higher than the upper threshold, and either state is possible for TP values between the 2 thresholds). The model fit to all 3 years of data included random logistic intercepts for each lake.

Year	Lower TP threshold (µg/L)	Upper TP threshold (µg/L)
2009	96.03 (41.87, 107.82)	434.93 (410.94, 644.15)
2010	49.97 (32.61, 124.21)	350.01 (330.02, 639.69)
2011	30.27 (19.22, 50.97)	366.36 (341.68, 622.97)
All Years	44.29 (42.30, 52.29)	437.03 (410.42, 480.20)

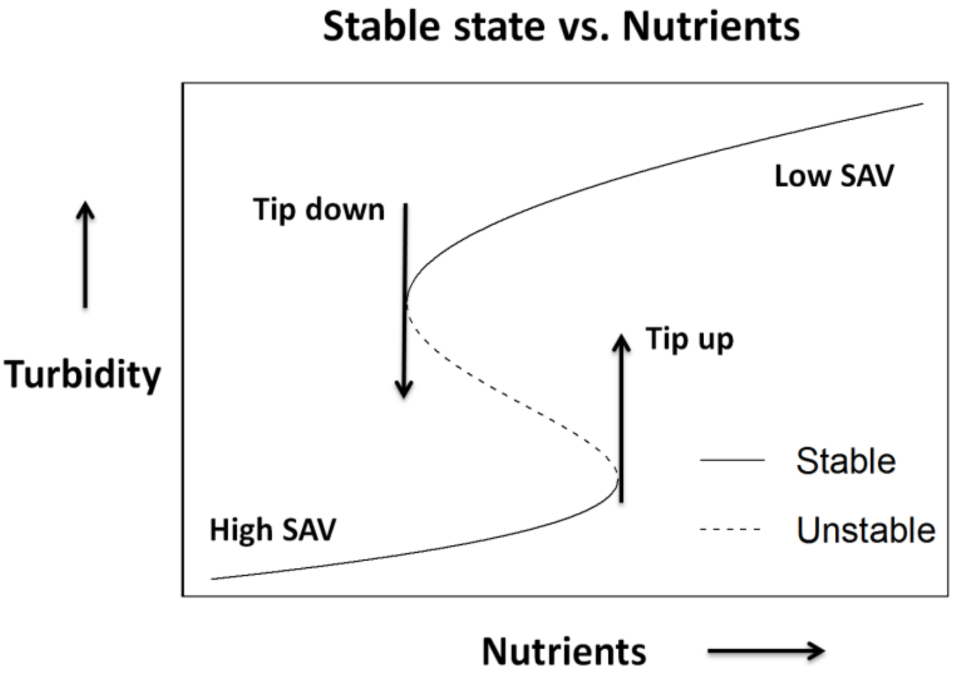

Figure 1. Bifurcation diagram from a theoretical model describing shallow lake dynamics. At low nutrient levels (left of “tip down” threshold), only the clear stable state exists (lower solid line). At high nutrient levels (right of the “tip up” threshold), only the turbid stable state exists (upper solid line). In between the 2 thresholds, 2 different stable states are possible under the same nutrient conditions, depending on whether initial turbidity levels lie above or below the unstable state (dotted lie) in this region of bistability.

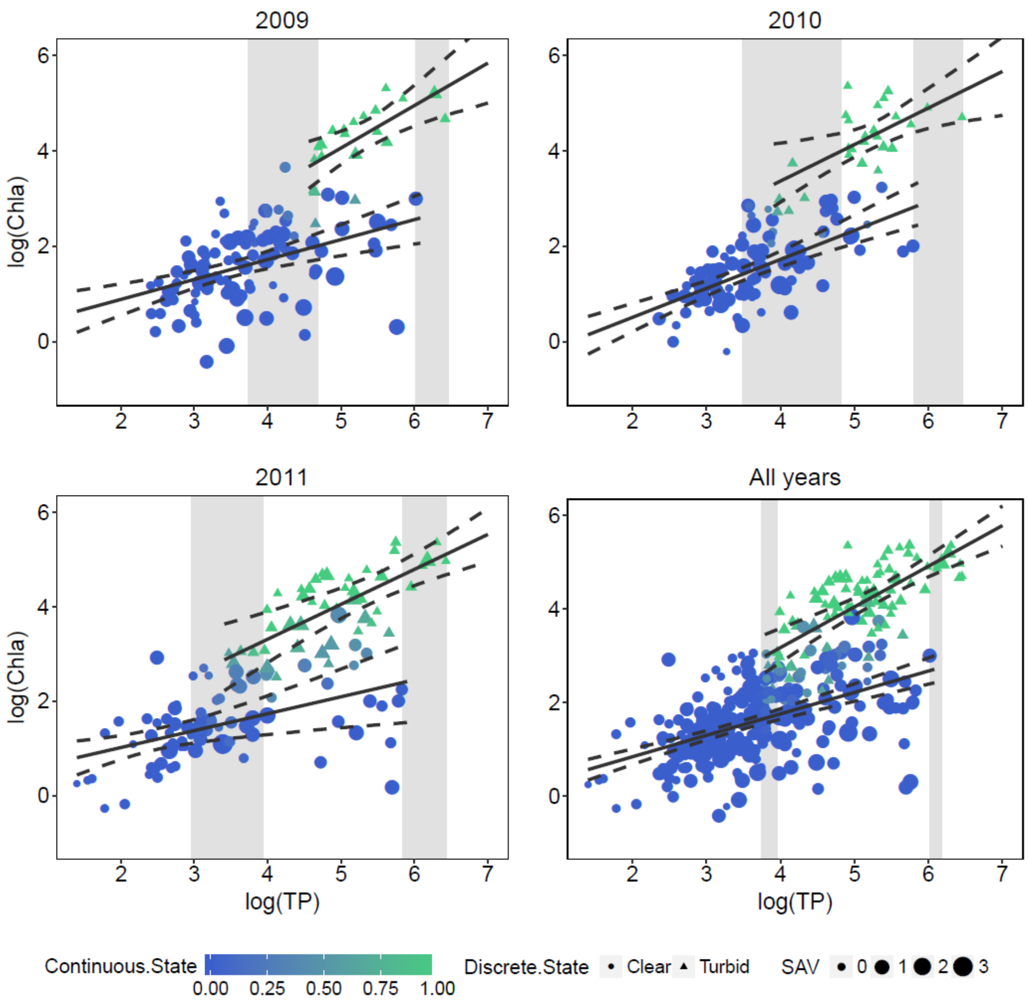

Figure 2. Bayesian latent variable regression (BLR) estimated steady state relationships between total phosphorus (TP) and chlorophyll *a* (Chla) for 3 different years (2009-2011) of shallow lake data collected in July in Minnesota, USA. Black solid (dashed) lines represent average (2.5th, 97.5th quantiles) estimated steady state relationships across all Markov chain Monte Carlo (MCMC) samples. Steady state lines end at the TP threshold point estimates, and gray bands represent 95% credible intervals for TP thresholds. Circular points represent lakes classified as clear (>50% of MCMC sampled states were clear), and triangular points represent lakes classified as turbid (>50% of MCMC sampled states were turbid). The average MCMC sampled state for each lake is shown on a blue to green color gradient (0=clear, 1=turbid). Point size is proportional to submerged aquatic vegetation (SAV, units: average kg/sample). The model fit to all 3 years of data included random logistic intercepts for each lake.

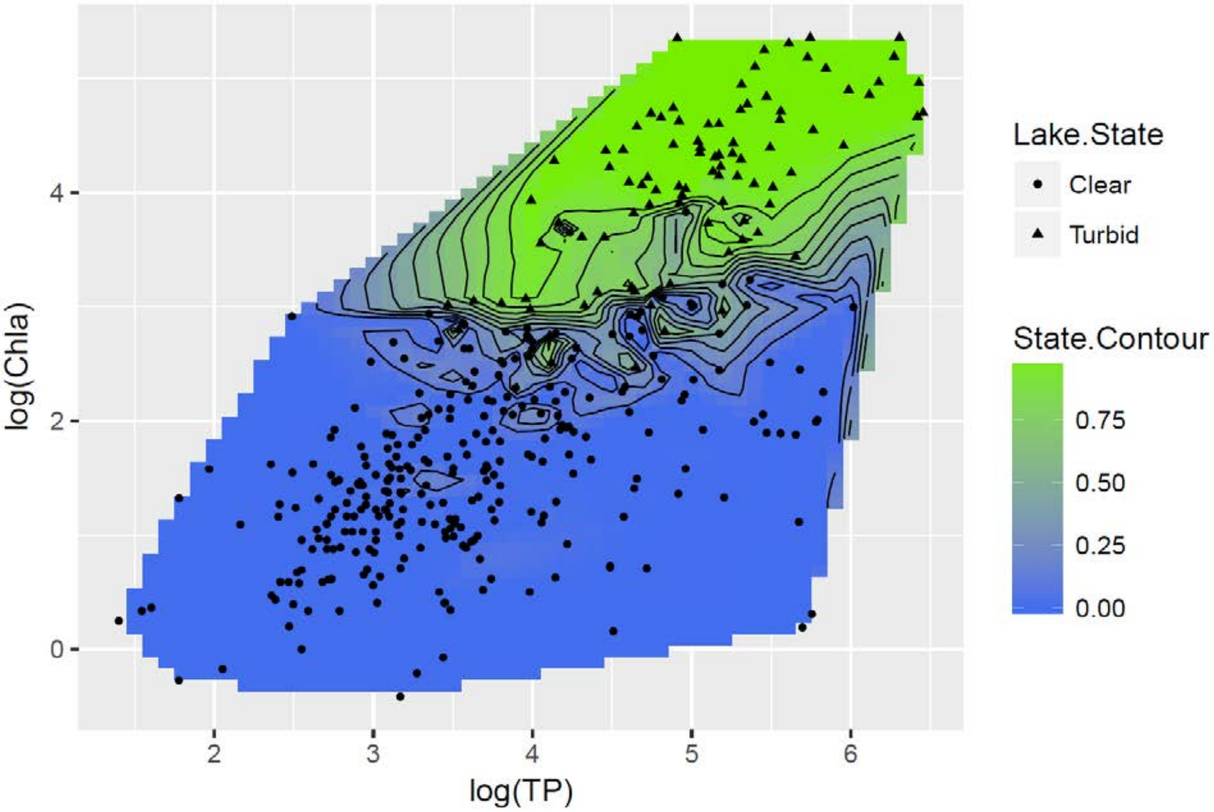

Figure 3. Heat map of all lake-year state classifications from separate fits to 3 different years (2009-2011) of shallow lake data collected in July in Minnesota, USA. Lakes are more frequently classified as clear (turbid) in blue (green) regions, where 0=clear and 1=turbid. The heat map was created using each lake's state on a continuous scale from 0-1 representing the proportion of Markov chain Monte Carlo (MCMC) samples in which the

FALL MOVEMENTS OF MALLARDS MARKED IN MINNESOTA

Bruce E. Davis

SUMMARY OF FINDINGS

During August-September of 2016, I marked 119 mallards (anas platyrhynchos) with tracking units. I obtained GPS locations from dataloggers recovered by hunters or uploaded through the Argos satellite system to yield 3506 locations on 44 birds. Locations within the state of Minnesota (n=2848) were used to examine habitat use. Marked birds were retained in Minnesota longer than expected; freeze up dates were later than average in 2016. When marked birds did leave the state, movements upon departure tended to be long with a mean distance 434 km between a bird’s last known location in Minnesota and its first known location outside of Minnesota. Marked birds used open water and emergent herbaceous wetland habitats for combined 55-80% of the time. Crop habitats were used most frequently at night (30% of proportional use) and more frequently for birds marked in the south hunting zone than for birds marked in the north hunting zone. Sample sizes were sufficient to detect differences in use of habitats among capture zones and time of day, but I did not detect differences in emigration rates between zones; given the late onset of winter, rates of emigration may have been similar between zones. For birds marked in 2016, biologically relevant differences were detectable when present.

INTRODUCTION

Distribution of waterfowl during fall migration and concurrent hunting seasons is affected by numerous factors. Wildlife managers are tasked with setting season dates, bag limits, shooting hours, and further restrictions on harvest. Availability of waterfowl throughout the hunting season (retention) is important to Minnesota waterfowl hunters. Understanding the chronology of immigration and emigration events and the factors affecting those events is imperative.

Many factors may impact emigration rates and use of habitats. Weather plays an important role in the timing of migration by waterfowl during fall; as winter weather severity increases, the probability of southward waterfowl migration also increases (Schummer et al. 2010). Repeated exposures to disturbance associated with hunting have been found to alter the distribution and habitat use and cause increased movements of wintering waterfowl (Dooley et al. 2010, Pease et al. 2005), but the effects of disturbance have not been investigated for waterfowl nearer their breeding habitats. Importantly, the effects of weather and anthropological disturbance are likely confounded; hunting seasons often coincide with changing weather patterns. In the presence of elevated human disturbance to waterfowl habitats that occurs during hunting seasons, it may be difficult to detect causes of temporal or spatial changes to a bird’s natural migration pattern. Numerous studies have been implemented to understand aspects of breeding waterfowl and some information is available on wintering waterfowl, but little work has been completed on waterfowl during migration periods. Due to their transient nature, waterfowl are inherently difficult to study during the migration periods. Thus, few studies have been undertaken to investigate patterns of fall migration.

In an effort to provide habitat to local and migrating waterfowl, retain waterfowl on the landscape throughout the duration of the season, provide hunting opportunities for its constituents, and to control waterfowl harvest, the Minnesota Department of Natural Resources has implemented numerous restrictions on duck harvest and disturbance to wetlands. Restrictions include establishment of waterfowl refuges, a 4 PM closure to duck hunting for the earliest portion of the duck season, designation of feeding and resting areas which restrict the use of motorized boats, a statewide ban on motorized decoys for the earliest portion of the season, and a ban on motorized decoys on state owned Wildlife Management Areas for the entire season.

The importance of the hunting regulations implemented in Minnesota to provide secure areas for ducks is unclear because fall emigration and factors affecting the chronology of fall migration are poorly understood. Restrictions on afternoon shooting hours unilaterally in Minnesota did result in 3-4% lower recovery rates (a proxy for harvest rates) than when sunset closures occurred, but the researchers were unable to detect a difference in annual survival rates (Kirby et al. 1983). Restrictions on shooting hours that are more restrictive than what is allowed in the federal framework have been in place since 1973 but their importance is unknown. Assessment of the effects of shooting hour restrictions and other hunting regulations on movement patterns warrants investigation. Better understanding of movement patterns gained from this work will allow managers to better set season dates and alter restrictions on harvest.

OBJECTIVES

Overall study objectives were to:

1. Better understand emigration chronology for mallards in Minnesota.
2. Estimate distances and directions moved by mallards in Minnesota.
3. Identify migration stopovers used by mallards in Minnesota.
4. Estimate use of habitats for birds while in Minnesota.

More specifically, during the pilot-year of this study, we seek to inform subsequent years of data collection by addressing these specific objectives:

5. Estimate variability in emigration, movement, and habitat use data within and among hunting zones.
6. Estimate rate of sample size reduction throughout the tracking period.
7. Evaluate alternative tracking units in terms of data quantity and quality.

STUDY AREA

Currently, Minnesota utilizes 3 zones to manage duck hunting seasons (Figure 1). Timing of seasons and restrictions on shooting hours differ among the zones. We attempted to mark equal numbers of birds in each hunting zone, but were unable to mark birds in the central zone.

METHODS

Marking

We attached 39 GPS-Argos backpack units (Lotek Wireless Inc., Newmarket, Ontario, Canada) to adult female mallards; these units log GPS data and then transmit that data back to the Argos system upon completion of their duty cycle. These units were 15 g and able to record about 100 GPS fixes and transmit those fixes to Argos satellites before exhausting their battery life.

Additionally, we marked 80 hatch year male mallards with GPS-archival backpack units (Lotek Wireless Inc.). These units record GPS location data at a user specified interval, but must be recovered to acquire data. These units weighed 11 g and were configured as backpack type transmitters. I selected hatch year males because they have the highest recovery rate of any mallard age-sex cohort. Apparent direct (within first hunting year after marking) recovery of hatch year male mallards banded in Minnesota based on band returns was predicted to be 18% and an additional 6% were expected to be recovered in the 2nd hunting season after deployment (USGS, Gamebirds data set).

GPS-logger or GPS-Argos backpack transmitter units receive satellite signals to estimate highly accurate locations; precision of locations is accurate to within a few meters. Of all available options, these units were deemed best suited for estimating detailed parameters associated with habitat use, use of refuge areas, local movements, and migration events. Birds were marked in conjunction with our current banding effort. We paid a $50 incentive for hunters returning tracking units.

Tracking

GPS-logger units were configured to attain location data every 11.5 hours; GPS-Argos units were configured to attain fixes every 22.5 hours and the units were set to begin this cycle at differing times. This allowed locations throughout the day and locations on each individual bird to shift over days and attain day and night fixes accordingly.

Movement Data

Estimated point locations were determined to be inside or outside the state of Minnesota. A bird was determined to have emigrated upon its permanent exit from the state. Movement direction was measured as the azimuth between the birds marking location and its first location outside the state upon permanent emigration.

Use of Habitats

Estimated point locations were overlaid on the 2011 National Land Cover Data layer and habitats were determined based on estimated point locations. Similarly, it was determined whether locations were on refuge or non-refuge locations and WMA or non-WMA locations based on appropriate GIS data layers.

Data Analyses - Movement Data

I determined date of permanent departure from the state (emigration) for each bird based on its location data. I used proportional hazards regression (Allison 1995) to examine variation in emigration rates due to the effects of the bird’s age and sex or its location of marking. I present product-limit emigration estimates (Kaplan and Meier 1958) for the marked sample. Further, I present a plot latitude of location data over time and a plot of the array of movement direction upon emigration from Minnesota.

Data Analyses - Use of Habitats

I divided the tracking period into 3 time periods based on hunting seasons: PREHUNT (the period before regular duck season was opened in Minnesota), HUNT (the period when regular hunting season was open anywhere in Minnesota), and POST (the period after regular duck season had closed anywhere in Minnesota). I divided location data in portions of the day as diurnal (sunrise to sunset) or nocturnal. I collapsed habitats to five basic categories for analysis including open water, forested or developed habitats, pasture habitats, crop habitats, or emergent marsh habitats. I determined use of habitats within the state of Minnesota based on 2848 location estimates from 44 birds using compositional analyses (Aebischer et al.1993). I determined diurnal and nocturnal proportional use of each bird in every habitat during each time period, I replaced zero values with 0.002 (an order of magnitude lower than the lowest nonzero proportion of a habitat used by any bird in a combination of any time period and portion of day. To remove the unit sum constraint, I constructed log ratios by dividing proportional use of each habitat by proportional use of emergent marsh habitat and used Napierian logarithms of these ratios as response variables. I used split-plot, repeated measures multivariate analysis of variance to test for overall effects of season (PREHUNT, HUNT, POST), portion of day (day or night), cohort of marked bird, or zone of capture. I fit a full model containing all 4 of these explanatory factors as well as a term for repeated measures among birds.

Models were fit using backwards-stepwise procedures. I present estimates of proportional use of each habitat, averaged across birds, from the untransformed data within levels of significant ($P \leq 0.050$) explanatory variables from the final fitted model.

Similarly, I examined use of refuge areas (areas closed to waterfowl hunting by statute or regulation) and use of WMAs using analysis of variance after constructing proportions as outlined above. I present proportional use of these habitats below.

RESULTS

Movement Data

I did not detect differences in rates of emigration among cohorts or zones of capture ($Ps>0.018$). Retention rates of marked birds in Minnesota remained > 80% until early November then declined to about 45% by mid-November, remaining birds left the state in early-December (Figure 2). Latitudes of marked birds declined throughout the season (Figure 3) and vectors of emigration flights were mostly long and southeasterly (Figure 4).

Use of Habitats

I did not detect differences among proportional use of habitats by seasons or cohorts ($Ps >0.090$), but proportional use of habitats differed among zones of capture ($P=0.018$) and portion of day ($P < 0.0001$). Use of crop habitats were higher for birds marked in the south capture zone than for birds marked in the north capture zone (Figure 5). Use of open water habitats were highest during the day; use of crop habitats were highest during the night (Figure 6).

Use of areas closed to hunting varied by zone of marking and season ($Ps<0.0029$). Proportional use of areas closed to hunting were 45% in the north hunting zone, whereas use of refuge areas was only 8% in the south hunt zone. Use of refuge areas was highest (44.9%) during the preseason period, but decreased to 22.6% and 27.3% during the hunting season and post-hunting periods, respectively.

Use of WMAs was 56%, 37%, and 27% during the preseason, hunting season, and post-hunting seasons, respectively. Use of WMAs during night was 39%, but 53% during the day.

DISCUSSION

Emigration rates were similar between zones of capture and cohorts; given the late onset of winter that occurred in 2016, this was not surprising. Temperatures were above normal through early December in northern Minnesota. When freeze up did occur in the north hunting zone it also occurred in much of the southern portion of the state shortly thereafter. I speculate that the extended retention time of the marked sample within Minnesota was likely due to the late onset of winter.

Use of open water and emergent wetland habitats was high; these estimates were based on the National Land Cover Database data currently available. More refined analyses of habitat use could be conducted if more informative and accurate GIS data layers are available.

ACKNOWLEDGMENTS

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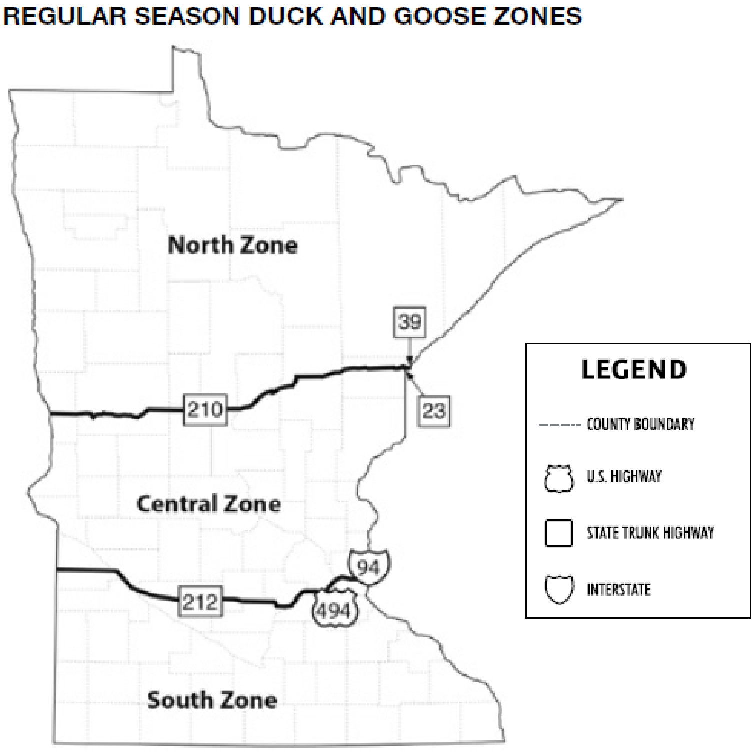

Figure 1. Minnesota waterfowl hunt zones boundaries, 2016.

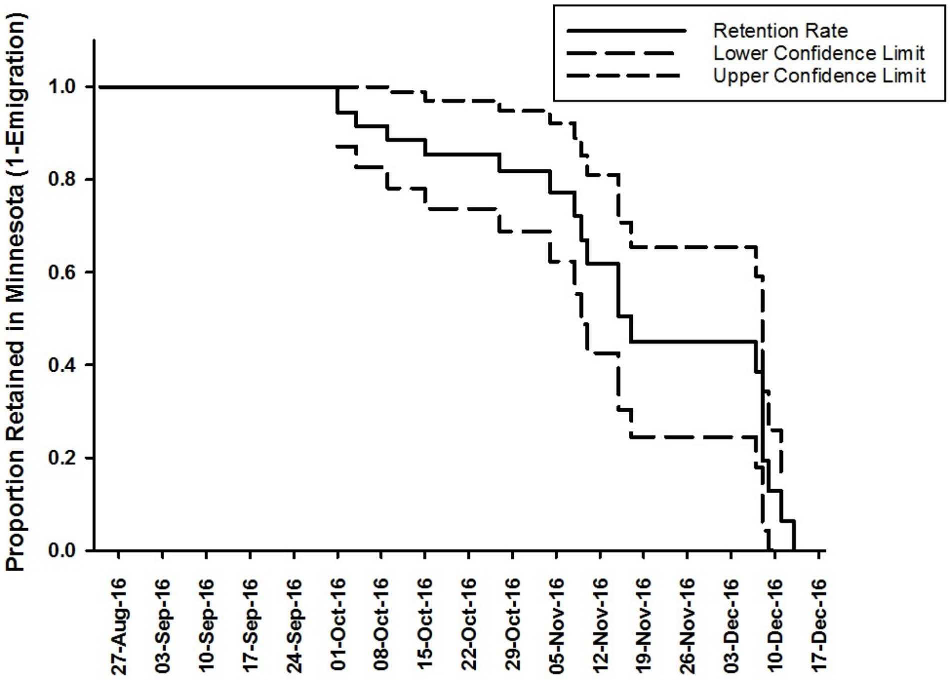

Figure 2. Retention curve for mallards marked with tracking units in Minnesota, 2016.

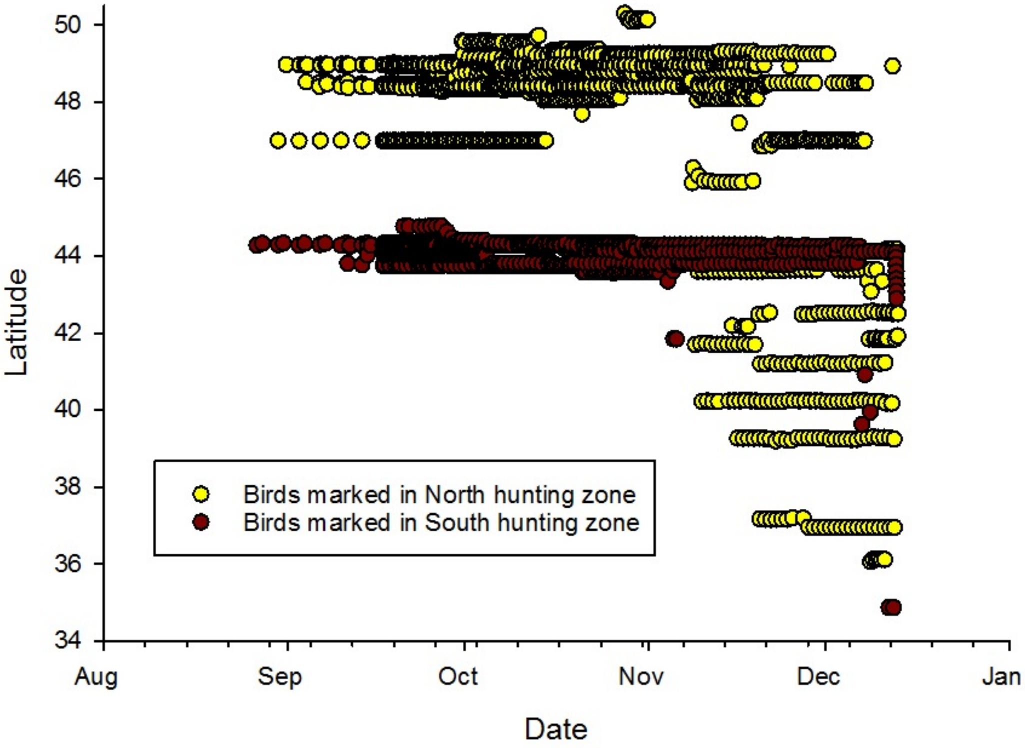

Figure 3. Scatter plot of location latitudes by date for mallards marked with tracking units in Minnesota, 2016.

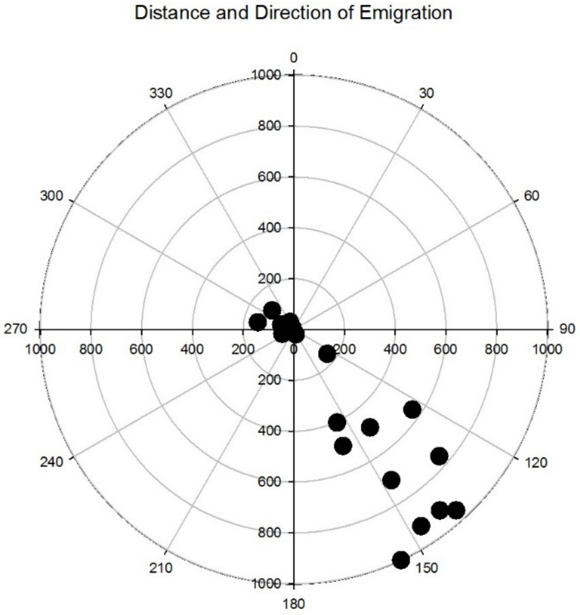

Figure 4. Polar plot of distance and direction of movement on permanent emigration (black dots) for mallards marked with tracking units in Minnesota, 2016. Concentric rings represent distances (km); azimuth (degrees) of movements are indicated on the outermost ring of the plot.

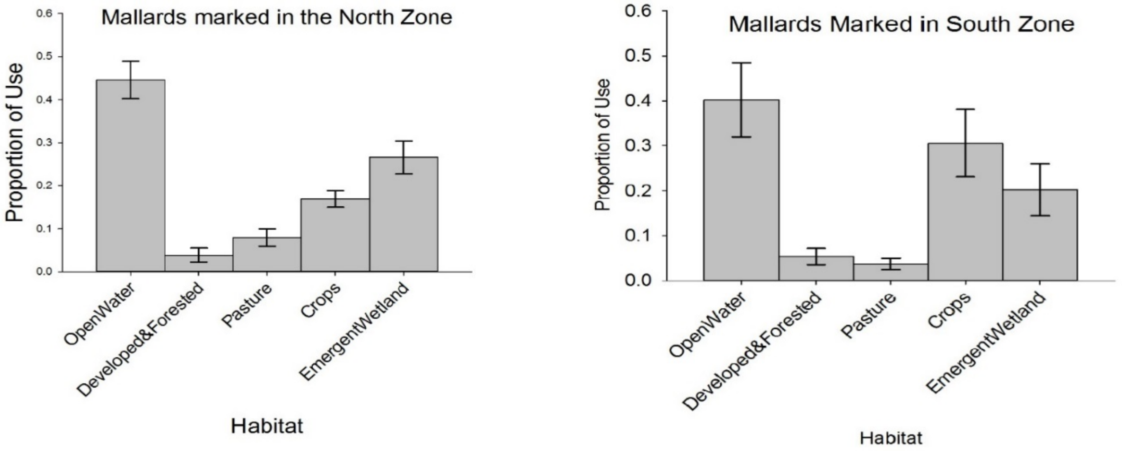

Figure 5. Proportional use of habitats by mallards marked in the Minnesota's north and south hunting zones, 2016. Proportions are expressed as an average across birds.

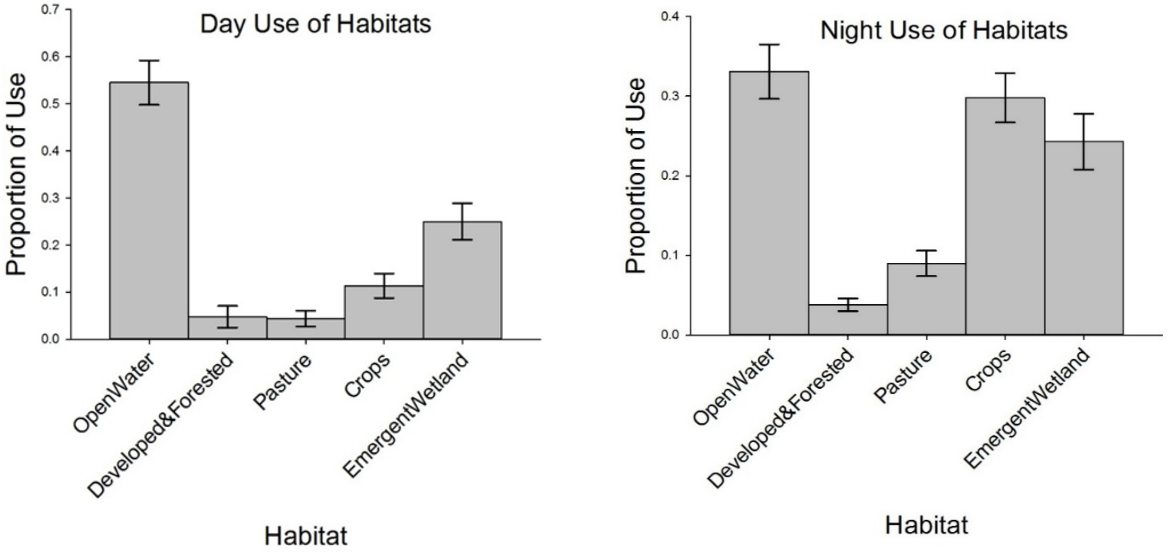

Figure 6. Proportional day or night use of habitats by mallards marked in the Minnesota's north and south hunting zones, 2016. Proportions are expressed as an average across birds.

DEVELOPING METHODOLOGIES FOR PREDICTING THE LOCATIONS OF WOOD DUCK BREEDING HABITAT COMPONENTS IN MINNESOTA

SUMMARY OF FINDINGS

There have been alterations to both aquatic and terrestrial habitats used by wood duck (*Aix sponsa*) hens and broods in Minnesota and the Upper Midwest. We initiated this study to develop methodologies that can be used to predict the locations and monitor spatiotemporal changes in the areal extent of wood duck breeding complexes. Specifically, we want to develop Light Detecting and Ranging (LiDAR) as a method to identify multiple habitat components and to monitor changes in these components from the contemporary period forward. We will provide better historical context regarding spatiotemporal changes in nesting habitat by analyzing Forest Inventory and Analysis (FIA) data with a quantitative method currently being developed. Our specific objectives are to (1) develop and evaluate spatial predictive models of habitat components that are important to breeding wood ducks (i.e., tree species [alternatively deciduous v. coniferous], diameter-at-breast height [DBH], tree canopy density, stand type, wetland type, water depth) based on LiDAR-generated metrics or other sources of spatial data (e.g., National Wetland Inventory [NWI], existing GIS layers, aerial photographs), (2) ascertain the optimal pulse density of LiDAR needed to accurately measure or classify each habitat component of importance to wood ducks (3) determine the generalizability of the LiDAR method for predicting the locations of habitat components by applying algorithms developed from data collected in the main study area (Cass County, Forest Ecological Province) to other sites in the Forest, Prairie, and/or Transition Provinces at which adequate LiDAR-cloud data have been obtained, (4) estimate the species- and DBH-specific proportions of trees with suitable cavities and detection probability of suitable cavities from empirical field data, and (5) determine whether there has been a change in the number of potential nest trees since the 1970s based on changes in FIA data.

We conducted vegetation surveys at 185 wetland plots during Summer 2016 and 152 forest plots during Fall 2016 and Spring 2017. Preliminary results suggest that the proportion of trees with suitable cavities varied by species, DBH class, and health status. Flights to collect LiDAR data were scheduled to occur during Fall 2016, but were postponed until Fall 2017. Thus, we could not associate ground-level vegetation data to LiDAR data. We will collect wetland and forest surveys and analyze FIA data during much of the latter half of 2017, and associate ground-level and LiDAR data starting during late Wiinter 2018.

INTRODUCTION

Some terrestrial and aquatic habitats used by wood duck hens and broods during the pre-nesting, nesting, and brood-rearing life-cycle phases have been altered substantially in Minnesota and the Upper Midwest. For example, there were decreases in the areal extent of some classes of aquatic habitats in northcentral Minnesota (Radomski 2006) and in the number of beaver impoundments in the forested portion of Minnesota between the early 1990s and 2002 (Dexter 2002, p. 52), both of which were used by wood duck broods (see McGilvery 1968,

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Bellrose and Holm 1994). Although the number of potential nesting trees for wood ducks was projected to increase both in Minnesota (Jaakko Pöyry Consulting, Inc. 1994) and the Upper Midwest (Denton et al. 2012*b*), there has been recent concern among MNDNR managers that harvesting relatively large-DBH trees of economically valuable species (e.g., aspen) in northern Minnesota will reduce the availability of cavity trees frequently used for nesting by some waterfowl (R. A. Norrgard and D. P. Rave, MNDNR, personal communication).

The ultimate goal of this project is to develop methodologies that can be used to predict the locations of the habitat components that compose wood duck breeding complexes (i.e., important habitats used during the pre-breeding to brood-rearing life cycle phases). These methodologies should have the (A) flexibility to identify both forested and non-forested habitat components that occur at different spatial scales, (B) accuracy to reliably quantify spatiotemporal changes in the characteristics (e.g., areal extent) of habitat components and (C) efficiency to collect habitat data over large spatial scales. It also would be beneficial to develop methodology to analyze habitat data that were collected in long-term standardized surveys that likely will be performed in the future.

Meeting all of these needs with a single methodology or existing dataset probably is not possible. Consequently, we will develop 2 methodologies for obtaining better knowledge regarding spatiotemporal changes in wood duck breeding-habitat components. We propose to develop LiDAR methodology to identify multiple habitat components and to monitor changes in these components from the contemporary period forward. This methodology also could be used to provide habitat trend information that can be used in MNDNR administrative efforts (e.g., subsection planning) and research (e.g., estimating habitat availability in resource selection studies; see Aebischer et al. [1993]).

We also propose to provide better historical context regarding spatiotemporal changes in nesting habitat by analyzing FIA data with a quantitative method currently being developed. Reliable FIA surveys have been conducted since the 1970s. We propose to conduct analyses of FIA data to identify spatiotemporal changes in nesting habitat components not characterized by LiDAR, at spatial scales smaller than those of previous investigations, and over a greater time period (i.e., since the 1970s). This methodology also will provide database queries that can be used in future monitoring efforts, and an insight of whether the predicted trend in the abundance of tree cavities (e.g., Denton et al. 2012*b*) is accurate.

GOALS AND OBJECTIVES

The ultimate goal of this project is to develop methodologies that can be used to predict the locations and monitor spatiotemporal changes in the areal extent of wood duck breeding complexes (i.e., important habitats during the pre-breeding to brood-rearing life cycle phases). Meeting this goal will require that we (1) identify the location and areal extent of breeding-habitat components in the main study area, (2) validate the predicted locations of wood duck breeding complexes with independent, empirical data from other sites, and (3) quantify the spatiotemporal trends in potential nesting trees in Minnesota over the long term. We will meet this goal using 2 sources of data (i.e., LiDAR and FIA). Our specific objectives are to:

1. Develop and evaluate spatial predictive models of habitat components that are important to breeding wood ducks (i.e., tree species [alternatively deciduous v. coniferous], DBH, tree canopy density, stand type, wetland type, water depth) based on LiDAR-generated metrics or other sources of spatial data (e.g., NWI, existing GIS layers, aerial photographs). This evaluation will include determining the accuracy with which each component can be predicted with LiDAR-cloud data.
2. Ascertain the optimal pulse density of LiDAR needed to accurately measure or classify each habitat component of importance to wood ducks.

3. Determine the generalizability of the LiDAR method for predicting the locations of habitat components by applying algorithms developed from data collected in the main study area (Cass County, Forest Ecological Province) to other sites in the Forest, Prairie, and/or Transition Provinces at which adequate LiDAR-cloud data have been obtained (e.g., J. Erb's study areas, MNDNR statewide elevation measurement project).
4. Estimate the species- and DBH-specific proportions of trees with suitable cavities and detection probability of suitable cavities from empirical field data.
5. Determine whether there has been a change in the number of potential nest trees since the 1970s based on changes in FIA data.

METHODS

Wetland Surveys

Initially, we used the available spatial data from NWI (Cowardin et al. 1979) to select 260 sampling locations in the study area. Initially, we stratified wetlands contained in the NWI GIS layer by NWI system, subsystem, and class (hereafter, wetland types). Unfortunately, information about wetland subclasses was not available in this GIS layer. We then randomly selected 260 2- X 2-m plots from the 9 major wetland types present in the study area: 60 plots from both the Lacustrine-Littoral-Emergent Vegetation and Palustrine-Emergent Vegetation, and 20 plots each from the Lacustrine-Limnetic-Unconsolidated Bottom, Lacustrine-Littoral-Unconsolidated Bottom, Palustrine-Forested, Palustrine-Shrub Scrub, Palustrine-Unconsolidated Bottom, Riverine-Upper Perennial-Unconsolidated Bottom, and Riverine-Lower Perennial-Unconsolidated Bottom wetland types. We selected more plots from the first 2 wetland types because we surmised that these habitats were more likely to be used by wood duck broods (e.g., Grice and Rogers 1965), and that there was a greater likelihood that these habitats would be structurally diverse and thus more challenging to identify from LiDAR signatures. We also specified that plots had to be ≥100 m apart to reduce the likelihood of non-independence among plots (i.e., sampling plots with similar vegetation structure).

Many relatively small, isolated wetlands were not delineated in the NWI layer, so we later selected 50 additional plots in these habitats from the MNDNR Hydrography GIS layer (MNDNR 2015). We randomly selected 1 plot per wetland that was 0.81–8.09 ha, ≤402 m from a road, and adjacent to public land. After initially selecting plots from both layers, we examined aerial photos to assess the accessibility of these locations. We attempted to sample all plots that initially appeared potentially accessible, but not those that appeared inaccessible.

We navigated to the approximate location of each plot center using a Garmin Montana GPS unit, and established a plot center. If the plot center was difficult to access (e.g., because of soft bottom substrate that could not be traversed on foot, dense vegetation that could not be penetrated via boat) or on or near an ecotone, we moved the plot location to a site that was as close as possible to the initial location, accessible, and in the interior of a somewhat homogeneous vegetation patch. Moving plots away from ecotones reduced the likelihood of misclassifying habitats (i.e., habitat misclassifications are more likely to occur near ecotones because the exact location of a sampled plot is difficult to determine with somewhat imprecise GPS units). We also moved some plots located in open water habitats to the nearest vegetated location within the wetland because the former habitat type is simple and easily identified with LiDAR data. Instead, we chose to dedicate the greatest sampling effort to vegetated plots.

For each plot, we recorded the date, start time, observers, plot number, whether wood ducks were observed within 100 m of plot, and if so, provided a count of individuals in each cohort (male, female, brood, unknown). We ascertained whether the NWI classification (system, subsystem, class) available on our GIS layer was correct at each plot (i.e., some wetlands may have changed since the original classification, or that the original classification may have been incorrect), and recorded the appropriate NWI wetland classification to the level of subclass. We classified the types of wood duck loafing structures present within the plot (7 classes: none, rock, log or stump, muskrat lodge, beaver lodge or dam, small island or tussock, barely or lightly vegetated shoreline), as well as the type of beaver modification, if any that had some influence on the plot (6 classes: none, water level, runs, tree removal, dam or lodge, food cache). We also obtained a location data for each plot center using a Geneq Sx Blue II GPS unit (20–50 cm accuracy in open habitats when data were obtained at 1 reading / 5 seconds for 1 min), and recorded the specific GPS unit used.

At each plot, we placed a 2- X 2-m Daubenmire square (Daubenmire 1959, Gilmore et al. 2008) so its center was located at plot center, and measured several habitat variables within the device. This square had 0.2 m delineations, which facilitated the measurement of several habitat variables. Specifically, we used these delineations to estimate the % coverage (5% increments) of 4 habitat classes (emergent, floating leaf, open water, shrub [woody vegetation ≤1.37 m tall]) that were present at or above the water surface, and of submergent plants, when possible to make reliable observations (i.e., at locations in which water turbidity or sun glare did not substantially hinder observability). Within the Daubenmire square, we also documented the dominant emergent cover type (14 classes: none, alder [*Alnus spp*.], Canada bluejoint grass [*Calamagrostis canadensis*], giant bur-reed [*Sparganium eurycarpum*], cattail [*Typha spp.*], ericaceous shrub, floating-leaf, giant reed grass [*Phragmites spp*.], rush [*Scirpus spp*.], reed canary grass [*Phalaris arundinacea*], sedge [*Carex spp*.], willow [*Salix spp*.], wild rice [*Zizania aquatica*], other). Other habitat components measured inside the Daubenmire square were the minimum depth of submergent vegetation and the height of emergent vegetation and shrubs (0.1 m increments with a 3-m ruler), tree canopy height (0.1 m increments for woody vegetation >1.37 m tall with a Suunto clinometer or with a 3-m ruler), mean tree canopy cover (with a spherical densitometer), and water depth (with either a 3-m measuring pole [(0.1 m increments] at relatively shallow plots or an Eagle FishEasy 245DS depth finder [0.03 m increments] at deeper locations).

Within the Daubenmire square, we also estimated vertical vegetation cover and structure using a round Robel pole (Robel et al. 1970) that had alternating 0.1-m white and black bands and narrow, vertical, and contrasting marks at the midpoint of each band. Because it was not possible for personnel to stand at plots in relatively deep water or where the soil substrate was soft, it was necessary to adapt this device so that it could be used by 2 people in a boat. This adaptation consisted of attaching a long wooden pole to the Robel pole in a perpendicular manner. One crew member extended the Robel pole to the corner of the Daubenmire square opposite the other crew member, and oriented this device upright to the water surface. The other crew member placed their sighting eye 0.8 and 1.6 m above the water surface with the aid of the 3-m ruler, and recorded the lowest decimeter or 0.5 decimeter mark that could be observed from diagonally across the Daubenmire square (2.8 m). Crew members switched assignments and took readings from across the opposite diagonal of the square. This approach generated 2 measurements from each observation height, all of which were averaged together.

Forest Surveys

We first obtained forest spatial data (e.g., stand age and location, forest cover type) from Cass County, State of Minnesota, and USDA Forest Service databases. Because of slight differences among these databases regarding the classification of forest cover types, we aggregated forest composition information from these databases into 5 cover types that are likely to be used by nesting wood ducks. These cover types are aspen-birch, lowland hardwoods, mixed conifer, northern-hardwoods, and oak). We also were interested in surveying only stands likely old enough to have developed the structures likely to be used by nesting wood ducks (i.e., Aspen-Birch ≥50 years, all other stand types ≥80 years).

To reduce the likelihood of underestimating the variability of habitat structure and sampling somewhat unrepresentative habitats when selecting survey sites, we specified that ≤2 plots per stand could be established, and that these plot centers must be both ≥50 m apart and ≥30 m from the nearest stand boundary. Using these criteria, we then stratified forest stands on public lands by cover type and age class, and randomly selected 300 forest stands (60 stands of each of the 5 types, *n* = 563 plots) to be surveyed. It was necessary to remove 19 plots from the sample because of nearby heritage sites, or scheduled timber harvesting (i.e., interpretation of habitat characteristics would be problematic if timber harvesting occurred between the times forest surveys were conducted and LiDAR data were collected).

We navigated to the selected plot centers using a Garmin Montana GPS, and established 20-m circular plots (0.126 ha) around those points. Plots located near ecotones were moved sufficiently into the forest interior as to avoid apparent edge effects of vegetation structure. We first recorded the plot identification number, date, start and end times of survey, visit number to the plot (first or second), observers, cloud cover (0.1 increments), and proportion of tree boles covered by snow or leaf-out (0, 0.01–0.10, 0.11–0.33, 0.34–0.66, 0.67–1.00). We obtained location data for each plot center using Geneq Sx Blue II (0.9–1.8 m accuracy under closed forest canopy when obtaining 1 reading / 5 seconds for approximately 15 min) and Geneq Sx Blue II + GNSS (0.5–0.9 m accuracy under closed forest canopy when obtaining 1 reading / 5 seconds for approximately 15 min) GPS units, and recorded the GPS make, model, and unit number used at each plot. We also classified the stand structure following U.S.D.A. Forest Service (2014; 5 classes: single story, two-storied, multi-storied, mosaic, unknown/unassessable;) and forest cover type following Eyre (1980). We assigned these specific cover types to 5 more general types (Appendix 1).

We then examined and measured individual tree stems within each plot following an established protocol (USDA Forest Service 2014), with some exceptions. Specifically, we surveyed only trees large enough to have cavities used by nesting wood ducks (i.e., ≥22.0 cm DBH [Haramis 1975]), and tall enough for DBH to be measured (≥1.37 m). Starting at the 0° azimuth within each plot, we proceeded clockwise, numbering each suitable tree stem, and recording the following data for each stem: species, DBH (0.1 cm increments), distance (0.1 m increments) and direction (1° increments) from plot center, health status (following Thomas 1979, Appendix 2), and crown class (5 classes: remnant, dominant, codominant, intermediate, overtopped; U.S.D.A. Forest Service 2014).

All field crew members then used binoculars to conduct a preliminary search of each tree in the plot to identify cavities that potentially were suitable for nesting by wood ducks. When a potentially suitable cavity was encountered, we used a Pyle Model PLCM22IR remote camera attached via a stiff, braided wire to a 15.24 m Crain CMR Series Measuring Ruler (*sensu* Waldstein 2012) to perform a more careful examination of the entrance and interior of the cavity. We first determined whether cavity entrance dimensions were suitable by attempting to pass a cardboard cut-out of the minimum usable dimensions (6 x 6 cm, Zwicker 1999, cited in Denton et al. 2012*b*) through the cavity opening. This cut-out was placed on the wire connecting the camera to the measuring ruler. We then examined cavity interiors with the camera to ascertain whether the following conditions had been met: bottom of cavity entrance was ≥0.6 m above ground level (Strom 1969), vertical depth (from the bottom of the cavity to the bottom of the entrance) was ≥10.2 cm to 4.5 m; Bellrose and Holm 1994 p. 176) and not hollow to the ground (Robb 1986, cited in Bellrose and Holm 1994, p. 178), horizontal depth (from inner bark of the entrance opening toward the back of the cavity) appeared large enough for hens to move from the entrance to the interior of the cavity, nest platform dimensions were ≥14 x 15, cm (Boyer 1974, Haramis 1975, Denton et al. 2012*a*), and the cavity did not contain standing water or excess debris (Sousa and Farmer 1983).

Field personnel ascertained whether (1) cavity dimensions were adequate to permit a wood duck to enter the cavity and access the likely nesting location and (2) structural impediments were likely to hinder nesting efforts, and used this information to classify the suitability of each examined cavity for wood duck nesting (4 levels: suitable, marginal, unsuitable, unknown). We considered a cavity to be suitable if all these conditions were met, and unsuitable if any of these conditions were not met. A cavity was classified as marginal if it were unclear whether all dimensional requirements were met (i.e, ≥1 dimensional measurement appeared to be close to some minimum or maximum value). Cavities typically were classified as unknown/unobservable if personnel were unable to completely observe the cavity, either because of cavity height or some structural attribute did not permit observation with the camera system. We considered a cavity to be unsuitable if any dimensional measurement were not met or if there were standing water or excess debris in the cavity. Field personnel also provided a cause for unsuitability (7 classes: entrance dimensions too small, insufficient horizontal depth, insufficient vertical depth, insufficient platform dimensions, too deep or hollow to the ground, standing water in the cavity, excessive debris in the cavity). Our assessment of the suitability of interior characteristics required some subjectivity because direct measurements could not be made with our camera system.

For each cavity inspected, we recorded tree number, cavity entrance type (3 classes: opening on the top, side, combination of top and side openings which are joined on the exterior of the tree), primary and secondary sources of cavity formation (11 classes: split, broken limb, broken top, woodpecker, fire, lightning, insect, logging wound, decay/rot, other, unknown), evidence of animal use (9 classes: eggshell/ membrane, nesting materials, hive or other insect structure, animal present, scratching at entrance, pecking at entrance, other, unknown, none), and animal taxa. We also measured cavity height with either a 15.24 m measuring ruler (±0.1 m) or Suunto clinometer (±0.5 m).

LiDAR Data Collection

MNDNR Resource Assessment Program (RAP) originally planned to have LiDAR and associated remote sensing data collected during aerial flights conducted by a contractor during Fall 2016. These data-collection flights were postponed until Fall 2017. This postponement will preclude us from associating LiDAR and field data until late Winter 2018.

STUDY AREA

The primary study area encompasses 202,342 ha in northeastern Cass County, Minnesota (Figure 1), but may be expanded if additional funds become available. Parts of Chippewa Plains, Pine Moraines-Outwash Plains, and St. Louis Moraine Ecological Subsections (Hanson and Hargrave 1996) occur within this area.

RESULTS

Wetland Surveys

We conducted surveys at 185 wetland plots during the late summer and early fall of 2017 (Table 1, Figure 2). Of the plots sampled, 30 had no vegetation at or above the water surface but 155 had some form of vegetation growth. In the latter plots, the dominant vegetation at or above the water surface were classified as: alder (*n* = 1), blue joint grass (*n* = 4), bur reed (*n* = 2), cattail *spp* (*n* = 16), ericaceous shrub (*n* = 5), floating leaf (*n* = 16), phragmites *spp* (*n* = 9), rush *spp* (*n* = 31), reed canary grass (*n* = 11), sedge *spp* (*n* = 21), willow (*n* = 2), wild rice (*n* = 40), and other vegetation (*n* = 1). We observed that 14 (7.6%) plots were modified by beaver, 11 (5.9%) had potential wood duck loafing sites, and wood ducks were present ≤100 m of 20 (10.8%) plots.

Forest Surveys

We surveyed 26 forest plots during Fall 2016 and an additional 126 plots during Spring 2017 (Figure 3). The forest cover types (Eyre 1980) of plots surveyed were classified primarily as aspen, northern red oak and sugar maple-basswood (Table 2). A total of 4,931 trees of 26 species were measured and inspected for cavities (Table 3). Of these trees, 536 had potential cavities that we inspected with the remote camera-system (724 total cavities, as many trees had multiple cavities). The majority of cavities were classified as unsuitable for nesting by wood ducks (*n* = 429; 66%), and the remainder were classified as suitable (*n* = 126; 17%), marginally suitable (*n* = 65; 9%), or of unknown suitability (*n* = 54; 8%).

FIA Analysis

We did not conduct any analyses of FIA data during FY17, but will perform analyses during FY 18.

DISCUSSION

Wetland Surveys

Initially, we randomly selected wetlands for sampling to obtain an adequate sample size for each NWI class, with special emphasis placed on those classes that are most likely to have diverse vegetation structure. However, these efforts were confounded in-part by limitations of the existing NWI spatial data. Specifically, we observed during field data collections that NWI classifications of some randomly selected plots were incorrect, which we attribute to a combination of misclassification of wetland habitats, habitat changes since the original classification, and projection error. Such discrepancies contributed in-part to the resultant allocation of samples (Table 1), as did our effort to sample relatively intensively in the important vegetation types within the study area so that these types could be identified with LiDAR. We chose to move some randomly selected plots that were originally located in open water habitats with no vegetative structure at or above the water surface (e.g., Unconsolidated Bottom class) to the nearest vegetated wetland location that had a relatively homogeneous structure. Presumably, wetland habitats with no surface vegetation should have a rather simple and readily identifiable LiDAR signature, whereas those with different types of vegetation will be diverse in structure and therefore will require greater sample sizes to identify with LiDAR.

During the upcoming field season, we will attempt to obtain an adequate sample size of the different vegetation types present in the study area, but this will be challenging because much of the available NWI GIS layer classifies wetlands only to the level of class, which usually provides little information about vegetation structure. However, the NWI layer for north-central Minnesota is being updated, so we will explore using any new spatial information for plot selection.

Forest Surveys

The proportion of trees with suitable cavities varied by species, DBH and health status (Tables 4 and 5). Our preliminary results indicate that most suitable cavities were produced in sugar maple (*Acer saccharum*), northern red oak (*Quercus rubra*) and American basswood (*Tilia Americana*, Table 4). Further, the percentage of trees with suitable cavities generally increased with DBH (Table 3). The average DBH of all trees sampled was 33.3 cm (range: 22.0–94.3 cm), but the average DBH of trees with suitable cavities was 42.3 cm (range: 22.8–73.6 cm). For most species, trees that were dying or dead had a greater percentage of suitable cavities than live, healthy, trees (Table 5).

We will explore ways to select plots to be surveyed during Fall 2017 of forest cover types and tree species-DBH classes that are underrepresented in our current sample. Unfortunately, some tree species (e.g., American elm, *Ulmus americana*) and relatively large-DBH trees in general are uncommon, which challenge our ability to obtain desired sample sizes.

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T. Kaebish and D. Kepler (MNDNR RAP) provided substantial insight regarding the potential uses of LiDAR data, and facilitated the use of their data in our research. We thank S. Mortenson (Leech Lake D.R.M.), J. Watts (Cass County M.I.S.), S. Dunham, J. Rickers, T. Tisler (U.S.D.A. Forest Service) for logistical support. J. Giudice and V. St.-Louis provided many suggestions regarding study design and sampling regime. We thank J. Lawrence provided helpful reviews of research proposals and an earlier draft of this report.

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Table 1. The proportion of wetlands in each National Wetland Inventory (NWI) classification (to class level), and number of plots selected to be surveyed and actually surveyed in the Cass County, Minnesota, USA study area during 2016.

NWI wetland classification	Proportion of wetlands in study area	Number of plots selected	Number of plots surveyed
Lacustrine limnetic unconsolidated bottom	0.522	20	4
Lacustrine littoral aquatic bed			7
Lacustrine littoral emergent vegetation	0.004	60	23
Lacustrine littoral unconsolidated bottom	0.020	20	13
Palustrine aquatic bed			13
Palustrine emergent vegetation	0.102	60	102
Palustrine forested	0.191	20	0
Palustrine scrub-shrub	0.130	20	7
Palustrine unconsolidated bottom	0.026	20	3
Riverine lower perennial emergent			7
Riverine Lower Perennial Rock Bottom			1
Riverine lower perennial unconsolidated Bottom	0.002	20	5
Riverine upper perennial unconsolidated bottom	0.003	20	0

Table 2. The forest cover type classification (Eyre 1980) and sample size of forest plots surveyed in Cass County, Minnesota, USA during 2016–2017.

Forest cover-type	Number of plots surveyed
Aspen	39
Black ash–American elm–red maple	11
Bur oak	9
Eastern white pine	3
Northern red oak	32
Paper Birch	13
Red maple	2
Red pine	7
Sugar maple	2
Sugar maple–basswood	32
White pine–northern red oak–red maple	2

Table 3. The number of stems counted in each tree species and diameter-at-breast-height (DBH, in centimeters) class within forest plots located in Cass County, Minnesota, USA during 2016–2017. In parentheses are the proportion of those trees with suitable cavities followed by the associated standard error. Dashed lines indicate no values for tree species-DBH classes with no trees sampled or no suitable cavities detected.

	DBH class (cm)						
Tree species	22–29	30–39	40–49	50–59	60–69	70–79	≥80
American basswood (*Tilia americana*)	357 (0.008, 0.005)	189 (0.032, 0.013)	101 (0.069, 0.025)	36 (0.139, 0.058)	12 (0.083, 0.08)	1 (0,–)	1 (0,–)
American elm (*Ulmus americana*)	15 (0,–)	2 (0,–)	1 (0,–)	–	–	–	–
Balsam fir (*Abies balsamea*)	80 (0,–)	13 (0,–)	2 (0,–)	–	–	–	–
Balsam poplar (*Populus balsamifera*)	7 (0,–)	10 (0,–)	4 (0,–)	–	–	–	–
Bigtooth aspen (*Populus grandidentata*)	105 (0,–)	118 (0.017, 0.012)	59 (0.017, 0.017)	23 (0.043, 0.042)	11 (0.091, 0.087)	3 (0,–)	–
Black ash (*Fraxinus nigra*)	156 (0,–)	29 (0,–)	7 (0,–)	2 (0,–)	–	–	–
Black cherry (*Prunus serotina*)	1 (0,–)	–	–	–	–	–	–
Box elder (*Acer negundo*)	3 (0,–)	1 (0,–)	1 (0,–)	–	–	–	–
Bur oak (*Quercus macrocarpa*)	140 (0.007, 0.007)	74 (0,–)	18 (0,–)	8 (0,–)	6 (0.333, 0.192)	–	–
Eastern cottonwood (*Populus deltoides*)	1 (0,–)	–	–	–	–	–	–
Eastern hophornbeam (*Ostrya virginiana*)	1 (0,–)	–	–	–	–	–	–
Eastern larch (*Larix laricina*)	–	1 (0,–)	1 (0,–)	–	–	–	–
Eastern white pine (*Pinus strobus*)	9 (0,–)	13 (0,–)	17 (0,-)	8 (0.125, 0.117)	11 (0,–)	4 (0.250, 0.217)	4 (0, -)
Green ash (*Fraxinus pennsylvanica*)	93 (0.011, 0.011)	47 (0,–)	18 (0,–)	6 (0,–)	–	–	–
Hackberry (*Celtis occidentalis*)	3 (0,–)	1 (0,–)	–	–	–	–	–
Jack pine (*Pinus banksiana*)	10 (0,–)	6 (0,–)	1 (0,–)	–	–	–	–
Northern pin oak (*Quercus ellipsoidalis*)	6 (0,–)	7 (0,–)	–	–	–	–	–

Northern red oak (*Quercus rubra*)	247 (0.008, 0.006)	287 (0.038, 0.011)	136 (0.044, 0.018)	26 (0.115, 0.063)	12 (0.167, 0.108)	1 (0,–)	–
Northern white-cedar (*Thuja occidentalis*)	4 (0,–)	1 (0,–)	3 (0,–)	1 (0,–)	–	–	–
Paper birch (*Betula papyrifera*)	282 (0.004, 0.004)	183 (0.005, 0.005)	28 (0,–)	1 (0,–)	1 (0,–)	–	–
Quaking aspen (*Populus tremuloides*)	210 (0,–)	289 (0.010, 0.006)	170 (0.053, 0.017)	34 (0.118, 0.055)	6 (0,–)	–	–
Red maple (*Acer rubrum*)	243 (0.008, 0.006)	116 (0.034, 0.017)	17 (0.118, 0.078)	1 (0,–)	2 (1,0)	–	–
Red pine (*Pinus resinosa*)	26 (0,–)	59 (0,–)	67 (0,–)	36 (0,–)	13 (0,–)	2 (0,–)	1 (0,–)
Sugar maple (*Acer saccharum*)	255 (0.016, 0.008)	138 (0.065, 0.021)	48 (0.271, 0.064)	18 (0.222, 0.098)	4 (0.75, 0.217)	–	1 (0,–)
White spruce (*Picea glauca*)	6 (0,–)	6 (0,–)	2 (0,-)	–	–	–	–
Yellow birch (*Betula alleghaniensis*)	13 (0.077, 0.074)	9 (0.111, 0.105)	6 (0.167, 0.152)	1 (0,–)	–	–	–
Unidentified ash *spp* (*Fraxinus spp*)	5 (0,–)	–	–	–	–	–	–
Unidentified pine *spp* (*Pinus spp*)	–	3 (0.333, 0.272)	–	–	–	–	–
Unidentified aspen *spp* (*Populus spp*)	6 (0,–)	16 (0.125, 0.083)	9 (0, –)	4 (0,–)	–	–	–
Unknown *spp*	8 (0,–)	2 (0,–)	–	–	2 (1,0)	–	–

Table 4. The percentage of trees by tree species that were sampled, the percentage of trees of each species with suitable cavities, and the percentage of trees of each species with suitable or marginal cavities that were detected within forest plots located in Cass County, Minnesota, USA during 2016–2017.

Tree species	% of all trees sampled	% of all trees with suitable cavities	% of all trees with suitable or marginal cavities
American basswood (*Tilia americana*)	14.14	18.97	16.28
American elm (*Ulmus americana*)	0.37	–	–
Balsam fir (*Abies balsamea*)	1.93	–	–
Balsam poplar (*Populus balsamifera*)	0.43	–	–
Bigtooth aspen (*Populus grandidentata*)	6.47	4.31	4.07
Black ash (*Fraxinus nigra*)	3.93	–	0.58
Black cherry (*Prunus serotina*)	0.02	–	–
Box elder (*Acer negundo*)	0.10	–	–
Bur oak (*Quercus macrocarpa*)	4.99	1.72	1.74
Eastern cottonwood (*Populus deltoides*)	0.02	–	–
Eastern hophornbeam (*Ostrya virginiana*)	0.02	–	–
Eastern larch (*Larix laricina*)	0.04	–	–
Eastern white pine (*Pinus strobus*)	1.34	1.72	2.33
Green ash (*Fraxinus pennsylvanica*)	3.33	0.86	0.58
Hackberry (*Celtis occidentalis*)	0.08	–	–
Jack pine (*Pinus banksiana*)	0.34	–	–
Northern pin oak (*Quercus ellipsoidalis*)	0.26	–	–
Northern red oak (*Quercus rubra*)	14.38	18.97	15.12
Northern white-cedar (*Thuja occidentalis*)	0.18	–	–
Paper birch (*Betula papyrifera*)	10.04	1.72	3.49
Quaking aspen (*Populus tremuloides*)	14.38	12.07	14.53
Red maple (*Acer rubrum*)	7.69	6.90	9.88
Red pine (*Pinus resinosa*)	4.14	–	–
Sugar maple (*Acer saccharum*)	9.41	26.72	25.58
White spruce (*Picea glauca*)	0.28	–	–
Yellow birch (*Betula alleghaniensis*)	0.59	2.59	1.74
Unidentified ash spp (*Fraxinus spp*)	0.10	–	–
Unidentified pine spp (*Pinus spp*)	0.06	0.86	0.58
Unidentified aspen spp. (*Populus spp*)	0.71	1.72	2.91
Unknown spp	0.24	0.86	0.58

Table 5. The species-specific number of suitable cavities detected; percentage of cavities in live, dying, and dead trees; and percentage of trees examined in the live, dying and dead classes in Cass County, Minnesota, USA during 2016–2017. Health status classifications (1–7 described in Appendix 2) were assigned to broader classifications as follows: live (1), dying (2), and dead trees (3–7). Tree species were included only if at least one suitable cavity was found.

Tree species	Number of suitable cavities	Cavities in live trees (%)	Live trees (%)	Cavities in dying trees (%)	Dying trees (%)	Cavities in dead trees (%)	Dead trees (%)
American basswood (*Tilia americana*)	22	50.00	85.22	27.27	9.90	22.73	4.88
Bigtooth aspen (*Populus grandidentata*)	5	–	51.41	20.00	27.90	80.00	20.69
Bur oak (*Quercus macrocarpa*)	2	66.67	86.59	33.33	12.20	–	1.22
Eastern white pine (*Pinus strobus*)	2	–	63.64	–	10.61	100.00	25.76
Green ash (*Fraxinus pennsylvanica*)	1	-	83.54	–	11.59	100.00	4.88
Northern red oak (*Quercus rubra*)	22	29.17	67.28	41.67	22.57	29.17	10.16
Paper birch (*Betula papyrifera*)	2	-	60.00	-	18.18	100.00	21.82
Quaking aspen (*Populus tremuloides*)	14	6.25	36.81	25.00	33.57	68.75	29.62
Red maple (*Acer rubrum*)	8	10.00	50.40	70.00	39.58	20.00	10.03
Sugar maple (*Acer saccharum*)	31	6.06	57.54	84.85	35.99	9.09	6.47
Yellow birch (*Betula alleghaniensis*)	3	33.33	55.17	33.33	37.93	33.33	6.90
Unidentified pine *spp* (*Pinus spp*)	1	-		-	-	100.00	100.00
Unidentified aspen *spp* (*Populus spp*)	2	–	5.71	–	–	100.00	94.29
Unknown *spp*	1	–	33.33	–	–	100.00	66.67

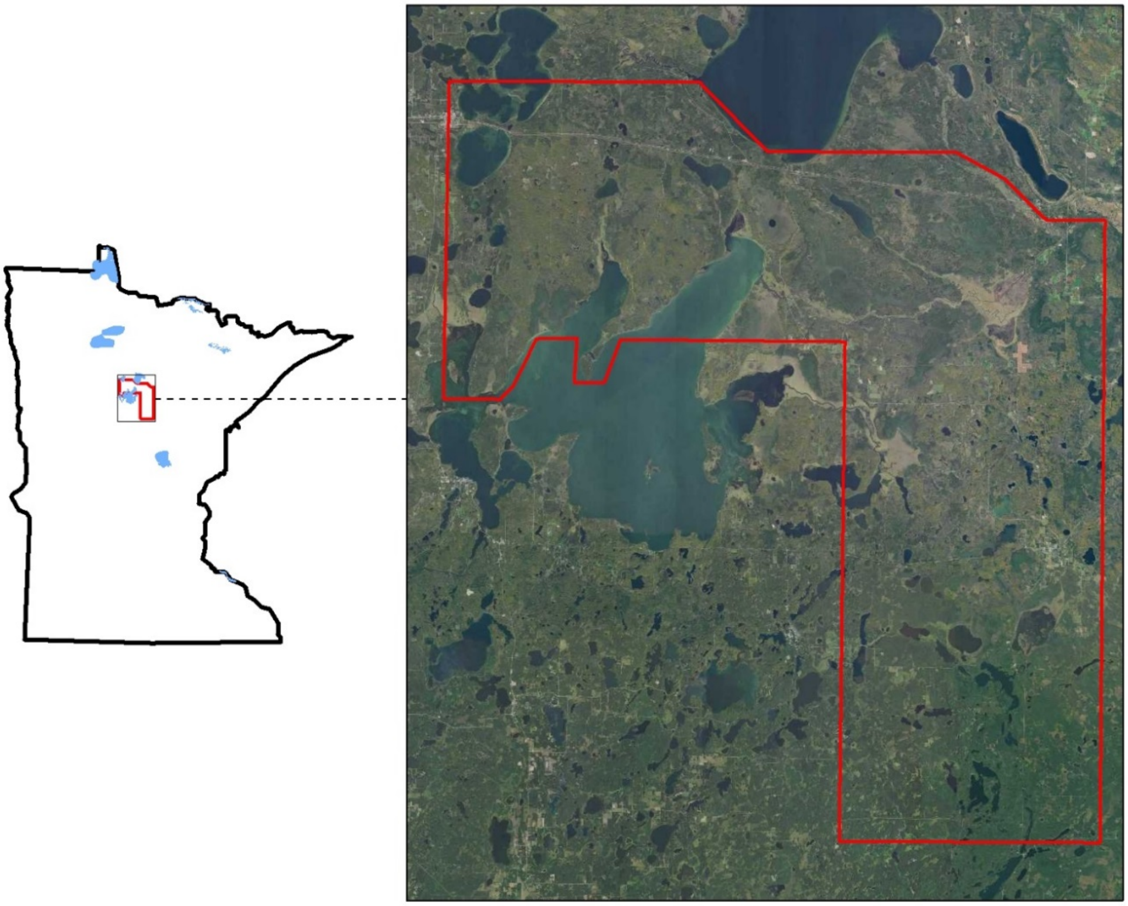

Figure 1. Location of the wood duck-LiDAR project in Cass County, Minnesota, USA.

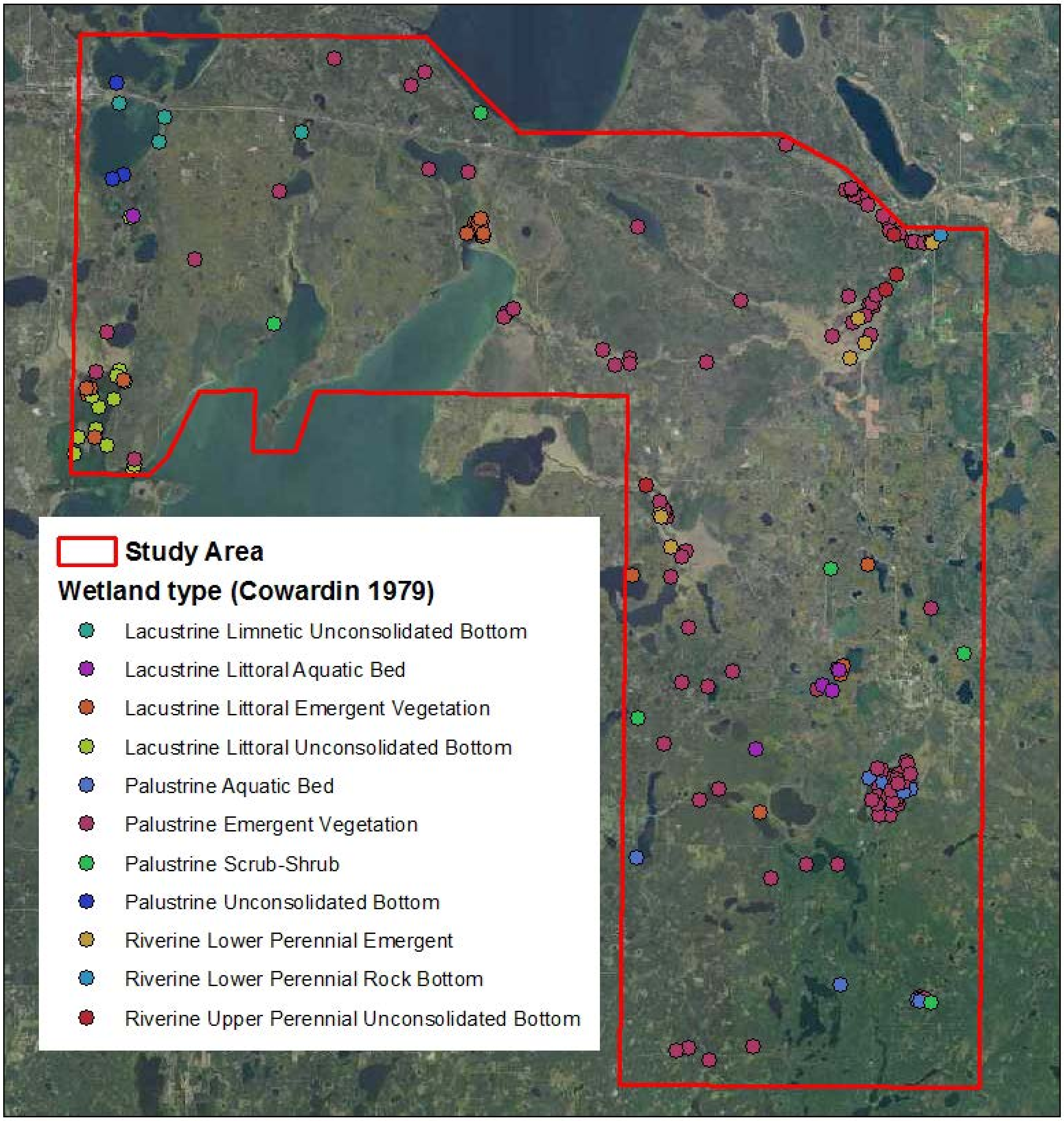

Figure 2. Location of wetland plots of different National Wetland Inventory classes (Cowardin et al. 1979) surveyed in in Cass County, Minnesota, USA during Summer and Fall 2016.

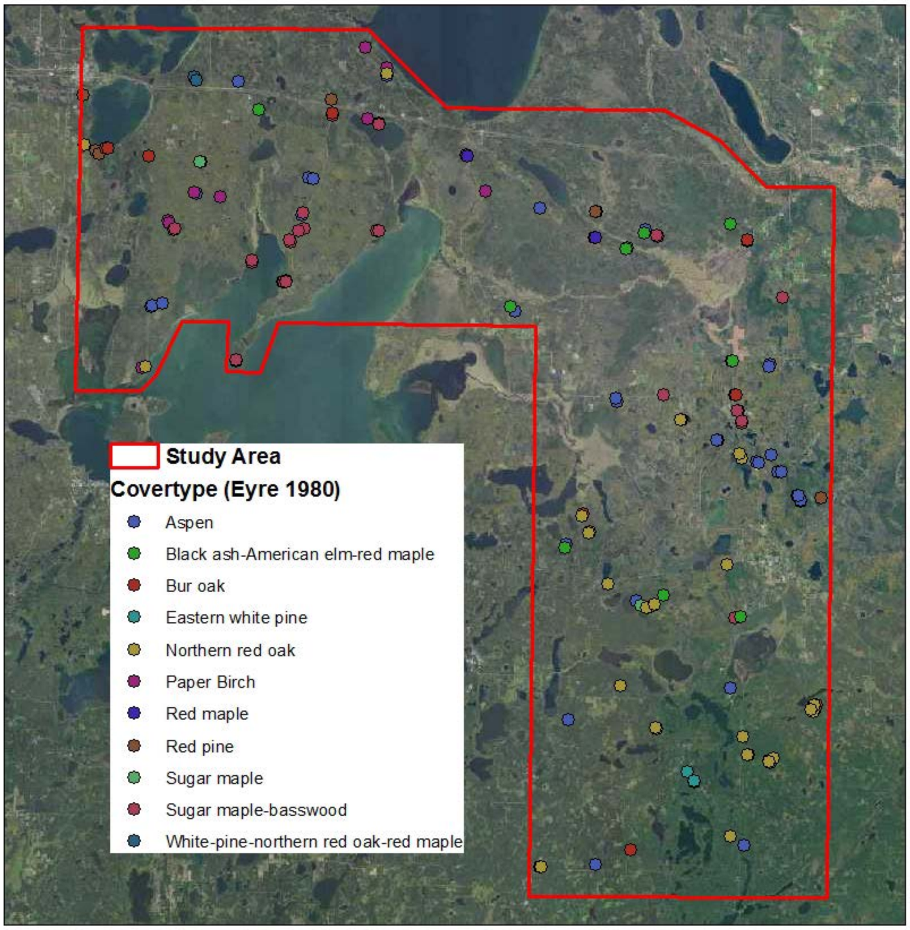

Figure 3. Location of forest plots of different cover types (Eyre 1980) that were surveyed in Cass County, Minnesota, USA during Fall 2016 and Spring 2017.

Appendix 1. Crosswalk between the Forest Cover Types of Eyre (1980) and the more general forest types used to classify stands from GIS databases.

General forest type	Eyre (1980) forest cover type
Aspen – birch	Aspen (16)
	Paper birch (18)
Mixed conifer	Red pine (15)
	White pine – northern red oak – Red maple (20)
	Eastern white pine (21)
Northern hardwood	Sugar maple – basswood (26)
	Sugar maple (27)
Oak	Norther pin oak (14)
	Bur oak (42)
	White oak (53)
	Northern red oak (55)
	Black oak (110)
Lowland hardwood	Black ash – American elm – red maple (39)
	Silver maple – American elm (62)
	Red Maple (108)

Appendix 2. Numerical codes used to classify the health status of trees (from Thomas 1979).

Health status	Description
1	Live tree that has no defects or injuries that will threaten its long-term health.
2	Live tree with defects that contribute to a decline in health. Indicators may include decay on the bole, fungi, large dead limbs, and substantial cracks.
3	Recently dead tree with bark, limbs, and twigs substantially intact.
4	Dead tree that has lost some limbs and almost all twigs.
5	Dead tree that has lost most limbs and all twigs...
6	Dead tree with a broken top and hard bole wood.
7	Dead tree with a broken top and soft bole wood.

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CONJOINED WHITE-TAILED DEER (*ODOCOILEUS VIRGINIANUS*) FAWNS[1]

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SUMMARY OF FINDINGS

In May 2016, conjoined white-tailed deer (*Odocoileus virginianus*) fawns were found deceased in southeastern Minnesota. The bodies of the fawns were joined ventrally and laterally with two separate necks and heads. This is the first case described of conjoined white-tailed deer brought to full-term gestation and delivered.

White-tailed deer are the most abundant and widely distributed species of the New World deer (Heffelfinger 2011). Most female white-tailed deer ≥2 yrs old carry twins (DeYoung 2011). Conjoined twins are rare in Cervidae, but more commonly reported in humans and domestic animals. Kompanje and Hermans (2008) found 19 cases of conjoined twins in non-domestic terrestrial mammals in the literature between 1671 and 2006. These reports included five occurrences in cervids: white-tailed deer, moose (*Alces alces*), and red deer (*Cervus elaphus*). Only 2 cases of conjoined twins were reported previously in white-tailed deer. According to criteria presented by Spencer (2003), both cases would be classified as Parapagus dicephalus- a body united ventrally with 2 separate heads. In Michigan, US, a mid-gestation, two-headed white-tailed deer fetus was found during necropsy of an adult doe (Fay 1960). A similar late-gestation fetus was reported in an adult doe in western South Dakota, US (Severson et al. 1972).

We present the first known case of conjoined white-tailed deer fawns, which were birthed. In late-May 2016, a mushroom gatherer found a 2-headed white-tailed deer fawn near Freeburg, Houston County, Minnesota, US. The specimen was found on the forest floor and was reported to be clean, dry, and freshly dead. No other deer or signs of parturition were noted in the area. The specimen was submitted to the Minnesota Department of Natural Resources (MNDNR) in excellent condition and was frozen until necropsy.

The specimen presented here demonstrated Parapagus dicephalus with ventral fusion along the length of the body (Figure 1). Whereas the previous cases reported in white-tailed deer had a shared neck and bifurcation in the proximal cervical vertebrae, these fawns had 2 separate necks and heads. External body parts appeared symmetrical and normal except for bifurcation of the neck. The fawns had a single umbilicus, which was raw and free of umbilical cord. There was one vagina and one anus with fecal pellets present. The pelage was typical of neonatal white-tailed deer with spot patterning that continued through both necks and heads. The fawns weighed 3.6 kg, which was slightly greater than neonatal birth-masses reported for fawns in northeastern Minnesota (Carstensen et al. 2009). Length from base of the tail to bifurcation of the neck was 32 cm and both neck and head lengths were 24 cm. Chest girth was 33 cm. We estimated the age of the fawns to be 0-4 days postpartum based on hoof growth (Sams et al. 1996). Given the timing, morphology, and evidence at the site of collection, we believe the fawns were carried full-term. The lungs sank when placed in water, indicating the fawns were delivered stillborn.

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Examination via 3D computed tomography and magnetic resonance imaging indicated duplication of skull, cervical vertebrae, several ribs, most thoracic vertebrae, and the first sternebrae. Caudal to the region of the 9th thoracic vertebra a single vertebral column was evident (Figure 2A). No other major skeletal anomalies were noted. The gastrointestinal tracts were separate, only the right tract was complete from esophagus to anus (Figure 2B). The left gastrointestinal tract consisted of 2 non-continuous segments, one comprised of a blind-ended esophagus and a second section of gut consisting of a dilated segment of forestomach with duodenum. This latter portion of gut also ended blindly. The liver was malformed and shared. Four segments of splenic tissue were present. Two separate hearts shared a pericardial sac (Figure 2C).

Spencer (2003) reviewed approximately 1,000 cases of conjoined human twins, including 305 parapagus, and postulated that all conjoined twins arise from 2 separate notochords (i.e., forerunner of vertebral column) on 2 originally separate embryonic discs. An alternative theory suggests that monozygotic conjoined twins occur when splitting of the embryo is incomplete at the primitive streak stage of development (Kaufman 2004). Although conjoined twins are thought to be most common and well-studied in humans, causes are speculative.

ACKNOWLEDGMENTS

We wish to acknowledge Kevin Serres for collecting and reporting the specimen, and Thomas Hemker and Donald Nelson of the MNDNR for aiding in reporting of the case. Robert Utne and Jessica Brooks of Wild Images In Motion Taxidermy mounted the specimen free of charge. The mount will be on display at the MNDNR headquarters in St. Paul, Minnesota, US. A skeletal preparation of the specimen will be housed at the University of Minnesota Veterinary Anatomy Museum in St. Paul, Minnesota, US. This study was funded by the Minnesota Department of Natural Resources.

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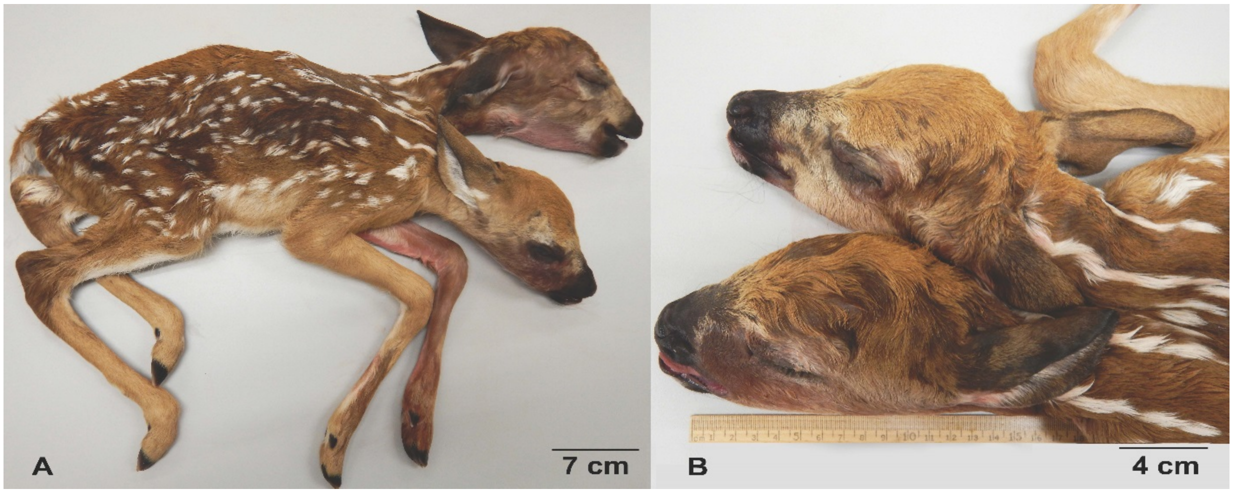

Figure 1. Stillborn, conjoined white-tailed deer fawns (*Odocoileus virginianus)* collected in Houston County, Minnesota, USA, during May 2016. A. Lateral view of entire specimen. B. Close-up of head region.

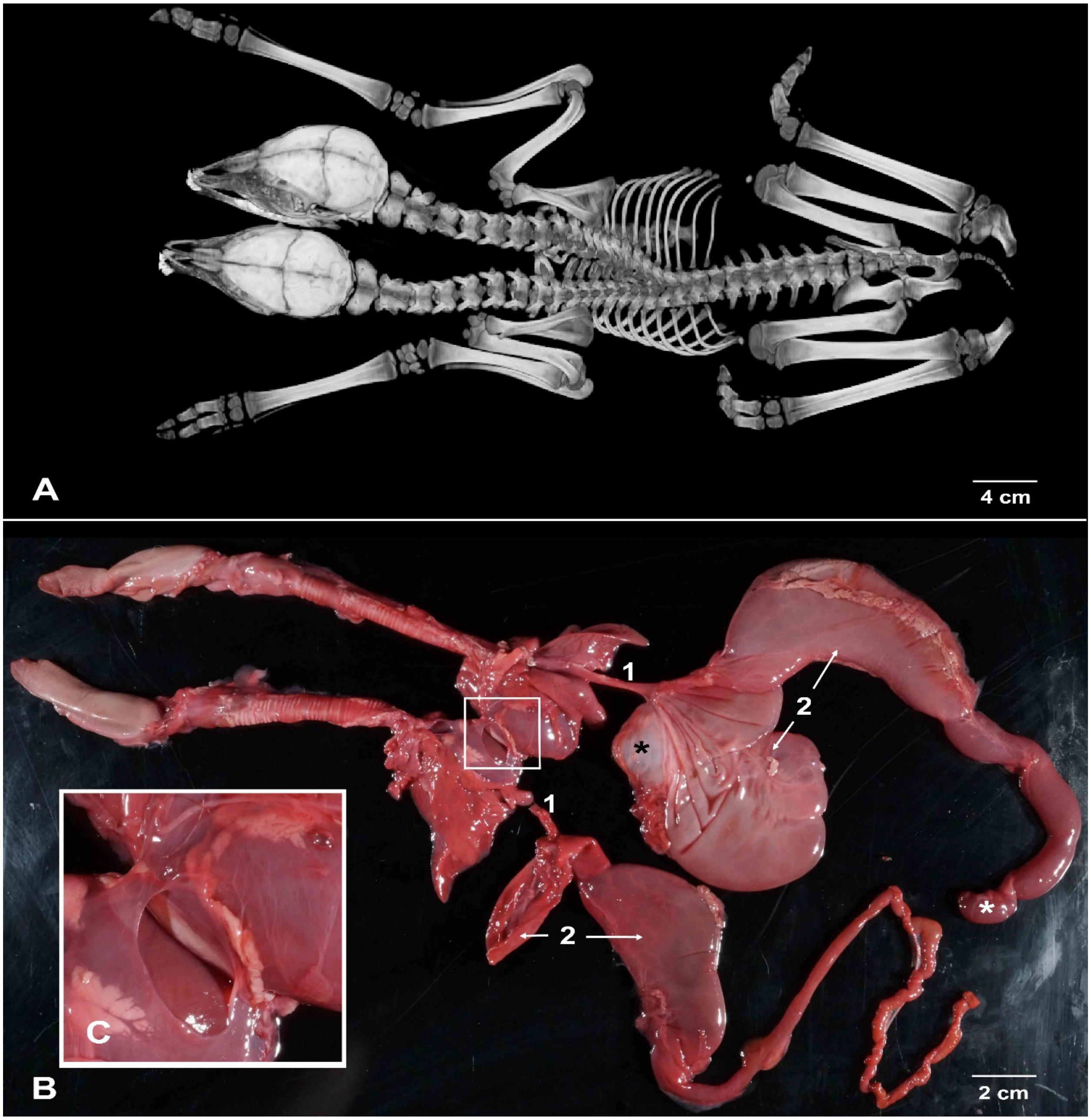

Figure 2. A. 3D computed tomography of stillborn, conjoined white-tailed deer fawns (*Odocoileus virginianus*) collected in Houston County, Minnesota, USA, during May 2016. This image documents the grossly visible separate head-neck regions and duplication of most thoracic vertebrae, which then become a single vertebral column. B. Viscera removed at necropsy from conjoined white-tailed deer fawns. Two esophagi (1) and two forestomachs (2) were present. The left head was connected with the abnormally developed upper visceral tracts-the esophagus ended in a blind sac (black asterisk) and the forestomach/upper duodenum was dilated and also ended blindly (white asterisk). The lower tract associated with the right head was complete. Note: the small separation seen in the esophagus of the lower tract and the removal of part of the lower gastrointestinal tract was done at necropsy. C. Enlarged view of boxed area showing separate hearts within a shared pericardial sac. Note: pericardial sac was incised at necropsy.

EVALUATION OF LOCALIZED DEER MANAGEMENT FOR REDUCING AGRICULTURAL DAMAGE CAUSED BY WHITE-TAILED DEER IN MINNESOTA

Gino J. 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 this study to assess the effectiveness of localized management of deer (i.e., targeted removal of deer in a limited area) to reduce damage to agricultural crops in southeast Minnesota. The objective was to evaluate the effectiveness of localized management for reducing fine-scale deer abundance and to examine whether damage caused by deer to agricultural crops is reduced on properties where deer densities are lowered. I completed 3 field seasons during 2014-2016. I used baited infrared camera surveys to estimate deer abundance on focal properties. I evaluated yields of corn in fenced and unfenced plots to estimate the impacts of browsing by deer. Corn yield loss was seemingly low on most properties, and there was no difference in corn damage between properties where localized management was used versus normal sport-hunting. Corn damage could not be explained solely by deer abundance at the property level. However, extra deer harvest opportunities were utilized when requested. Deer management was >2 times as intensive on properties where integrated management was used versus normal sport-hunting. The results of this study will provide a basis for improving the framework for future application of localized management in agricultural regions.

INTRODUCTION

Damage caused by white-tailed deer can be severe in the United States with ≥$100 million lost annually by agricultural producers (Conover 1997). Results from previous studies have demonstrated only 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.

Minimizing damage caused by deer is an important consideration in managing their populations in Minnesota. In many deer permit areas in Minnesota, deer are managed at or near population goals annually. However, complaints of deer damage from agricultural producers are common. During years 2003-2012, wildlife managers fielded an average of 130 complaints annually about damage caused by deer. 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 a distinct cluster of complaints occurs in the southeast region of the state (Nelson and Engel 2013).

In southeast Minnesota, the majority of complaints involve standing row crops and alfalfa in the field. Farmers who enter into a Cooperative Damage Management Agreement with the Minnesota Department of Natural Resources (MNDNR) are eligible for cost-sharing to install 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 southeast Minnesota involve the use of localized deer damage management techniques such as shooting permits and depredation permits (herein, localized management).

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 2012, an average of 95 shooting permits for nuisance deer were issued annually for use during summer and winter (Nelson and Engel 2013). 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 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 property to harvest up to 5 antlerless deer in addition to established bag limits during regular hunting seasons; up to 75 deer could be harvested on an individual property using depredation permits. To be eligible, applicants had to demonstrate: 1) a history of deer damage documented through complaints to the MNDNR Area Wildlife Office, 2) crop losses, 3) enrollment in a Cooperative Damage Management Agreement with MNDNR including a plan for deer hunting management, and 4) hunting was allowed on the property during the previous hunting season.

Localized management in southeast Minnesota increased deer harvest on individual properties from previous years and anecdotally landowners and hunters involved in the program were satisfied (Luedtke 2013). However, the effect of localized management on agricultural damage caused by deer is unknown. Also, logistical limitations and eligibility guidelines restrict the number of properties where depredation permits may be issued annually. Given the onerous nature of administering localized management from an agency perspective, it is important to establish whether such management aids in reducing agricultural damage as intended.

The purpose of this study is to evaluate whether localized management of deer reduces agricultural damage and to provide a basis for improving the framework for future application of localized management in Minnesota. No previous studies have examined the effectiveness of localized management for reducing damage to agricultural crops. Other research has suggested that using recreational hunting to institute localized management of overabundant deer and effectively reduce damage may be difficult (Simard et al. 2013). If localized management can be used to minimize damage, these techniques should be used wherever feasible in Minnesota. Otherwise, alternative strategies for balancing local deer populations with social carrying capacity should be explored.

OBJECTIVES

1. To evaluate the effects of localized white-tailed deer management techniques – including shooting permits and depredation permits – on localized deer densities in southeast Minnesota.
2. To quantify the amount of damage caused by white-tailed deer to corn crops relative to localized management in southeast Minnesota.

STUDY AREA

This 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, alfalfa, and small grains. Steep ravines cut by narrow streams are interspersed throughout the uplands. Ravines are rocky and primarily forested by mature hardwoods (Omernik and Gallant 1988).

Pre-fawn deer densities in the southeast Minnesota deer permit areas included in this study averaged 5 deer per km^2 (Grund 2013), which represents the highest deer densities found in the farmland zone of Minnesota. An average of 1.5 deer per km^2 was harvested in these deer permit areas during 2012, which was nearly twice the statewide average (McInenly 2013).

METHODS

Experimental Design

My objective was to evaluate the effectiveness of localized management for reducing fine-scale deer abundance and to examine whether damage caused by deer to agricultural crops is reduced on properties with higher management intensity. Therefore, I examined deer depredation to crops and deer abundance on individual focal properties in southeast Minnesota. On properties used as treatments, localized management strategies were integrated with regular sport-hunting. On control properties, normal sport-hunting was allowed by the landowner. I included 7 focal properties in the study, including 4 treatments and 3 controls.

Data Collection

Deer Abundance Estimates on Focal Properties

To aid in estimating deer abundance and management intensity (i.e., deer harvested per deer available for harvest) on focal properties, I used baited infrared camera surveys to obtain estimates of the abundance of deer at a fine scale in the area of crop fields designated for evaluation. This method of survey was conducted according to previous research by Jacobson *et al* (1997) and a pilot study I conducted in southeast Minnesota during 2013 (G. D'Angelo, unpublished data). The abundance of deer in an area can be determined using baited surveys, where bucks can be uniquely identified by antler characteristics and their number used to infer the number of does and fawns visiting a bait site repeatedly. 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. Deer abundance estimates were obtained 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 those that caused damage during the growing season and were 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. I present estimates of deer abundance as deer per camera to standardize across the range of property sizes in the study.

Management Intensity

I asked agricultural producers to report deer harvested on their properties by season. I quantified management intensity as: number of deer harvested divided by the total number of deer estimated to be on the property via infrared camera surveys. Herein, I describe properties under the 2 aforementioned management strategies: hunting (herein HUNT, i.e., hunting conducted by sport-hunters during the regular season framework), or integrated management (herein INT, i.e., hunting was integrated with localized management strategies including depredation and shooting permits outside of the regular season framework).

Corn Evaluations

Within each field, I delineated 8 plots, which were stratified into interior (>10 m from the field edge) and edge (0-5 m from the field edge). Each plot included 2 paired 5-m X 5-m subplots (~6/1000th acre) separated by 5 m and within the same rows of corn. One subplot of each pair was fenced to exclude deer and the other subplot was an unfenced control. Within each pair, the treatment and control were assigned randomly. Square exclosures were constructed with 2-m high heavy-duty plastic mesh attached to 4 2.4-m u-posts. Exclosures surrounding subplots were approximately 6 m X 6 m to reduce the effect of fencing on plants within the subplot. Exclosures were installed immediately following planting and herbicide treatment or initial cultivation for control of weeds. When necessary, exclosures were removed for $\leq$24 hours to allow farmers to conduct additional field treatments. I evaluated corn crops near the estimated date of plant maturity before senescence (approximately 130 days after planting). Within each subplot I recorded the number of rows, number of plants, and for 30 randomly selected plants, I measured plant height, level of herbivory per plant, and classified the quality of each ear of corn relative to damage caused by deer. I estimated grain yield (total seeds produced per 30 plants) for fenced and unfenced subplots, and calculated the percent corn loss for each fenced and unfenced plot as: ((total seeds in fenced plot minus total seeds in unfenced plot) divided by total seeds in the fenced plot) multiplied by 100. I consulted with the agricultural producer to determine the variety of corn planted in each field.

RESULTS AND DISCUSSION

I conducted this study beginning in spring 2014 through 2016, including 3 growing seasons for corn and 3 deer hunting seasons. HUNT was used to manage deer on 3 properties and INT was used on 4 properties. In each year, I sampled 112 subplots in corn fields including 56 unfenced subplots and 56 fenced subplots. In 2014, I excluded from analysis 2 pairs of fenced and unfenced subplots (i.e., 4 subplots total) on 1 property because the growth of corn plants was severely affected by soil erosion. In 2015, I excluded from analysis 2 pairs of fenced and unfenced subplots (i.e., 4 subplots total) on 1 property because of damage caused by raccoons (*Procyon lotor*). In 2016, I excluded from analysis 1 pair of fenced and unfenced subplots on 2 properties (i.e., 2 subplots on each property) because of damage caused by raccoons.

Deer abundance via infrared camera surveys was similar among HUNT and INT properties across years (Table 1, $t = 1.105$, $df = 19$, $P = 0.283$). Among HUNT properties, deer abundance was similar among years (2014: $\bar{x} = 20$ deer per camera, SE = 4; 2015: $\bar{x} = 25$ deer per camera, SE = 2; 2016: $\bar{x}$ = 26 deer per camera, SE = 5; $F_{2,6}$ = 0.668, P = 0.547). Also, among INT properties deer abundance was similar among years (2014: $\bar{x}$ = 20 deer per camera, SE = 3; 2015: $\bar{x}$ = 21 deer per camera, SE = 5; 2016: $\bar{x}$ = 21 deer per camera, SE = 3; $F_{2,9}$ = 0.026, P = 0.974).

Agricultural producers on INT properties used extra deer harvest opportunities in all years. Management intensity on INT properties was >2 times the management intensity on HUNT properties (Table 1, HUNT: $\bar{x}$ = 0.15, SE = 0.02; INT: $\bar{x}$ = 0.37, SE = 0.05; t = -3.838, df = 19, P = 0.001). Among HUNT properties, management intensity was similar among years (2014: $\bar{x}$ = 0.19, SE = 0.02; 2015: $\bar{x}$ = 0.13, SE = 0.05; 2016: $\bar{x}$ = 0.13, SE = 0.01; $F_{2,6}$ = 1.416, P = 0.313). Among INT properties, management intensity was similar among years (2014: $\bar{x}$ = 0.44, SE = 0.10; 2015: $\bar{x}$ = 0.42, SE = 0.06; 2016: $\bar{x}$ = 0.24, SE = 0.04; $F_{2,9}$ = 3.323, P = 0.154).

Despite increased harvest pressure for deer on INT properties versus HUNT properties during all years, corn yield loss did not differ between management strategies (Table 2; HUNT: $\bar{x}$ = 12.1, SE = 2.8; INT: $\bar{x}$ = 8.1, SE = 2.9; $F_{1,160}$ = 0.685, P = 0.409), among properties ($F_{6,155}$ = 0.519, P = 0.793), or among years (All years: $\bar{x}$ = 10.2%, SE = 2.0; 2014: $\bar{x}$ = 6.5%, SE = 2.6; 2015: $\bar{x}$ = 15.9%, SE = 3.9; 2016: $\bar{x}$ = 8.2%, SE = 3.8; $F_{2,159}$ = 2.00, P = 0.138). Corn yield loss was >3.5 times greater for edge plots versus interior plots ($F_{1,160}$ = 8.57, P = 0.004) when pooled by property across all 3 years. Notably, during 2015 when corn yield loss was greatest overall, corn yield loss was nearly 5 times greater on edge plots versus interior plots for all properties ($F_{1,52}$ = 8.60, P = 0.005).

The primary objective of this study was to evaluate the effectiveness of localized management for reducing fine-scale deer abundance and to examine whether damage caused by deer to agricultural crops was reduced on properties where deer densities were lowered. Deer abundance was similar among all properties in this study during all years, despite management intensity on INT properties being 2-3 times greater than on HUNT properties. Generally, deer densities in southeast Minnesota were high relative to other regions of the state (D'Angelo and Giudice 2016). Although a higher proportion of deer estimated to be using INT properties were harvested annually, deer on adjacent properties likely filled any voids created by localized management. Property sizes in the region were generally smaller than deer home ranges (Stewart et al. 2009), which complicates reducing deer densities sufficiently on focal properties. Agricultural fields were highly interspersed and bordered with forested cover for deer, so even at lowered deer densities, damage can occur since deer can access fields frequently. Temporary reductions in deer abundance on INT properties may have reduced annual corn losses since deer harvest on these properties occurred throughout the corn growing season. The level of corn damage that may have occurred had localized management not been used is not known.

Overall corn yield loss was seemingly low on most properties. There was no difference in corn damage between properties where localized management was used versus normal sport-hunting, and the level of corn damage could not be explained by deer abundance at the property level. Plots along the edge of corn fields experienced greater losses of corn. Our results demonstrate that this trend occurred on most properties in each year of our study. Deer typically cause greater damage on field edges, especially those nearer escape cover (DeVault et al. 2007, Stewart et al. 2007, Hinton et al. 2017), likely because risk (e.g., predation, hunting) is less.

Extra deer harvest opportunities were used by landowners when requested. Management was more intensive on INT properties versus HUNT properties. Also, deer were harvested earlier and more continuously throughout the growing season, corn drydown period, and crop harvest seasons on INT properties. Increased deer harvest pressure on INT properties may have prevented corn damage from being worse had additional deer not been harvested. Therefore, extra opportunities to harvest deer should be afforded on properties where landowners consult with MNDNR staff about their concerns for potential deer damage. These concerns are legitimate as my data demonstrated. Landowners are basing their concerns on prior experiences and current conditions. The results of this study will provide a basis for improving the framework for future application of localized management in agricultural regions. Wildlife managers with local knowledge may be best suited to make recommendations about deer management strategies. Within the regulatory framework, they should be afforded the ability to adapt deer harvest permit levels for specific situations to most effectively minimize crop losses and to foster positive relationships with agricultural producers and hunters.

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Table 1. Estimates of the abundance of white-tailed deer, management intensity of deer, and corn damage caused by deer on 7 privately owned properties in southeast Minnesota, during 2014-2016.

Property	Deer management strategy[1]	Estimated deer abundance (deer per camera)[2]			Management intensity[3]			% Corn loss[4]		
		2014	2015	2016	2014	2015	2016	2014	2015	2016
A	HUNT	26	27	30	0.16	0.08	0.13	7	37	8
B	HUNT	22	26	33	0.21	0.23	0.12	-1	29	8
C	HUNT	13	22	16	0.21	0.07	0.14	14	0	6
D	INT	26	35	19	0.35	0.28	0.32	24	0	6
E	INT	21	17	14	0.39	0.50	0.29	-6	23	15
F	INT	22	18	23	0.28	0.54	0.19	0	4	16
G	INT	11	12	29	0.74	0.36	0.16	12	16	-2

[1] On properties with HUNT management deer harvest was conducted by sport-hunters during the regular season framework. On properties with INT management deer harvest was through integrated methods including by sport-hunters during the regular season framework and using depredation and shooting permits outside of the regular season framework.

[2] Deer abundance estimated from infrared camera surveys indexed as deer per camera with camera densities of 1 camera per 65 ha on each focal property.

[3] Proportion of the number of deer estimated to be using a property that were harvested.

[4] Negative values indicate higher average yield estimates in unfenced subplots versus subplots fenced to exclude deer.

Table 2. Estimates of corn damage caused by white-tailed deer on edge and interior sampling plots on 7 privately owned properties in southeast Minnesota, during 2014-2016. Edge plots were along the field edge and interior plots were >10 m from the field edge.

		% Corn loss								
		All			Edge			Interior		
Year	Deer management strategy[1]	n	$\bar{x}$	SE	n	$\bar{x}$	SE	n	$\bar{x}$	SE
2014	HUNT	24	6.6	3.1	12	3.8	4.4	12	9.5	4.3
	INT	30	6.5	4.1	15	11.1	7.1	15	1.8	3.9
2015	HUNT	24	22.2	6.7	12	35.9	12.0	12	8.6	3.3
	INT	30	10.7	4.5	14	19.4	8.5	16	3.2	3.0
2016	HUNT	24	7.4	3.1	12	9.2	5.6	12	5.6	2.7
	INT	30	8.7	6.5	14	18.1	13.3	16	8.0	3.2

[1] On properties with HUNT management deer harvest was conducted by sport-hunters during the regular season framework. On properties with INT management deer harvest was through integrated methods including by sport-hunters during the regular season framework and using depredation and shooting permits outside of the regular season framework.

EVALUATING INSECTICIDE EXPOSURE RISK FOR GRASSLAND WILDLIFE ON PUBLIC LANDS

Nicole M. Davros and Katelin Goebel[1]

SUMMARY OF FINDINGS

Increasing evidence suggests that acute toxicity to pesticides may be more important than agricultural intensity in explaining declines in grassland-dependent wildlife. Although neonicotinoids (systemic insecticides routinely used on corn and soybeans) are currently under scrutiny for their effects on birds and pollinators, other insecticides are commonly used in Minnesota's farmland regions that may also have negative effects on non-target organisms. Minnesota Department of Natural Resource (MNDNR) wildlife managers and members of the public have reported concerns about foliar-application insecticides in particular. Such insecticides are used on a variety of crops but their use has been especially important for controlling soybean aphid outbreaks in Minnesota's farmland regions. Concerns have previously been raised about the impacts of chlorpyrifos, a broad-spectrum organophosphate, and other foliar-application insecticides on water quality and human health, prompting the Minnesota Department of Agriculture (MDA) to release guidelines for voluntary best management practices for their use. Although lab studies have shown chlorpyrifos and other insecticides used to target aphids are highly toxic to non-target organisms, including economically important game species and pollinators, fewer studies have investigated the environmentally-relevant exposure risk of free-ranging wildlife to these chemicals. Our research project will assess the direct and indirect exposure risk of grassland wildlife to common soybean aphid insecticides along a gradient from soybean field edge to grassland interior. The data we obtain on the environmentally-relevant exposure risk of wildlife to these insecticides will be used to help natural resource managers and private landowners better design habitats set aside for grassland wildlife in Minnesota's farmland region.

INTRODUCTION

Grassland habitat loss and fragmentation is a major concern for grassland-dependent wildlife throughout the Midwestern United States (U.S.). In particular, habitat loss due to agricultural intensification has been implicated as a primary reason for the declines of many grassland nesting birds (Sampson and Knopf 1994, Vickery et al. 1999). However, concerns are increasingly being raised about the impacts of pesticides on birds and other wildlife in agriculturally-dominated landscapes (e.g., Hopwood et al. 2013, Hallmann et al. 2014, Main et al. 2014, Gibbons et al. 2015), and some evidence exists that acute toxicity to pesticides may be more important than agricultural intensity in explaining grassland bird declines in the U.S. (Mineau and Whiteside 2013).

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Soybean aphids were first discovered in southeastern Minnesota during 2000 and subsequently spread throughout the farmland zone by 2001 (Venette and Ragsdale 2004). Although these aphids pose significant risks to agriculture, their presence does not automatically translate to reduced yield or income (Vennette and Ragsdale 2004). In response to concerns over yield loss, the University of Minnesota Extension Office (hereafter, UM Extension) released guidelines on how to scout for aphids and when to consider treatment for infested fields (UM Extension 2014). Foliar applications of insecticides using boom sprayers or planes are common treatment methods when chemical control of aphids is considered necessary. The 2 most common insecticides used are chlorpyrifos and lambda-cyhalothrin (MDA 2005, MDA 2007, MDA 2009, MDA 2012, MDA 2014a) but bifenthrin use has also been reported (R. Riley, personal communication; E. Runquist, unpublished data). Withholding times vary by chemical (lambda-cyhalothrin: 45 d; chlorpyrifos: 28 d; bifenthrin: up to 14 d); thus, the timing of product use within the growing season should be considered. If retreatment is necessary due to a continued infestation, landowners are encouraged to use an insecticide with a different mode of action to prevent the development of resistance (UM Extension 2014). Therefore, multiple chemicals may be used on the same field at different times of the year in some situations. Alternatively, landowners may choose to use a product that combines 2 or more chemicals together (e.g., chlorpyrifos + lambda-cyhalothrin), and such products are readily available on the market.

Lambda-cyhalothrin (common trade names include Charge, Demand, Excaliber, Grenade, Hallmark, Icon, Karate, Kung-fu, Matador, Samurai, and Warrior) is a broad-spectrum pyrethroid insecticide that affects the nervous systems of target- and non-target organisms through direct contact, ingestion, and inhalation [National Pesticide Information Center (NPIC) 2001]. Although lambda-cyhalothrin is considered low in toxicity to birds, it is highly toxic to pollinators such as bees (NPIC 2001). Further, field studies have shown lower insect diversity and abundance in fields exposed to lambda-cyhalothrin (Galvan et al. 2005, Langhof et al. 2005, Devotto et al. 2006). Because insects are an especially important source of protein for birds during the breeding season, fewer insects could mean reduced food availability for fast-growing chicks.

Bifenthrin (common trade names include Bifenture, Brigade, Discipline, Empower, Tundra, and Xpedient) is a broad-spectrum pyrethroid insecticide that affects the central and peripheral nervous systems of organisms by contact or ingestion (Johnson et al. 2010). Bifenthrin is low in toxicity to birds, including game species such as bobwhite quail (*Colinus virginianus*) and mallards (*Anas platyrhynchos*)(LD_{50} values of 1800 mg/kg and <2150 mg/kg, respectively; Johnson et al. 2010). However, there are exposure risks for birds that feed on fish and aquatic insects because bifenthrin is very highly toxic to aquatic organisms (Siegfried 1993, Johnson et al. 2010). Some terrestrial insects are also susceptible to bifenthrin (Siegfried 1993). Bifenthrin is very highly toxic to bumblebees, with one study showing 100% mortality by contact (Besard et al. 2010).

Chlorpyrifos (common trade names include Dursban, Govern, Lorsban, Pilot, Warhawk, and Yuma) is a broad-spectrum organophosphate insecticide that also disrupts the normal nervous system functioning of target- and non-target organisms through direct contact, ingestion, and inhalation (Christensen et al. 2009). Although first registered for use in the U.S. in 1965, its use as an ingredient in residential, pet, and indoor insecticides was removed in 1997 (except for containerized baits) due to human health concerns (Christensen et al. 2009, Alvarez et al. 2013 and references therein, MDA 2014b). Further, MDA recently released guidelines for best management practices for the use of chlorpyrifos due to water quality concerns (MDA 2014b). Lab studies have shown chlorpyrifos to be toxic to a variety of aquatic and terrestrial organisms (reviewed in Barron and Woodburn 1995), and some bird and beneficial insect species are especially susceptible to acute toxicity from chlorpyrifos exposure (Christensen et al. 2009, MDA 2014a). Chlorpyrifos is very highly toxic to gallinaceous bird species such as the ring-necked pheasant (*Phasianus colchicus*) and domesticated chickens (*Gallus gallus domesticus*), with a lethal dose causing death in 50% of treated animals (LD_{50}) of 8.41 mg/kg and 32-102 mg/kg, respectively (Tucker and Haegele 1971, Christensen et al. 2009). Several other bird species are also particularly susceptible to chlorpyrifos, including American robins (*Turdus migratorius*), common grackles (*Quiscalus quiscula*), and mallards (Tucker and Haegele 1971, Christensen et al. 2009). Yet few field studies have been able to document direct mortality of birds from chlorpyrifos exposure (e.g., Buck et al. 1996, Martin et al. 1996, Booth et al. 2005), and an ecotoxological risk assessment conducted by Solomon et al (2001) concluded that the available evidence did not support the presumption that chlorpyrifos use in agroecosystems will result in extensive mortality of wildlife. However, chlorpyrifos exposure leading to morbidity (e.g., altered brain cholinesterase activity, altered behaviors, reduced weight gain) has been documented in both lab and field studies (McEwen et al. 1986, Richards et al. 2000, Al-Badrany and Mohammad 2007, Moye 2008). Thus, sub-lethal effects leading to indirect mortality (e.g., via increased predation rates) may be a concern for wildlife exposed to chlorpyrifos.

Minnesota DNR wildlife managers and members of the public have reported concerns about the effects of these soybean aphid insecticides on non-target wildlife, including economically important bird and pollinator species. The common public perception is that indiscriminate spraying without first scouting for aphid outbreaks has become the norm and fewer birds and insects are observed after spraying has occurred. Yet little is known about the actual exposure risk of birds and terrestrial invertebrates to these insecticides in Minnesota's grasslands. Distances reported for drift from application of foliar insecticides vary widely in the literature (5-75 m; Davis and Williams 1990, Holland et al. 1997, Vischetti et al. 2008, Harris and Thompson 2012), and a recent butterfly study in Minnesota found insecticide drift on plants located up to 1600 m away from potential sources (E. Runquist, personal communication). The distance of travel for spray drift is dependent on several factors including droplet size, boom height or width, and weather conditions (e.g., humidity, wind speed, dew point) at the time of application. Guidelines for pesticide application are readily available to landowners and licensed applicators (MDA 2014b, MDA 2014c) so that the likelihood of spray drift can be minimized but there is likely large variation in typical application practices.

OBJECTIVES

Our goal is to assess the environmentally-relevant exposure risk of grassland wildlife to commonly-used soybean aphid insecticides, especially chlorpyrifos, in Minnesota's farmland region. In particular, we will:

1) Quantify the concentration of insecticides along a gradient from soybean field edge to grassland interior to assess the potential for grassland wildlife (particularly nesting birds and their young, and beneficial insects) to be exposed to chemicals directly via contact with spray drift and indirectly through insect prey items exposed to the insecticides.
2) Quantify and compare the relative abundance, richness, diversity, and biomass of invertebrate prey items along a gradient from soybean field edge to grassland interior prior to and post-application to assess the indirect impact of the insecticides on food availability for grassland nesting birds and other wildlife.

STUDY AREA

Our study is being conducted within the south-central and southwest regions of Minnesota's farmland zone (Figure 1). These regions are intensively farmed, and corn and soybeans combined account for approximately 75% of the landscape [U.S. Department of Agriculture

(USDA) 2013a, USDA 2013b]. Acres set aside as grassland habitat on public and private land account for 5.8% and 4.6% of the landscape, respectively, in these regions (Davros 2015). Since 2003, these regions have also experienced some of the highest estimated use of chlorpyrifos and lambda-cyhalothrin (MDA 2005, MDA 2007, MDA 2009, MDA 2012, MDA 2014a).

METHODS

Experimental Design

A treatment study site will consist of a MNDNR Wildlife Management Area (WMA) immediately adjacent to and downwind from a soybean field that will be sprayed to control for aphids. We are working in close consultation with wildlife managers and private landowner cooperators to choose 6-8 treatment sites. We will choose sites dominated by a diverse mesic prairie mix containing warm-season grasses and forbs because this mix is commonly used by MNDNR managers and agency partners in the farmland zone to restore habitats for the benefit of grassland birds and beneficial insect species. We will also chose 2-4 control study sites with similar site characteristics except that control sites will not be sprayed with any chemical to control aphids.

Field sampling will occur during summer 2017 and 2018, and approximately half of the study sites will be sampled each year. Within each treatment site prior to spraying, we will establish sampling stations at distances of <1 m, 5 m, 25 m, 50 m, 100 m, and 200 m along each of 3 transects. If the site is large enough, we will also establish a station at a distance of 400 m along each transect. This design will give us a total of 18-21 stations per site. We will establish transects and stations the same way within control sites. At all sites, transects will run perpendicular to the edge of the soybean field and will be spaced 100 m apart to reduce the likelihood of duplicate insecticide exposure from the spraying event.

Data Collection

To assess the potential for direct exposure of birds and other wildlife to soybean aphid insecticides (hereafter, target chemicals), we will deploy passive sampling devices (PSDs) to absorb any chemical drift that occurs. The PSDs will be placed in treatment fields on the morning of but prior to spraying of soybeans. They will be made of Whatman^{TM} Qualitative Filter Paper (grade 2) that is attached to 0.5 in^2 hardwire cloth formed to a cylinder shape to approximate the size and shape of a large songbird or a gamebird chick. We will place the PSDs at two heights (ground and mid-canopy) at each of the sampling stations. Ground-level sampling will help represent ground-nesting birds and other wildlife that spend the majority of their time on the ground (e.g., gamebirds, small mammals, many species of invertebrates). Mid-canopy sampling will help represent above-ground nesting birds and many species of spiders and insects. We will retrieve the PSDs from the field ≤1 h after spraying and properly store them for later chemical analysis. All ground-level and mid-canopy samples will be analyzed independent of one another. At control sites, we will place PSDs at both ground and mid-canopy levels at each of the stations. We will leave the PSDs on site for the same amount of time as PSDs at treatment sites before we collect and store them for later analysis.

To assess the potential for birds and other insectivorous wildlife to be exposed to the target chemicals indirectly via consumption of prey items, we will sample invertebrates ≤2 h post-spraying at each of the sampling stations. We will sample ground-dwelling invertebrates using a vacuum trap and canopy dwelling invertebrates using a sweepnet. Vacuum trap and sweepnet samples will both be taken along 60 m transects to the left side of the sampling stations and parallel to the soybean field. We will combine vacuum trap and sweepnet samples taken from the same station during the same time period into one sample and properly store them for later chemical analysis. We will sample control sites using the same methods and timing, with the timing based on when we deploy the PSDs at these sites.

To quantify and compare the abundance, richness, diversity, and biomass of invertebrate prey items, we will collect vacuum trap and sweepnet samples from the <1-5 m, 25 m, and 100 m distances along the 3 transects at each site (total = 9 stations/site). The <1 m and 5 m distances will be combined into 1 transect parallel to the soybean field for this effort. We will collect these samples 1-3 d prior to spraying and between 3-5 d and 19-21 d post-spraying at treatment sites. Samples will be taken along 40 m transects but on the right side of the sampling stations and parallel to the soybean field. We will combine vacuum trap and sweepnet samples into 1 sample per station per sampling period and store them in ethanol for later sorting and identification. We will place emphasis on 3 invertebrate orders important in the diets of grassland nesting birds: Araneae (spiders), Orthoptera (grasshoppers, crickets, and katydids) and Coleoptera (beetles). All individuals from these orders will be sorted and identified to at least the family level for analysis. Quantifying the spider community will allow us to examine potential impacts on an additional trophic level since spiders are an important predator of insects.

We will use portable weather meters (Kestrel 5500AG Agricultural Weather Meters) to measure relevant weather data (e.g., temperature, wind speed, wind direction, humidity, dew point) along the center transect at the <1 m, 100 m, and 200 m stations during the deployment of PSDs and during insect sampling periods at each site.

At each site, we will also collect vegetation data 1-3 days prior to spraying at all sampling stations and again at 3-5 d and 19-21 d post-spraying at the reduced subset of sampling stations coinciding with invertebrate sampling efforts. Data collected will include percent canopy cover, maximum height of live and dead vegetation, litter depth, and vertical density. We are still developing our methods for vegetation data collection but we will likely use the program SamplePoint (Booth et al. 2006) to estimate percent canopy cover as it provides a more objective measure than visual estimation techniques.

We will send the PSD samples and invertebrate samples to an external lab to be analyzed using a solvent-based extraction method. Extracts will be concentrated by evaporation and then analyzed using a gas chromatography/mass spectrometry-negative chemical ionization (GC/MS-NCI) method. Although our experimental design will focus on soybean fields sprayed with foliar insecticides to control aphids, the chemical analyses will allow us to quantify additional pesticides (e.g., neonicotinoids, fungicides) at minimal extra cost. Obtaining information about other pesticide exposure will be valuable supplementary information in support of other Section of Wildlife research and management goals.

Data Analyses

We will use mixed regression models to examine factors related to risk of direct and indirect exposure of wildlife to target chemicals. Chemical concentration will be the dependent variable. We will specify distance from soybean field edge and canopy height (when relevant) as a fixed effect. We may also include other covariates such as site, ordinal date, vegetation data, and weather condition variables where appropriate. We will use similar models to examine differences in the abundance, richness, diversity, and biomass of Aranaeans, Orthopterans and Coleopterans. We will use the sampling period (i.e., 1-3 d prior to spraying, and 7-9 d or 18-20 d post-spraying) as a repeated measure in these analyses, specifying a covariance structure [e.g., autoregressive 1 (AR1)] when appropriate.

RESULTS AND DISCUSSION

To date, we have surveyed 12 farmer cooperatives in 12 counties to gather more specific information about chemical spraying (e.g., type of insecticide, application method) in southern

Minnesota. Congruent with MDA's pesticide usage reports (MDA 2007, MDA 2009, MDA 2012, MDA 2014), the coops reported that chlorpyrifos, lambda-cyhalothrin, and bifenthrin have been the most commonly-used foliar soybean insecticides in recent years. Additionally, we learned that neonicotinoids have also been used in the chemical mixes used as foliar treatment of crop pests. This information is contrary to the widespread belief that neonicotinoids are only used as a prophylactic seed treatment to treat plants systemically.

We also surveyed landowners adjacent to potential WMA study sites to learn more about their soybean aphid spraying practices and to ask for their cooperation with our study (see Appendix 1) since cooperation will be key to timing our field sampling. We mailed 221 letters during the first week of March 2017; 24 letters were returned as undeliverable. The overall response rate for the first mailing was 24.4%. In early April, we sent a second round of 164 letters and had a response rate of 6.1%. Some landowners opted to call us instead and provide their renter's contact information; however, not all landowners provided renter information when they returned the survey by mail. Overall, we were able to identify 11 landowners adjacent to and upwind from a WMA during 2017 who are willing to be cooperators with our study. We are currently contacting these landowners again to determine if they have planted soybeans this year and whether they will be spraying their soybeans for aphids this growing season. Several landowner cooperators have indicated that they do not plan on scouting for aphids. Rather, they plan to spray regardless of infestation levels. This approach to soybean management may be a primary reason why reports of aphid resistance to pyrethroid insecticides are increasing in Minnesota and parts of North Dakota this year (UM Extension 2017).

Further results are forthcoming as no field sampling has occurred yet. Our first year of field sampling will occur during late summer 2017 once soybean aphid spraying begins. A second season of field sampling is also planned for summer 2018.

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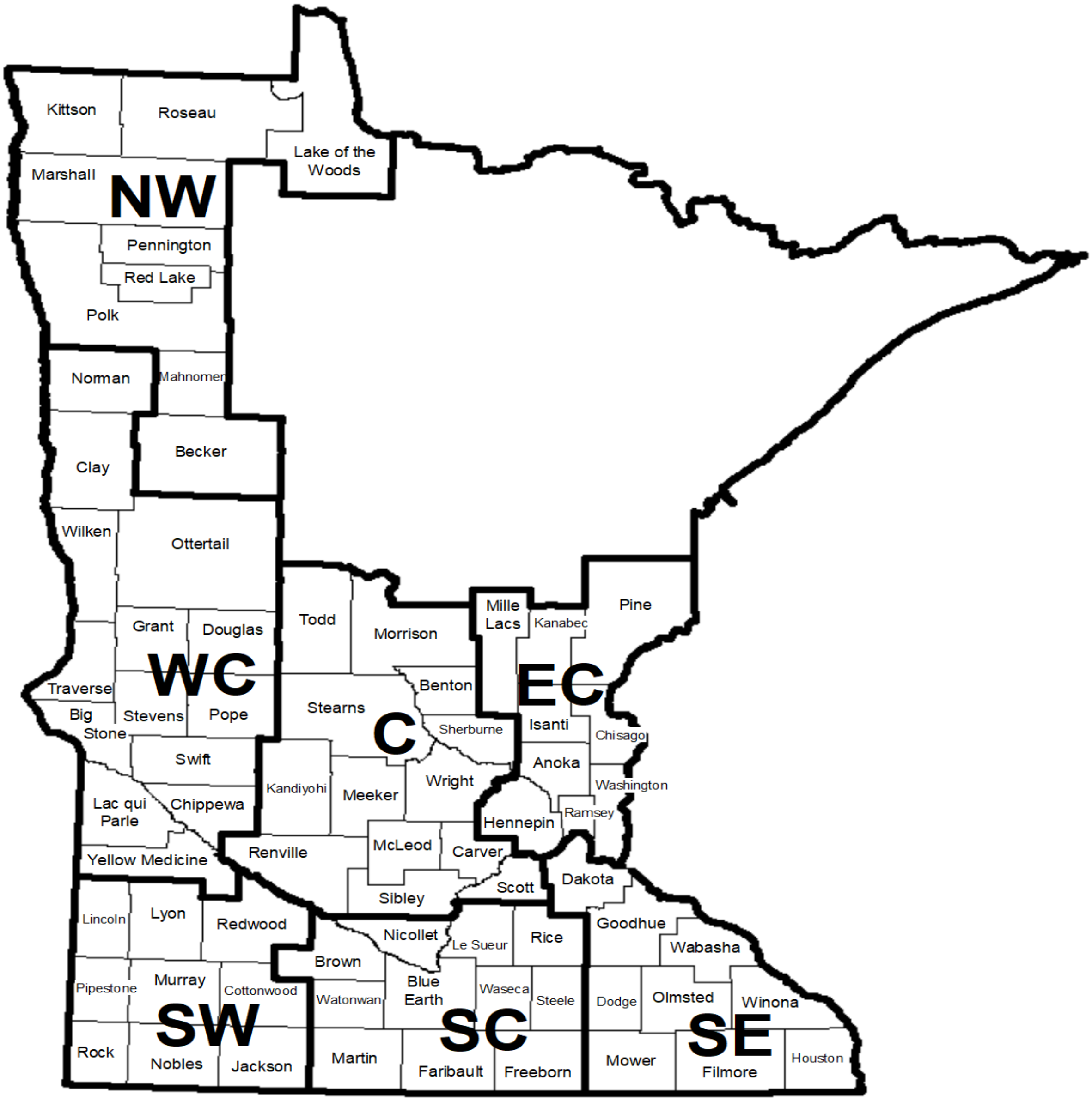
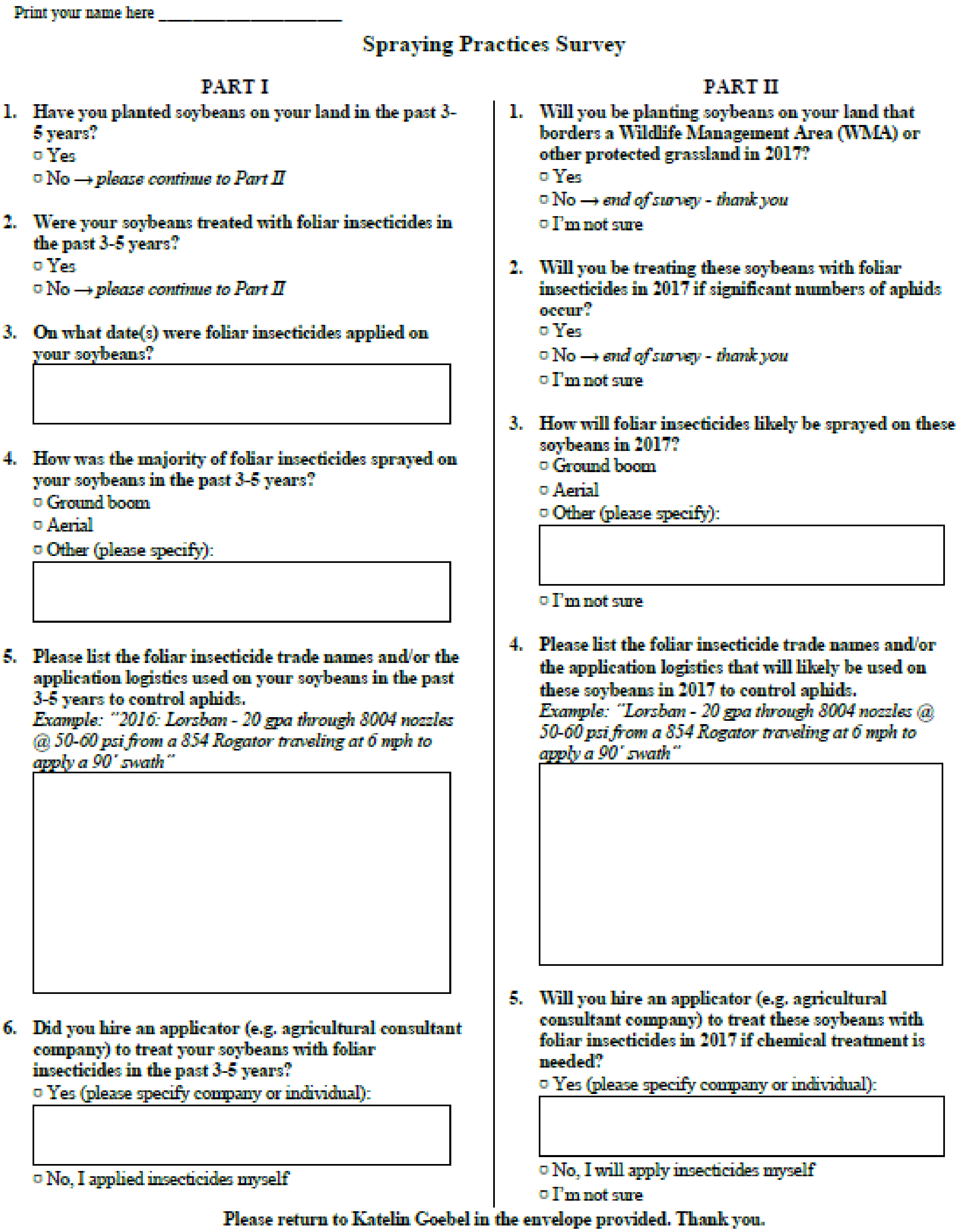


Figure 1. Minnesota's agricultural regions as outlined in MNDNR's annual August Roadside Surveys.

Appendix 1. Survey sent to neighboring landowners (i.e., private landowners with property immediately adjacent to potential Wildlife Management Area study sites) in March and April 2017 to assess soybean aphid spraying practices and to solicit cooperation for summer 2017 sampling efforts.



Print your name here ____________________

Spraying Practices Survey

PART I

1. **Have you planted soybeans on your land in the past 3-5 years?**
   ○ Yes
   ○ No → *please continue to Part II*

2. **Were your soybeans treated with foliar insecticides in the past 3-5 years?**
   ○ Yes
   ○ No → *please continue to Part II*

3. **On what date(s) were foliar insecticides applied on your soybeans?**

4. **How was the majority of foliar insecticides sprayed on your soybeans in the past 3-5 years?**
   ○ Ground boom
   ○ Aerial
   ○ Other (please specify):

5. **Please list the foliar insecticide trade names and/or the application logistics used on your soybeans in the past 3-5 years to control aphids.**
   Example: "2016: Lorsban - 20 gpa through 8004 nozzles @ 50-60 psi from a 854 Rogator traveling at 6 mph to apply a 90' swath"

6. **Did you hire an applicator (e.g. agricultural consultant company) to treat your soybeans with foliar insecticides in the past 3-5 years?**
   ○ Yes (please specify company or individual):
   ○ No, I applied insecticides myself

PART II

1. **Will you be planting soybeans on your land that borders a Wildlife Management Area (WMA) or other protected grassland in 2017?**
   ○ Yes
   ○ No → *end of survey - thank you*
   ○ I'm not sure

2. **Will you be treating these soybeans with foliar insecticides in 2017 if significant numbers of aphids occur?**
   ○ Yes
   ○ No → *end of survey - thank you*
   ○ I'm not sure

3. **How will foliar insecticides likely be sprayed on these soybeans in 2017?**
   ○ Ground boom
   ○ Aerial
   ○ Other (please specify):
   ○ I'm not sure

4. **Please list the foliar insecticide trade names and/or the application logistics that will likely be used on these soybeans in 2017 to control aphids.**
   Example: "Lorsban - 20 gpa through 8004 nozzles @ 50-60 psi from a 854 Rogator traveling at 6 mph to apply a 90' swath"

5. **Will you hire an applicator (e.g. agricultural consultant company) to treat these soybeans with foliar insecticides in 2017 if chemical treatment is needed?**
   ○ Yes (please specify company or individual):
   ○ No, I will apply insecticides myself
   ○ I'm not sure

Please return to Katelin Goebel in the envelope provided. Thank you.

Print your name here ____________________

Contact Information Form

1. **May we contact you to identify foliar insecticide spraying date(s) in the summer of 2017?**
   ○ Yes
   ○ No

2. **What is the best way to reach you?**
   ○ Home phone
   ☐☐☐ – ☐☐☐ – ☐☐☐☐

   ○ Cell phone
   ☐☐☐ – ☐☐☐ – ☐☐☐☐

   ○ Both home & cell phones

3. **In order to identify the exact date(s) of spraying, how often are you comfortable with us contacting you during the late summer of 2017?**
   ○ Weekly
   ○ Semi-weekly
   ○ As often as necessary as the spraying date approaches (no more than once daily)

4. **Would you like to receive a paper copy of the LCCMR work plan for our project?**
   This can also be found at: http://www.lccmr.leg.mn/projects/2016/work_plans_may/_2016_03n.pdf
   ○ Yes
   ○ No

5. **Would you like to receive a paper copy of your responses to the Spraying Practices Survey and Contact Information Form?**
   ○ Yes
   ○ No

6. **If you rent your land, please provide the name and address of your renter so we may send them a letter and survey:**

Please return to Katelin Goebel in the envelope provided. Thank you.

NESTING AND BROOD-REARING HABITAT SELECTION AND SURVIVAL RATES OF RING-NECKED PHEASANTS IN PRAIRIE RECONSTRUCTIONS IN SOUTHWEST MINNESOTA

Nicole M. Davros and Lindsey N. Messinger

SUMMARY OF FINDINGS

Ring-necked pheasant (*Phasianus colchicus*) responses to the amount of grassland in the landscape have been well documented but we lack current information on the individual components of reproductive success (e.g., nest success, brood success, chick survival) that are driving pheasant population dynamics in Minnesota. From early spring 2015 through spring 2017, we radiocollared 122 hens on 2 study sites in southwestern Minnesota and monitored them during nesting and brood-rearing each year. We collected data on nest site selection and hen, nest, brood, and chick survival each year. In 2016 and 2017, we also collected data on brood-rearing habitat selection. Video cameras were used to document nest predation events in 2015 and 2016. Preliminary descriptive findings are described within this report as this study is ongoing and final results are pending. Ultimately, the results will be used to better understand the factors that limit reproductive success of pheasants so that natural resource managers can prioritize their grassland management and land acquisition strategies.

INTRODUCTION

Ring-necked pheasant population dynamics are driven largely by variation in survival rates, and predation is the primary cause of mortality for hens and their young (Peterson et al. 1988, Riley et al. 1998). Predator control efforts can help improve reproductive output over short time periods, but such efforts are economically and ecologically inappropriate over the long-term and at the landscape scale (Chesness et al. 1968, Riley and Schulz 2001). Management aimed to increase pheasant populations has instead focused primarily on providing abundant nesting cover to minimize the effects of predation and maximize reproductive success. As acres enrolled in the Conservation Reserve Program (CRP) and similar cropland retirement programs decline in Minnesota, providing suitable habitat on public lands to sustain populations will become more critical for mediating the effects of predation on pheasant population dynamics. However, the interaction between habitat and predation will no doubt remain. Thus, gaining new insights into the relationship between pheasant habitat selection and subsequent survival rates will be important for improving wildlife management strategies on publicly owned lands.

Predation during the nesting season is a major factor affecting pheasant population dynamics. Nest predation is the leading cause of nest failure for many grassland-nesting birds, including pheasants (Chesness et al. 1968, Clark et al. 1999) and can limit productivity. Additionally, hens take only short recesses from incubating which puts them at greater risk to predation during nesting (Giudice and Ratti 2001, Riley and Schulz 2001). Management efforts aimed at increasing patch size and reducing edge effects are assumed to alleviate rates of predation on birds and their nests (e.g., Johnson and Temple 1990, Sample and Mossman 1997, Winter et al. 2000); however, the composition of the landscape surrounding a patch (Clark et al. 1999, Heske et al. 2001) and the vegetation within a patch (Klug et al. 2009, Lyons 2013) also play important roles in determining susceptibility to nest predation.

Recent advances in video camera technology have allowed better monitoring of bird nests and provided evidence that nest predator communities are more complex than previously thought (Pietz et al. 2012). In particular, the predators associated with nest depredation events can vary with the structure and diversity of nesting cover (e.g., percent cover of litter, forbs, or cool-season grasses; Klug et al. 2009, Lyons 2013). Thus, management actions attempting to mitigate the impact of predators may not necessarily reduce rates of nest predation but rather create a spatial or temporal shift in the nest predator community and susceptibility to nest predation (Benson et al. 2010, Thompson and Ribic 2012). Nest predator communities also vary across regions and habitats and results from studies of other species or in other states may not be entirely applicable to Minnesota's pheasant population (Thompson and Ribic 2012). Understanding how management at the site level (e.g., vegetation structure, composition, and diversity) impacts the dynamics of nest predation is an important but as of yet unintegrated step in our ability to manage habitat for increased productivity of pheasants and other grassland birds (Jiménez and Conover 2001).

Chick survival is also a vital component of pheasant population dynamics but it remains poorly understood (Riley et al. 1998, Giudice and Ratti 2001). Assessing the causes of pheasant chick mortality has been difficult because many previous studies have relied on estimates of brood survival (e.g., the proportion of broods in which ≥1 chick survived to a certain age) rather than survival of individual chicks within a brood (e.g., Meyers et al. 1988, Matthews et al. 2012; but see Riley et al. 1998). Using brood survival estimates is likely unreliable because brood mixing can occur (Meyers et al. 1988; N. Davros, unpublished data). Further, lack of data on individual chicks (e.g., body condition, cause of death) prevents us from understanding the role of different factors (e.g., exposure, food limitation, predation) that lead to variation in recruitment. Evidence that predation is the leading cause of chick mortality for grassland gamebirds in North America is well-established (e.g., Riley et al. 1998, Schole et al. 2011). Food availability has been implicated as an important factor explaining chick survival for many gamebird species in Europe (Green 1984, Hill 1985, Potts 2012); however, strong evidence that food is a major limiting factor for survival of chicks in North America is still lacking. Moreover, food availability and rates of predation likely interact in relation to vegetation structure and composition and confound conclusions from chick survival and food resource studies (Hill 1985). Finally, death from exposure has been shown to decrease chick survival rates, especially after periods with increased precipitation when chicks are still very young and unable to fully thermoregulate (Riley et al. 1998, Schole et al. 2011). Risk of exposure and starvation may interact to decrease chick survival, but few studies have been able to directly address this question (but see Riley et al. 1998). Therefore, better data are needed to understand the interplay between these potential limiting factors on brood habitat selection and chick survival in different grassland habitat types within Minnesota's pheasant range.

Minnesota Department of Natural Resources (MNDNR) wildlife managers in the farmland region have indicated a need for more information on pheasant nesting, brood habitat suitability, and chick survival in relation to management activities. Indeed, better understanding the factors that limit brood production and chick survival will help natural resource agencies prioritize their management strategies at both the local (e.g., forb interseeding or other grassland reconstruction activities) and landscape (e.g., acquisition priorities) levels in this new era of reduced CRP acreages. Additionally, obtaining data on individual components of pheasant population dynamics will aid in future assessment of MNDNR management activities [e.g., Prairie Plan implementation (Minnesota Prairie Plan Working Group 2011), conservation grazing, forb interseeding] and agricultural land use practices (e.g., pesticide use) on Minnesota's pheasant population.

OBJECTIVES

Our overall objective is to evaluate the relative importance of within-patch diversity [e.g., sites dominated by smooth brome (*Bromus inermis*), warm-season grasses, and high diversity grass-forb mixtures] within Wildlife Management Area (WMA) project areas on pheasant productivity. Specifically, we will:

1. Evaluate pheasant nest site selection and nest, brood, chick, and hen survival in relation to vegetation cover and composition.
2. Evaluate pheasant brood-rearing habitat selection in relation to vegetation cover and composition.
3. Evaluate the relative importance of different factors (e.g., predation, weather) on nest, brood, chick, and hen survival.

Results from a pilot study during the 2015 breeding season allowed us to refine methods and protocols for the study's expansion in 2016 and 2017, and the 2017 field season was still underway at the time of this report. Therefore, we present only preliminary results here. A more complete evaluation of results is pending further data analyses.

STUDY AREA

Our study is being conducted in the southwest region of Minnesota. Topography ranges from flat to gently rolling. This region is intensively farmed, and corn and soybeans combined account for approximately 75% of the landscape (U.S. Department of Agriculture 2013a, U.S. Department of Agriculture 2013b). Grassland habitats, including those on private land [CRP, Reinvest in Minnesota (RIM), Conservation Reserve Enhancement Program (CREP), and Wetlands Reserve Program (WRP)] and public land [MNDNR Wildlife Management Areas (WMA) and U.S. Fish & Wildlife Service (USFWS) Waterfowl Production Areas (WPA)] account for 6.3% of the landscape in this region (Davros 2016). The southwest region lies within the core of Minnesota's pheasant range, and MNDNR's 2016 August roadside counts indicated 96.0 pheasants per 100 mi driven (Davros 2016).

We selected 2 WMA project areas as study sites. Each study site is about 9 mi^2 in size and contains extensive amounts of permanently protected grassland habitat. The Lamberton WMA study site (Redwood County) is a large, nearly contiguous WMA complex with >1,100 acres of permanently protected upland and wetland habitats. The Worthington Wells study site (Nobles County) has >1,500 acres of permanently protected habitat that spans multiple WMAs, the Okabena-Ocheda Watershed District, and USFWS lands.

METHODS

Data Collection

We captured hen pheasants in each study site during 5 time periods: 2 February – 15 April 2015, 7 October – 11 November 2015, 11 January – 29 April 2016, 26 September – 15 November 2016, and 18 March – 14 April 2017 (hereafter referred to as spring 2015, fall 2015, spring 2016, fall 2016, and spring 2017, respectively). We used 2 capture techniques: baited walk-in traps and netting via nighttime spotlighting from a 6-wheel utility-task vehicle (UTV). We weighed each hen to the nearest 5.0 g, measured the right tarsus to the nearest 0.5 mm, banded her with a uniquely numbered aluminum leg band, and fitted her with a 16.0-g necklace-style VHF radiotransmitter with integrated mortality switch [Advanced Telemetry Systems (ATS), Isanti, MN] before release.

We began radiotracking hens 3-5 times per week in late April each year to determine the onset of incubation. We assumed incubation had begun when a hen's radio signal was projected from the same location for several consecutive days. We flushed hens from their nests between incubation day 5-20 to determine clutch size and floated a subset of eggs to estimate hatch dates (Westerskov 1950, Carroll 1988). We marked the location of nests using a global positioning system (GPS) receiver. We also placed flagging ≤5 m from nests to aid relocation efforts. If a hen began making large daily movements prior to being flushed, we assumed her nest failed and we waited for her to resettle and begin incubating again before attempting another flush. We used the homing technique on radiocollars emitting a mortality signal to retrieve the collars. We used the condition of the hen's body and/or radiocollar (e.g., teeth marks, feathers plucked, body intact but frozen, frayed collar, missing crimp) and nearby evidence (e.g., predator scat, den site) to determine alive/dead status and potential cause of death, if applicable.

During 2015 and 2016 only, we placed miniature color video cameras (GE 45231 MicroCam Wired Color Camera, Louisville, KY) at a random subset of nests in an attempt to document nest predation events (Cox et al. 2012). Cameras were placed at nests at the same time that hens were flushed to float eggs, and our total time at the nest was ≤20 min. We placed cameras 1-5 m away from the nest bowl at a height of approximately 0.3 m. Cameras had infrared light-emitting diodes (LEDs) to allow recording at night and were connected to digital video recorders (Model MDVR14H, Super Circuits, Austin, TX) with SD memory cards and deep-cycle marine batteries housed in waterproof containers >20 m from nests. Video footage was later reviewed in the office and relevant video clips were archived.

Near the estimated hatch date of known nests, we monitored hen activity 2-3 times daily to pinpoint a hatching event. We assumed hatching was occurring when a hen's signal fluctuated in intensity (Riley et al. 1998). We captured 1-3 chicks by hand between day 0-2 (day 0 = hatch day) once the hen and her brood had moved away from the nest. We used 2 techniques to capture chicks. The first technique involved flushing the hen from her brood and using a decoy and playback to call chicks in. The second technique involved flushing the hen from her brood just before sunrise while she was brooding them and capturing chicks by hand as they scattered. We never captured more than 50% of the brood at one time. We also never kept the hen away from her brood for >30 minutes to minimize risk of hypothermia for the chicks. We discontinued chick capture attempts for a particular brood if we were unsuccessful at capturing any chicks by the end of day 2.

We transported captured chicks in a small cooler or waist belt heated with hand-warmers to a nearby field truck for processing. We determined the mass of each chick to the nearest 0.1 g and we measured tarsus length to the nearest 0.5 mm before suturing a 0.65-g backpack-style VHF radiotransmitter without mortality sensor (ATS, Isanti, MN) to the chick's back (Burkepile et al. 2002, Dahlgren et al. 2010). Handling time lasted <5 min per chick and all chicks were returned to the hen within 30-60 min of capture. We followed the methods of Riley et al (1998) to return chicks to the hen.

We triangulated hens and their broods 2-3 times daily ≥3 times per week. We took each bearing from ≥100 m away to reduce disturbance to the hen and her brood. We then used specialized computer software (LOAS, Ecological Software Solutions LLC) to generate estimated locations. We monitored hens and their broods through the first 4 weeks post-hatching. On day 14 and day 30, we flushed the hen just before sunrise to determine brood status and size.

To estimate individual chick survival, we listened for the signal of each radiomarked chick every 1-3 days in conjunction with monitoring the hen. We relied primarily on fluctuation in the chick's signal to determine if it was alive and moving. If the signal indicated that the chick was not moving, we used the homing technique to locate the transmitter and we searched the area for a carcass and evidence relating to the cause of death.

We collected vegetation data at the nest site within 7 days of hatching for successful nests. For nests that failed, we also collected vegetation data at the nest site ≤7 days after the estimated hatch date. At each nest site, we visually estimated percent canopy cover (Daubenmire 1959) of grasses, forbs, litter, bare ground, woody vegetation, and other (e.g., logs, rocks) using a 0.5 m^2 sampling quadrat. We estimated percent cover on an overlapping basis using 8 classes: 0%, 0.1-10%, 11-25%, 26-50%, 51-75%, 76-90%, 91-99%, and 100%. We estimated litter depth to the nearest cm and we counted the number of grass and forb species to determine species richness within the quadrat. We also recorded visual obstruction readings (VOR; Robel et al. 1970) in the 4 cardinal directions to determine the vertical density of vegetation to the nearest 0.5 dm around the nest and we recorded the maximum height of live and standing dead vegetation within 0.5 m of the Robel pole. We repeated these sampling efforts at 2 random points within 15 m of the nest site.

To evaluate brood habitat selection, we collected vegetation data at 5 estimated brood locations (hereafter, brood points) and 10 random points outside of each brood's biweekly home range until each brood was 4 weeks old. First, we mapped each brood's estimated locations in a Geographic Information System (GIS; ArcMap 10.2, ESRI, Redlands, CA) to estimate their biweekly home range. We defined each biweekly home range as the area bounded within all estimated brood points for that 2-week time period. We placed a 100 m buffer around the home range and used a random point generator in ArcMap to select 10 random points outside of the home range for comparison. We restricted the selection of random points so that they were within the same habitat type (e.g., grassland). Roadsides were considered as available grassland habitat and included in sampling efforts. We then collected vegetation data at each brood point and each random point within 7 days of the biweekly interval. At each brood point, we sampled 1 center point and 3 equidistant points 10 m away to capture the spatial variation of a brood location. We estimated percent canopy cover, litter depth, species richness, VOR, and maximum height of live and dead vegetation using the same methods described above for nest site selection. We repeated this sampling scheme at each of the 10 random points associated with each brood's biweekly home range. We restricted the sampling of brood habitat selection to field types other than row crops. If a hen and her brood spent more than 50% of their time in a row crop field during the 2-week period of observation, we did not include them in habitat sampling efforts. If more than one hen with a similar-aged brood was using the same habitat patch during the same time period, we only sampled 5 additional random points within that patch. Finally, we did not collect brood habitat data if a hen lost her entire brood within the first week of each 2-week observation window.

Data Analyses

To date, we have conducted preliminary analyses on hen survival and nest survival. We also calculated basic descriptive statistics for nest site selection. Data proofing for 2016 and data collection for 2017 were still ongoing at the time of this report; thus, not all analyses have included the 2016 and/or 2017 data and not all research objectives are addressed below.

We conducted a preliminary survival analysis to evaluate adult hen survival during the nesting and brood-rearing phases (15 April – 15 October; hereafter, breeding season) only. For hens captured in 2015 and 2016, we estimated cumulative survival using a Kaplan-Meier analysis approach in R v3.3.2 (R Core Team, 2016). The Kaplan-Meier approach assumes a known fate for each individual. As such, 10 individuals were censored at various intervals during the analysis period when they were reported missing and not relocated or their fate was otherwise reported as uncertain (e.g., slipped radiocollar, radiocollar malfunction, etc.). Individuals with capture and mortality or censor events occurring outside of the analysis period were excluded from the analysis.

Using the 2015 data only, we conducted a preliminary nest survival analysis using the logistic-exposure method (Shaffer 2004) to estimate daily survival rates (DSR) of nests. We used a constant survival model (PROC GENMOD; SAS v9.3; SAS Institute, Cary, NC) which assumes that survival is constant across time and does not include any nest-specific explanatory variables.

RESULTS AND DISCUSSION

We captured 122 hens during the 5 trapping periods across both sites from spring 2015 to spring 2017 (Table 1). The baited walk-in traps were not a productive capture technique. We speculate that pheasants were not motivated to use bait due to mild winter conditions with above-average food availability each year. Only 3 hens were captured using the walk-in traps (2.5%) whereas 119 hens (97.5%) were captured by spotlighting. We ended spotlighting capture efforts at the onset of the breeding season which limited our ability to increase sample sizes. In the future, we would consider using baited walk-in traps in late winter if weather conditions were severe enough to warrant this method. Winter conditions are considered severe for pheasants when snow is ≥6 inches deep and temperatures reach ≤0° F.

In 2015 and 2016, cumulative survival during the breeding season (183 day period pooled across years) for adult hens (*n* = 64) across study areas was 0.79 (CI: 0.69-0.90; Figure 1). During the 2 breeding seasons, 20% of marked individuals (*n* = 12) suffered a known mortality event (Figure 2). Of these mortality events, 75% were attributed to predation events, 17% to human causes (specifically, vehicle collision and agricultural equipment), and <1% to research-related marking. Although the Kaplan-Meier survival method provides a quick estimate of hen survival, the strict assumptions of this model may be inappropriate given our dataset. Because nearly 14% of individuals were censored during this analysis due to unknown fates (in particular, slipped radiocollars; Figure 2), subsequent survival analyses will work to include expert knowledge to incorporate uncertainty in fate to refine survival estimates (A. Norton, personal communication).

Due to mortalities (*n* = 3) and dropped collars (*n* = 2; unknown causes), we were able to monitor only 15 hens during the 2015 nesting season. One unmarked hen was flushed incidentally during field work and her nest was also monitored. Therefore, we monitored a total of 22 nests from 16 hens. Four nests were abandoned presumably due to research-related activities; therefore, we excluded them from our analysis of nest success. Twelve of 18 nests hatched successfully (67% apparent nest success). The 2015 DSR was 0.9406 ± 0.41 (range: 0.8731-0.9729) which results in an 11.7% overall nest success rate when extrapolated to a 35-day nesting cycle (12 days laying + 23 days incubation). We used a constant survival model due to our low sample sizes; however, future analyses will examine the role of vegetation, spatial (e.g., distance to edge), and temporal (e.g. nest age, ordinal date, year) covariates on nest DSRs. In particular, time-specific patterns of nest survival have been documented in several duck and passerine species (Grant et al. 2005, Grant and Shaffer 2012) and such analyses are likely more appropriate for pheasants given their long nesting cycle and extended breeding seasons.

During 2015 and 2016 only, we placed video cameras on approximately 40% of nests each year. Most hens were tolerant of cameras but a few hens did abandon their nests. However, these hens likely did not abandon due solely to cameras as hens not receiving cameras at their nests were also prone to abandonment, especially if flushed during early incubation. Notable observations included a rooster visiting a hen at her nest almost daily during late incubation (Figure 3) and a chick appearing on video 3 h prior to its hen leading her brood away from another nest. We potentially captured 2 predation events on camera in 2016 but the video qualities were low due to vegetation growth and windy conditions which greatly reduced our ability to clearly view activity at the nests. Although all nests were visible when cameras were first placed, the rapid growth of vegetation during the nesting cycle quickly impacted our ability to view nest contents or activities in the immediate area. Windy conditions often compounded our inability to review camera footage by causing vegetation to blow in front of the camera. In the future, we would consider using cameras again to document nest predation events but we would alter our camera set-up (e.g., distance to nest, height of camera) to reduce the impact of vegetation and wind on the quality of the footage.

We captured and tagged 81 chicks between day 0-2 during the 2015-2017 breeding seasons. During the 2016 and 2017 field seasons, we recaptured 3 chicks between day 12-15 and replaced their 0.65 g transmitters with sutured 1.1-g backpack-style transmitters (ATS, Isanti, MN). Recapturing radiomarked chicks at this age was relatively easy and seems like a viable option to replace lighter transmitters with heavier ones that have a longer battery life, thereby allowing monitoring of chicks beyond 4 weeks of age in future work. Similar to our hen survival analyses, future chick survival analyses will use additional information from these individuals to refine survival estimates when fates are uncertain.

We collected vegetation data from 19 nest sites in 2015. We calculated means and standard errors (SE) for 2 groups of comparisons: nest sites versus random points (Table 2), and successful versus depredated nests (Table 3). We included all nests regardless of nest fate (e.g., successful, depredated, abandoned, other failure) for the comparison of nest sites versus random points. Hens seemed to use nest sites with slightly less grass cover, lower total species richness, lower grass species richness, and shallower litter depth compared to random points nearby. Hens that successfully hatched a nest in 2015 appeared to use nest sites with less grass and forb cover but more standing dead vegetation cover, reduced species richness of both grasses and forbs, and reduced VOR. Sample sizes for both of these comparisons are low, however, and more data are needed to make formal comparisons.

The last field season of data collection is currently underway. The final results from this study will relate pheasant survival rates to nesting and brood-rearing habitat selection. Ultimately, the information gained will help managers better understand the factors that may limit pheasant productivity so that they can prioritize their management activities in an era of reduced grassland habitat on the landscape.

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Table 1. Ring-necked pheasant hen captures by season[a] and method[b] in southwestern Minnesota, 2015-2017.

	Spring '15		Fall '15[c]		Spring '16		Fall '16[c]		Spring '17[c]		
	BWT	Spot	BWT	Spot	BWT	Spot	BWT	Spot	BWT	Spot	Totals
Study Area											
Lamberton	2	8	.	8	1	12	.	12	.	18	61
Worthington Wells	0	10	.	11	0	15	.	9	.	16	61
Totals	2	18	.	19	1	27	.	21	.	34	122

[a] Season dates include: Spring 2015 = 2 February-15 April; Fall 2015 = 7 October-11 November; Spring 2016 =11 January-29 April; Fall 2016 = 26 September-15 November; Spring 2017 = 18 March-14 April.

[b] Capture methods included: baited walk-in traps (BWT) and netting via nighttime spotlighting from a utility-task vehicle (Spot).

[c] Walk-in traps were not used during fall trapping efforts or during Spring 2017.

Table 2. Descriptive statistics for vegetation surveys at sites used for nesting by ring-necked pheasant hens and nearby random points (≤15 m away) as a comparison in southwestern Minnesota. Data are shown for 2015 only.

	Nest sites (*n* = 19)		Random points (*n* = 19)	
	Mean	SE	Mean	SE
% Canopy cover[a]				
Grasses	3.4	0.30	3.6	0.18
Forbs	0.9	0.22	1.0	0.20
Standing dead	1.6	0.14	1.5	0.14
Species richness				
Total	3.3	0.62	3.6	0.51
Grasses	1.4	0.14	1.7	0.18
Forbs	1.8	0.59	1.8	0.44
Litter depth (cm)	2.8	0.43	3.3	0.46
VOR (dm)[b]	4.9	0.50	5.1	0.42

[a] Means and SEs for canopy cover measurements were transcribed into cover classes for analysis and have not been back-transcribed. Cover classes include: 0 = 0%, 1 = 0.1-10%, 2 = 11-25%, 3 = 26-50%, 4 = 51-75%, 5 = 76-90%, 6 = 91-99%, and 7 = 100%.

[b] VOR is the average visual obstruction reading as determined by using a Robel pole.

Table 3. Descriptive statistics for vegetation surveys at successful versus depredated nest sites of ring-necked pheasants in southwestern Minnesota during summer 2015 only.

	Successful Nests (*n* = 9)		Depredated Nests (*n* = 4)	
	Mean	SE	Mean	SE
% Canopy cover[a]				
Grasses	3.4	0.44	3.8	0.85
Forbs	0.9	0.20	1.8	0.48
Standing dead	1.7	0.17	1.0	0.00
Species richness				
Total	3.1	0.56	6.8	1.65
Grasses	1.4	0.18	1.8	0.48
Forbs	1.6	0.53	4.8	1.93
Litter depth (cm)	2.3	0.55	2.3	1.41
VOR (dm)[b]	4.3	0.53	6.2	1.95

[a] Means and SEs for canopy cover measurements were transcribed into cover classes for analysis and have not been back-transcribed. Cover classes include: 0 = 0%, 1 = 0.1-10%, 2 = 11-25%, 3 = 26-50%, 4 = 51-75%, 5 = 76-90%, 6 = 91-99%, and 7 = 100%.

[b] VOR is the average visual obstruction reading as determined by using a Robel pole.

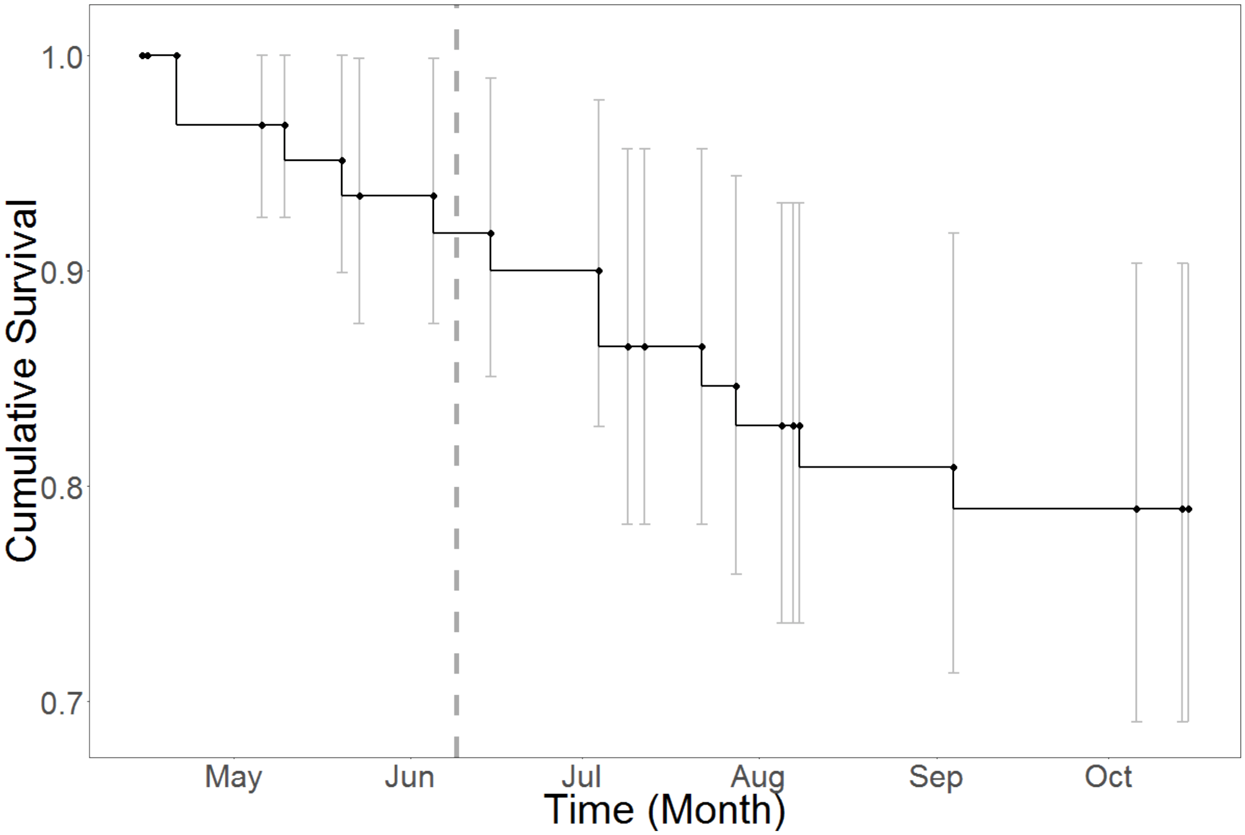

Figure 1. Cumulative survival of radiocollared ring-necked pheasant hens during the 2015 and 2016 breeding seasons (15 April – 15 October) in southwest Minnesota. Points represent survival estimates at intervals where mortality events took place. Error bars (vertical gray lines extending from each point) represent the upper and lower 95% confidence interval for each survival estimate. The 10-year average (2007-2016) for peak hatch of pheasant nests in Minnesota, as estimated by MNDNR's annual August roadside count surveys, is 12 June and is shown with the vertical gray dashed line.

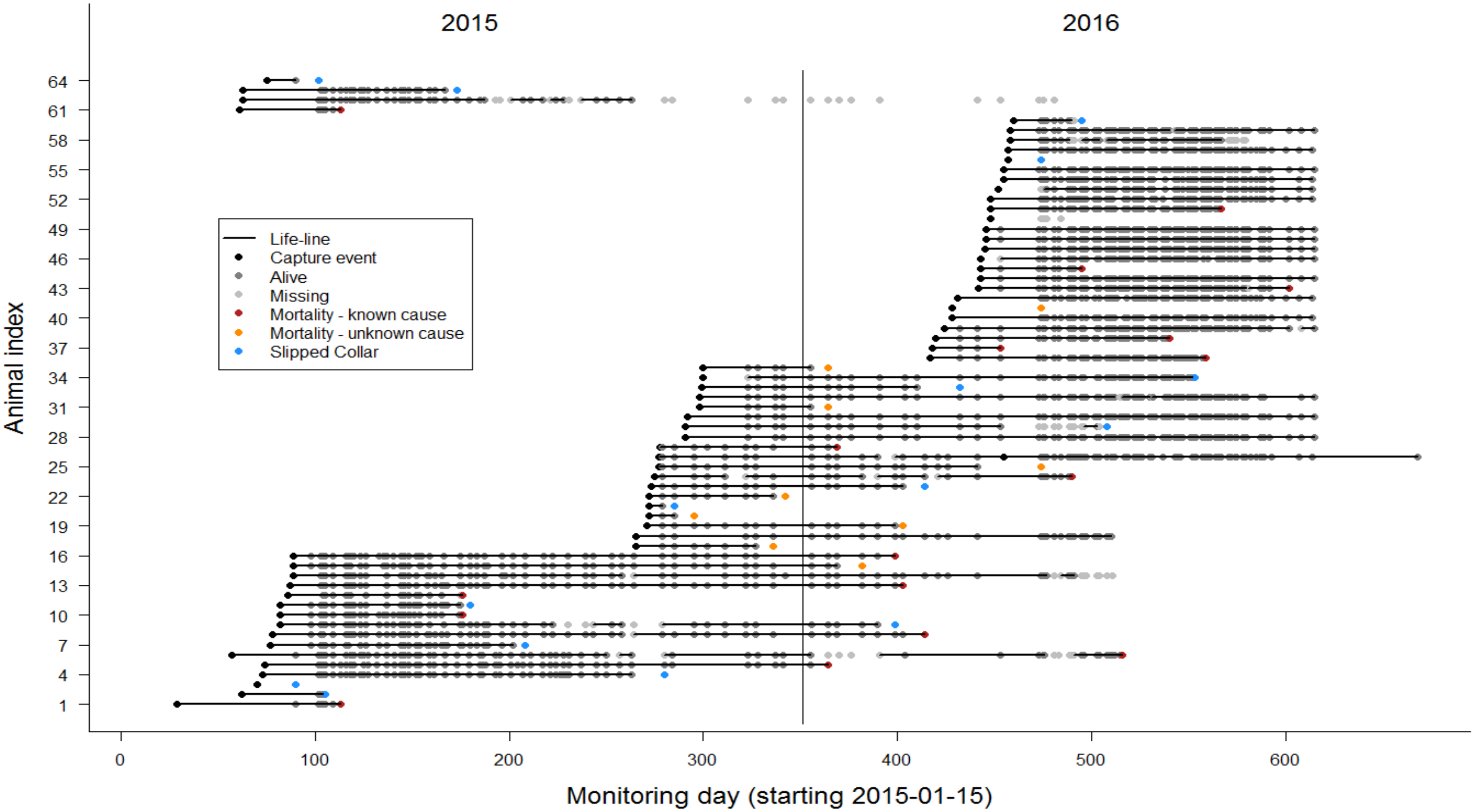

Figure 2. Survival and monitoring history of 64 ring-necked pheasant hens captured and radiocollared from 1 January 2015 – 15 October 2016 in southwest Minnesota. Each row in the figure represents an individual hen. For each individual, the first black point represents the capture date. Subsequent points indicate the monitoring frequency and status of each hen over time. Points for each hen are connected by a black “life-line” if the hen status is alive at both points. Breaks in the line indicate periods were the individual was monitored but not located and the line ends with a mortality event or the end of the monitoring interval specified above. The terminal point for each hen is colored to indicate the cause of mortality [red = known-cause mortality events (e.g., predation, vehicle collisions, mowing/haying operations, etc); orange = unknown-cause mortality events or end of monitoring status; blue = slipped radiocollar; dark gray = alive; light gray = missing].

Figure 3. A ring-necked pheasant rooster visits a hen at her nest during late incubation in southwest Minnesota during May 2015.

ASSESSING THE CONTRIBUTION OF HARVEST MORTALITY TO OVERALL MORTALITY RATES OF GRAY AND FOX SQUIRRELS ON PUBLIC LANDS IN MINNESOTA

Ryan G. Tebo and John H. Giudice

SUMMARY OF FINDINGS

Squirrel hunting is a popular activity in Minnesota, but hunter perceptions of squirrel decline on publicly-owned land near the Twin Cities Metropolitan Area of Minnesota (hereafter, metro) has led interested user groups to voice concern over squirrel populations in recent years. To explore these concerns, we conducted a study to compare squirrel mortality rates on a site with heavy hunting pressure (Whitewater Wildlife Management Area (WWMA) to a paired non-hunted site (Whitewater State Park (WSP) in southeastern Minnesota. From 1 July 2015 – 18 September 2015 and 2 May 2016 – 16 September 2016, we trapped and radiocollared 107 gray and fox squirrels (*Sciurus carolinensis* and *S. niger*, respectively) on WWMA and 101 gray squirrels on WSP. Using survival data from the first year of the study, we failed to find evidence that survival probabilities differed between sites during most of the monitoring period (1 July 2015 – 12 April 2016). The estimated survival probability for late summer (1 July 2015 – 18 September 2015) on both sites was 0.916 (85% CI: 0.871-0.946) and for the late fall through early spring monitoring interval (1 November 2015 – 12 April 2016) was 0.835 (85% CI: 0.752-0.892). However, during the first 6 weeks of the 2015 hunting season, 13 squirrels were harvested on WWMA and the survival probability was estimated to be 0.529 (85% CI: 0.398-0.645). Conversely, survival probability was estimated to be 0.955 (85% CI: 0.929-0.971) on WSP during the same time period. We again saw a high number of squirrels harvested during the first 6 weeks of the 2016 hunting season (10), but harvests were also recorded as late as 21 February 2017. Estimates of survival from the first year of the study may show a positive bias due to the high proportion of animals with unknown fates (e.g. missing animals, unrecoverable collars in mortality mode). Monitoring of squirrels radiocollared in 2016 has recently come to an end and with the addition of the second year of data we intend to construct more sophisticated survival models that reflect likely variation in survival probabilities over space, time, and individual covariates.

INTRODUCTION

Small game hunting is a popular recreational activity in Minnesota with approximately 292,000 hunters buying licenses each year since the late 1990s (Dexter 2009, Dexter 2014). Nearly 15% of small game hunters pursue gray and fox squirrels with an estimated take of 5.1 gray and 3.9 fox squirrels per hunter (Dexter 2009, Dexter 2014). The combined gray and fox squirrel harvest and the number of squirrel hunters has each declined by 13.8% since the late 1990s; however, the combined take per hunter has declined by only 3.3% during this same time (Dexter 2009, Dexter 2014). In an effort to better understand barriers to hunter participation, the Minnesota Department of Natural Resources (MNDNR) conducted a survey of squirrel hunters (Dunbar 2009). More hunters in the metro responded that they believed squirrel populations were declining (51%) as compared to other hunters statewide (19%). Metro hunters also indicated that they had limited access to private land and heavy hunting pressure existed on publicly-owned land (Dunbar 2009).

Many factors cause squirrel populations to fluctuate naturally (e.g., mast abundance, population density, disease outbreaks; see Barkalow et al. 1970, Nixon et al. 1974, Nixon et al. 1975, Healy and Welsh 1992, Descamps et al. 2009, Vander Haegen et al. 2013), and population dynamics are determined by reproduction, immigration, emigration, and mortality. Although squirrels are considered a game species where hunting mortality is often assumed to be compensatory to natural mortality, previous research suggests hunting mortality can be additive to non-hunting mortality in exploited squirrel populations (Herkert et al. 1992). If so, wildlife managers can alter hunting regulations to adjust the contribution of hunting mortality to overall mortality rates.

The number of gray and fox squirrels harvested during fall hunting season is correlated with pre-hunt densities and the amount of effort expended by hunters early in the hunting season (Nixon et al. 1975). A study in Virginia found the rate of population turnover, the annual mortality rate, and the rate of juvenile recruitment was higher in hunted squirrel populations (Mosby 1969). In an Ohio study, approximately 50% of the harvest was comprised of squirrels born the previous spring and summer and there was no difference observed in the harvest sex ratio for young-of-the-year or adult squirrels (Nixon et al. 1975). Annual mortality rates were 70-80% for young-of-the-year squirrels and nearly 80% for adult squirrels, with hunting accounting for nearly 60% of the annual mortality across all age classes (Nixon et al. 1975). The researchers concluded that the population could not sustain itself with this level of mortality and that squirrels were likely immigrating from surrounding habitats (Nixon et al. 1975). In Illinois, the annual mortality rate for fox squirrels was 79% on a hunted site and 44% and 45% on non-hunted sites, while mortality rates outside of the hunting season were not significantly different (Herkert et al. 1992). Reproductive intensities did not differ between the sites and the researchers also concluded that the hunted population was being sustained by immigration (Herkert et al. 1992). In a prior Minnesota study, researchers found an annual survival rate of 27% for gray squirrels on a hunted site with 74% of all mortalities attributed to hunting (Longley 1963). The annual survival rate was 48% the following year when the site was not hunted (Longley 1963). Reproductive rates were 2.4 young per litter regardless of the prior year’s squirrel hunt status (Longley 1963).

The MNDNR Section of Wildlife has considered changes to the squirrel season structure in the metro based on the aforementioned survey results. However, because a paucity of information exists with respect to the impacts that the current hunting regulations have on squirrel population growth rates, no changes have been made to date. This study intends to assess the contribution of harvest mortality to overall mortality rates of gray and fox squirrels on public lands in Minnesota. Prior to initiating this expanded research project, we completed a pilot study to evaluate squirrel trapping, handling, and tracking methods (see Curtis and Davros 2014).

OBJECTIVE

1. Assess mortality rates of radiocollared squirrels across multiple seasons (fall/early hunting season, winter/late hunting season, spring, and summer) on heavily-hunted public land and nearby non-hunted land.

STUDY AREA

Our study was conducted in southeastern Minnesota during 2015-2017. Within the study area, we chose 2 sites in close proximity to achieve a paired design of 1 treatment (i.e., hunted) and 1 control (i.e., non-hunted) site. Whitewater Wildlife Management Area, which receives pressure from squirrel hunters, was the treatment site. Whitewater State Park is not open to hunting and was used as the control site.

METHODS

Trapping

We trapped gray and fox squirrels during July-September 2015 and May-September 2016. We used wire box traps (48 x 15 x 15 cm; 2.5 x 1 cm mesh) baited with sunflower seeds, dried corn, peanut butter, and/or black walnuts. Using the MNDNR Forest Inventory layer (where available) or the MNDNR Landcover layer, we selected oak habitat within our sites in ArcGIS 10.2 (ESRI, Redlands, California, USA). In oak habitat areas where terrain allowed, we created a grid of points 25 m apart and placed traps at these points. In areas with challenging terrain or insufficient contiguous oak habitat, traps were selectively placed at locations that researchers deemed to be likely squirrel-use areas. We checked traps at least twice per day (i.e., late morning and late evening) to reduce the amount of time squirrels remained in the traps. We closed traps during inclement weather. We also removed traps before the weekends, thereby allowing us to clean and repair traps before changing sites, reducing the risk of theft, and reducing disturbance to WSP and WWMA visitors during peak visitation days.

Upon capture, we identified the sex of each squirrel and determined the reproductive status of females. We weighed squirrels in the trap to the nearest 10 g using a digital hanging scale. We used a modified handling cone to restrain squirrels, which allowed us to handle and radiocollar without sedation (Koprowski 2002). Handling cones were constructed of denim with hook and loop straps to help secure the squirrel and a zipper opening to allow access to the head and neck during collar attachment. Once in the handling cone, a removable plastic funnel was attached around the squirrel's neck to protect handlers from bites during collaring (McCleery et al. 2007). We only collared squirrels weighing ≥300 g in an effort to keep the transmitter weight below 3% of the animal's body mass. Squirrels received a 7-g, 10-g, or 13-g VHF necklace-style radiocollar (models M1525 and M1535, Advanced Telemetry Systems, Isanti, MN) depending on the weight of each individual squirrel. Each transmitter was equipped with an integrated mortality sensor that changed the pulse rate of the signal if an animal did not move for 12 h. Expected battery life was 362 days for the 13-g collar and 302 days for the 7- and 10-g collars. Upon completion of the handling procedure, all squirrels were immediately released. All non-target captures were released immediately. No specimens were collected.

Tracking

Following capture, radiocollared squirrels were monitored for mortality weekly using a vehicle-mounted non-directional radiotelemetry antenna system. Beginning on the first day of the squirrel hunting season (19 September 2015 or 17 September 2016), squirrels were monitored twice weekly until the end of October. Squirrels were then monitored once weekly until the end of December, and once biweekly until 1 March. When a radiocollar transmitted a mortality signal, we used homing techniques in an attempt to recover the collar and determine the cause of mortality. Squirrels remaining on the air beyond 1 March continued to be monitored biweekly until death or battery failure.

Data Analysis

Mortality is a continuous time process, but in this study we observed it discretely and incompletely (interval-censored and truncated monitoring data with staggered entry, missing animals, unknown fates, and uneven monitoring intervals). This data-collection design shares many similarities with nest-survival studies (Heisey et al. 2007). Therefore, we used a logistic-exposure modeling approach (Shaffer 2004) with interval- or right-censoring of animals with unknown fates (Bunck et al. 1995) to conduct an exploratory analysis of the survival process using monitoring data from squirrels captured during the 2015 trapping season. We used the glm function in the R programming language (R Core Team 2016) with a user-defined link function (https://rpubs.com/bbolker/logregexp) to estimate daily survival rates (DSR) as a function of site, study time (relative to 01 July 2015) or time groups (seasons), and individual covariates (e.g., sex, body mass, collar weight, capture location [x,y], distance from capture location to public roads). For the exploratory analysis, we did not attempt to model competing risks (harvest vs. natural mortality) and instead focused on estimating overall survival probabilities on the 2 study sites. However, for analysis purposes, we treated data from the WWMA during the first 6 weeks of the hunting season (19 Sep 2015 – 31 Oct 2015) separately because all observed harvest mortalities in 2015 occurred during this period. Thus, we used 2 datasets for survival estimation: 1) WWMA data from the first 6 weeks of the hunting season, and 2) all remaining data from both sites (which generally described the natural mortality process). We used Akaike's Information Criterion (AIC) to select among competing models, including a null model with constant daily survival. For inference, we used the most parsimonious model (fewest model parameters) that was within 2 AIC units of the best-approximating model (lowest AIC value).

An important assumption in this type of analysis is that censoring is independent of fate. Overall, 41% of our study animals had some form of censoring that reflected unknown fates (either interval status or final fate). Further, there were 4 times as many "missing" animals on WWMA (vs. WSP) that disappeared during the hunting season. Thus, we were concerned that censoring might not be independent of fate, which can lead to positively biased estimates of survival. We used a replicated imputation algorithm with simulated survival parameters (informed by the data) to construct complete histories for each animal with missing data or unknown fates. We used this procedure to construct 300 replicate monitoring datasets, which we fit to the models selected above (based on the observed data) to produce estimates of daily survival probabilities. We used the estimates from the imputation datasets as our point estimate and the conditional variance formula to compute the variance in the daily survival rate. We then extrapolated both imputed and observed daily survival rates to obtain annual survival probabilities, which we used to quantify potential biases due to censoring. We did not attempt to vary the simulated survival parameters (other than allowing the survival process to vary as a function of binomial variation); thus, differences between imputed and observed annual survival probabilities do not include uncertainty in the simulated survival parameters. We have included the information gleaned from the first monitoring year's exploratory analysis and are currently working on more sophisticated methods to analyze the full monitoring dataset. However, this approach was sufficient to inform our exploratory analysis and provide guidance on analysis options for the full monitoring dataset (years 2015-2017).

RESULTS AND DISCUSSION

Trapping and Monitoring

We successfully captured 132 squirrels (119 gray, 13 fox) during the 2015 and 2016 trapping seasons on WWMA, 107 of which received radiocollars. In an effort to distribute captures throughout WWMA, trapping locations were spread out across the unit (Figure 1). Fifty-one males and 56 females were collared. Based on their lactation status, 21 female squirrels that received collars (38%) showed evidence of prior reproductive activity. Weights of collared squirrels on WWMA ranged from 410-920 g ($\bar{x}$ = 655 g).

One hundred twenty-four squirrels (123 gray, 1 fox) were captured on WSP during the 2015 and 2016 trapping seasons, 101 of which received radiocollars. Trapping locations were spread out across the interior of WSP (Figure 2). Fifty males and 51 females were collared. Of 49 radiocollared females checked for reproductive status, 22 (45%) showed signs of prior reproductive activity. Weights of collared squirrels on WSP ranged from 370-840 g ($\bar{x}$ = 649 g).

From 1 July – late August 2015, trapping success was very low on both sites; only 10 squirrels had been captured on each site (3,564 trap checks; 0.56% capture rate). However, following this period of low capture success, we switched to fresh-picked walnuts as our bait type. In the remaining 3 weeks of the 2015 trapping season following the bait change, 46 and 39 squirrels were captured on WWMA and WSP, respectively (1,181 trap checks; 7.2% capture rate). Thereafter, we used fresh walnuts as the predominant bait-type when they were available. The preference for walnuts may have been due in part to a seasonal behavior change, where caching food became the ultimate goal of squirrels once trees began producing the year's walnut crop. Overall capture rates for WWMA and WSP for the duration of the study were 1.99% (6,629 trap checks) and 5.02% (2,470 trap checks), respectively. Two squirrels on WWMA and 4 squirrels on WSP originally captured in 2015 were recaptured during the 2016 trapping season and fitted with new radiocollars.

Twenty-nine (27.88%) squirrels were harvested by hunters on WWMA (Figure 3). In addition, 9 (8.65%) animals were lost to natural mortality events, 9 (8.65%) squirrels dropped their collars, 13 (12.50%) collars were unrecoverable (e.g., in a tree or inaccessible), 1 (0.96%) collar was recovered but fate could not be determined, 19 (18.27%) squirrels went missing due to unknown reasons, and 24 (23.07%) squirrels went missing due to presumed collar battery failure (Figure 3). On WSP, 21 (20.79%) squirrels were lost to natural mortality events, 15 (14.85%) squirrels dropped their collars, 20 (19.80%) collars were unrecoverable, 8 (7.92%) squirrels went missing due to unknown reasons, and 37 (36.63%) squirrels went missing due to presumed collar battery failure (Figure 3). We censored 3 of the 107 radiocollared squirrels on WWMA from the survival study due to capture complications.

Of squirrels captured in 2015, all known harvest mortalities (13; 100%) occurred within the first 6 weeks of the hunting season (Figure 4). The last observed harvest mortality was 27 October 2015. In 2016, however, 10 of 15 (67%) known harvests occurred during the first 6 weeks of the hunting season. The latest harvest mortality recorded was 21 February 2017. In addition, 8 of 12 (67%) squirrels that went missing for reasons not deemed to be collar battery failure went missing during the first 6 weeks of the hunting season in 2015. Four of 7 (57%) missing squirrels did so during the first 6 weeks of the 2016 hunting season. Although it cannot be confirmed, we believe it is very likely some, if not most, of these squirrels were harvested.

Despite efforts to inform and encourage hunters to report harvests of radiocollared squirrels, only 4 of the 29 (14%) known harvests were reported. Signs placed throughout WWMA and at popular parking areas asked hunters to report the harvest of any radiocollared squirrels. Additionally, squirrels captured in 2016 each received a unique numbered ear tag that included a call back telephone number. However, most radiocollars from harvested squirrels were found cut off in the presumed location of harvest, or in parking lots. Other squirrels were found deceased with noticeable gunshot wounds, but were left in the woods or unrecovered by hunters.

The majority (85.29%) of unrecoverable collars were in trees. Whether these squirrels died or dropped their collars is unknown; however, we believe that a majority of these losses are due to dropped collars. We base this assumption on numerous examples where collars fluctuated between mortality and normal signal, an indication that the collars were still subject to movement in the tree cavities the collars were presumed to be in. On another occasion, a collar was emitting a mortality signal until researchers reached the tree; at that point the collar switched out of mortality mode and an uncollared squirrel exited a cavity in the tree. Finally, one collar that had been unrecoverable in a tree for months was found on the ground by researchers in late March, with clear signs that the zip-tie attachment had been chewed through.

Survival Analysis

Our analysis of the first year of survival data failed to present strong evidence that the log odds of survival varied as a function of site (excluding the first 6 weeks of the hunting season on WWMA), time, season (again, excluding the first 6 weeks of the hunting season on WWMA), or the individual covariates we examined for squirrels captured during the first year of the study. Thus, we used constant-survival models (null models) to make inferences on the survival process, at least for our exploratory analysis. We acknowledge that constant daily survival is unlikely to be true, especially over long periods of time, but given the paucity of monitoring data for some seasons (late summer on both sites and winter-spring on WWMA) and concern about the amount of censoring in our data, it was a reasonable starting point for the exploratory analysis. With the second year of survival data, we anticipate being able to construct more sophisticated survival models that reflect likely variation in survival probabilities over space, time, and individual covariates (e.g., age, sex, distance). For example, we found weak signals that suggested the log odds of harvest decreased with distance from capture site to public roads (i.e., access points for hunters) on WWMA (Figure 5), and the log odds of survival in WSP and WWMA (excluding the first 6 weeks of hunting season) was lower during late summer compared to fall, winter, and early spring (Figure 6). The latter might reflect dispersal and greater vulnerability of juvenile squirrels to natural mortality events, but we lacked a sufficient sample size during this time period (due to trapping challenges) to precisely estimate the effect on survival probabilities.

The estimated daily survival rate (DSR) on WWMA during the first 6 weeks of the 2015 hunting season was 0.985 (85% CI: 0.978-0.990). Conversely, the estimated DSR for WWMA during other times of the year and in WSP was 0.999 (85% CI: 0.998-0.999). Extrapolating the DSRs to seasonal time intervals resulted in an estimated survival probability of 0.529 (85% CI: 0.398-0.645) for WWMA during the first 6 weeks of the hunting season, compared to 0.955 (85% CI: 0.929-0.971) for WSP during the same time period (Figure 7). The estimated survival probability for late summer (1 July 2015 – 18 Sep 2015) on both sites was 0.916 (85% CI: 0.871-0.946) and for the late fall through early spring monitoring interval (1 Nov 2015 – 12 Apr 2016) was 0.835 (85% CI: 0.752-0.892).

Not surprisingly, our imputation analysis suggested that censoring was not independent of fate and, thus, survival estimates based on the censored data were positively biased for both sites (Figure 8). The degree of bias should be interpreted cautiously because it was based on one set of survival parameters (assumptions). Nevertheless, it suggests that censoring by itself may not be sufficient to generate accurate estimates of survival given the limitations of our data (e.g., the presence of many animals with unknown fates and the likely lack of independence between censoring and fate). One potential solution is to use a Bayesian integrated survival analysis where the probability of relocation and survival for animals with unknown fates is estimated via a mark-resight approach (Walsh et al. 2015). We are exploring this option for analyzing the full monitoring dataset (years 2015-2017). Despite the potential bias in survival estimates caused by unknown fates, it is still clear that squirrels subjected to hunting pressure have much lower survival rates during the first 6 weeks of the season.

The survival estimates derived from this study will improve our understanding of the extent to which hunter harvest affects overall mortality rates in gray and fox squirrels. Our results will be used by MNDNR’s Section of Wildlife to determine if adjustments to squirrel harvest regulations are warranted or if further research on squirrel populations and their habitat is needed.

ACKNOWLEDGMENTS

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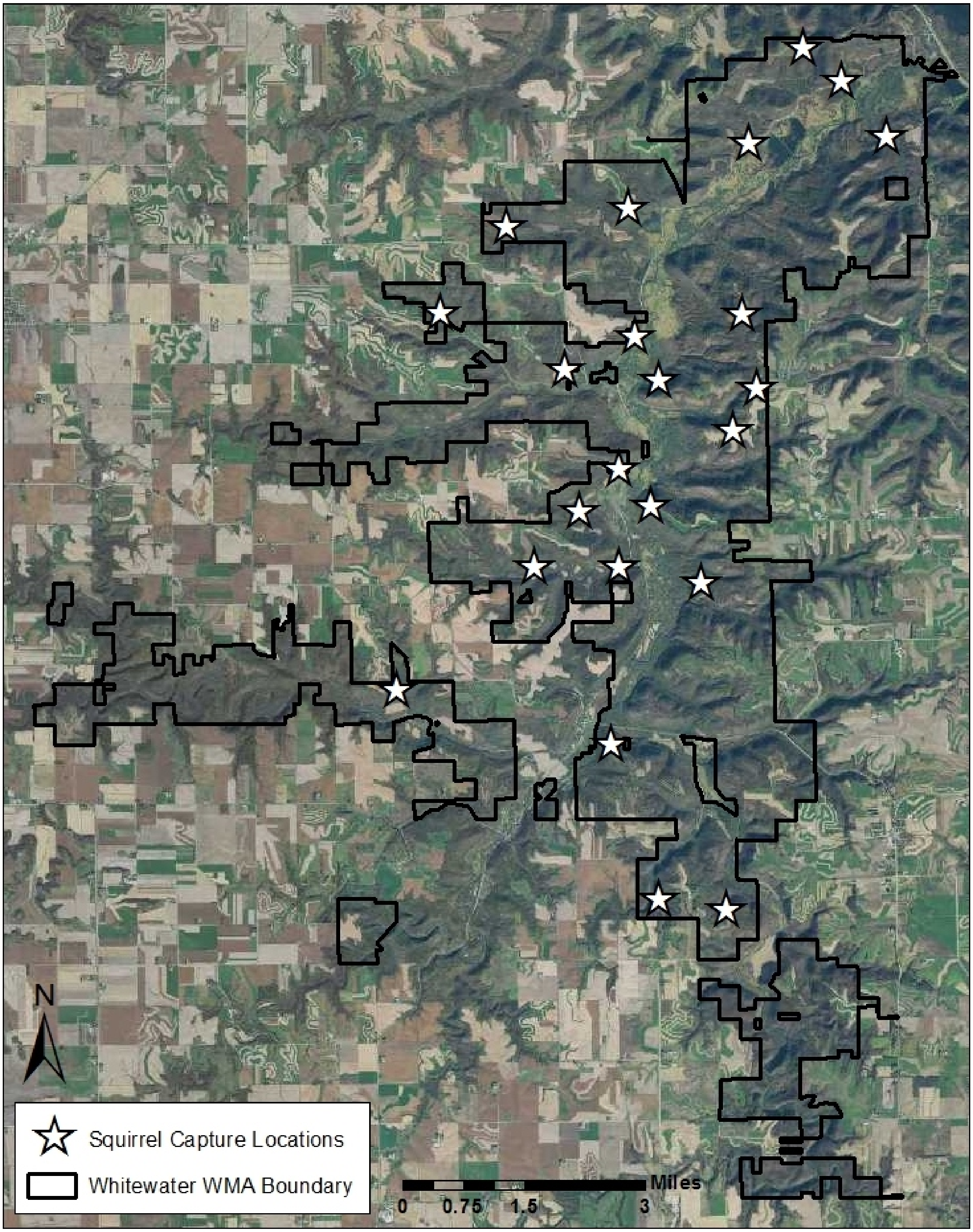

Figure 1. Capture locations of gray and fox squirrels (as indicated by white stars) during 11 July – 17 September 2015 and 24 May – 14 September 2016 in Whitewater Wildlife Management Area (WWMA), Minnesota (outlined in black).

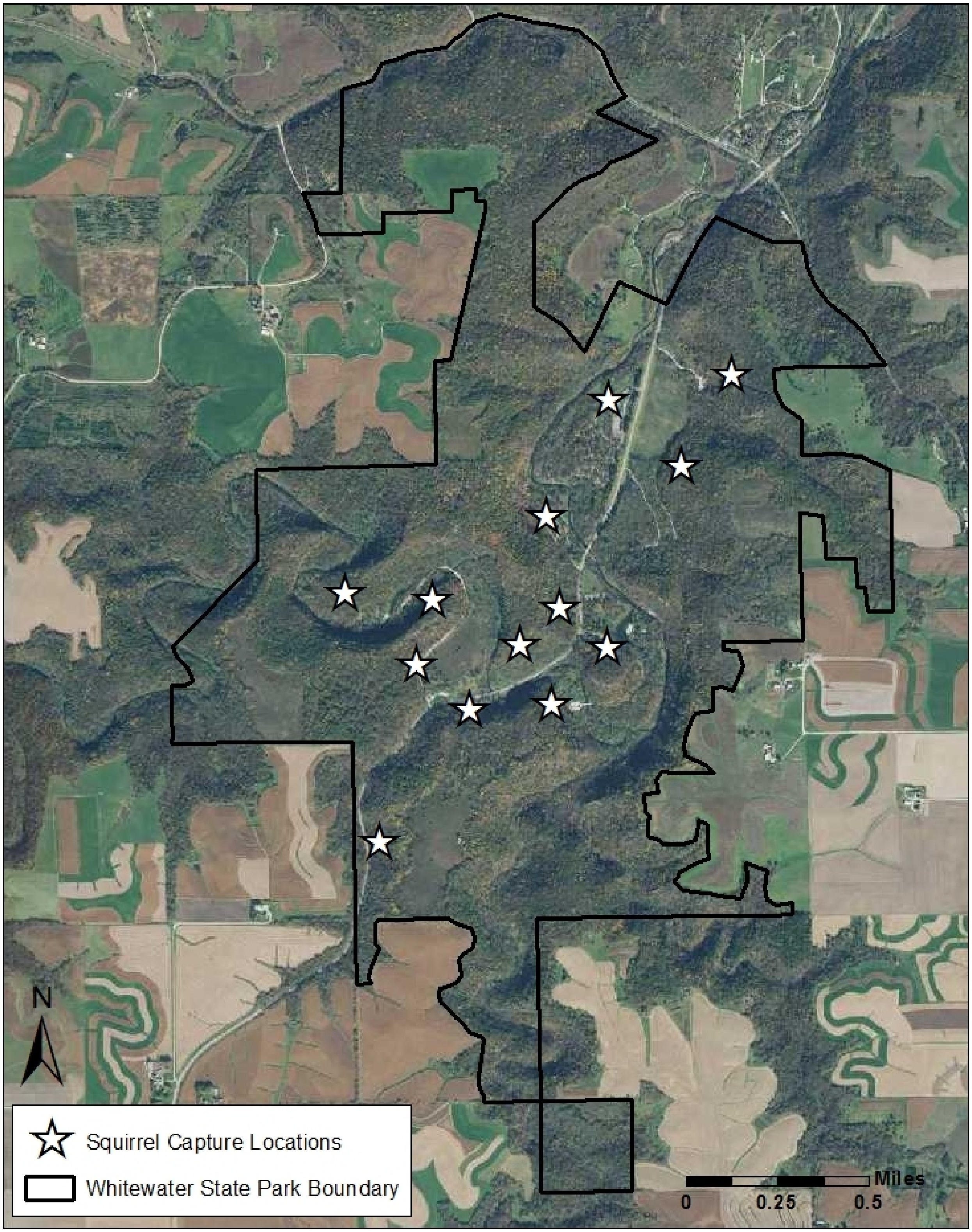

Figure 2. Capture locations of gray and fox squirrels (as indicated by white stars) during 6 August – 16 September 2015 and 2 May – 15 September 2016 in Whitewater State Park, Minnesota (outlined in black).

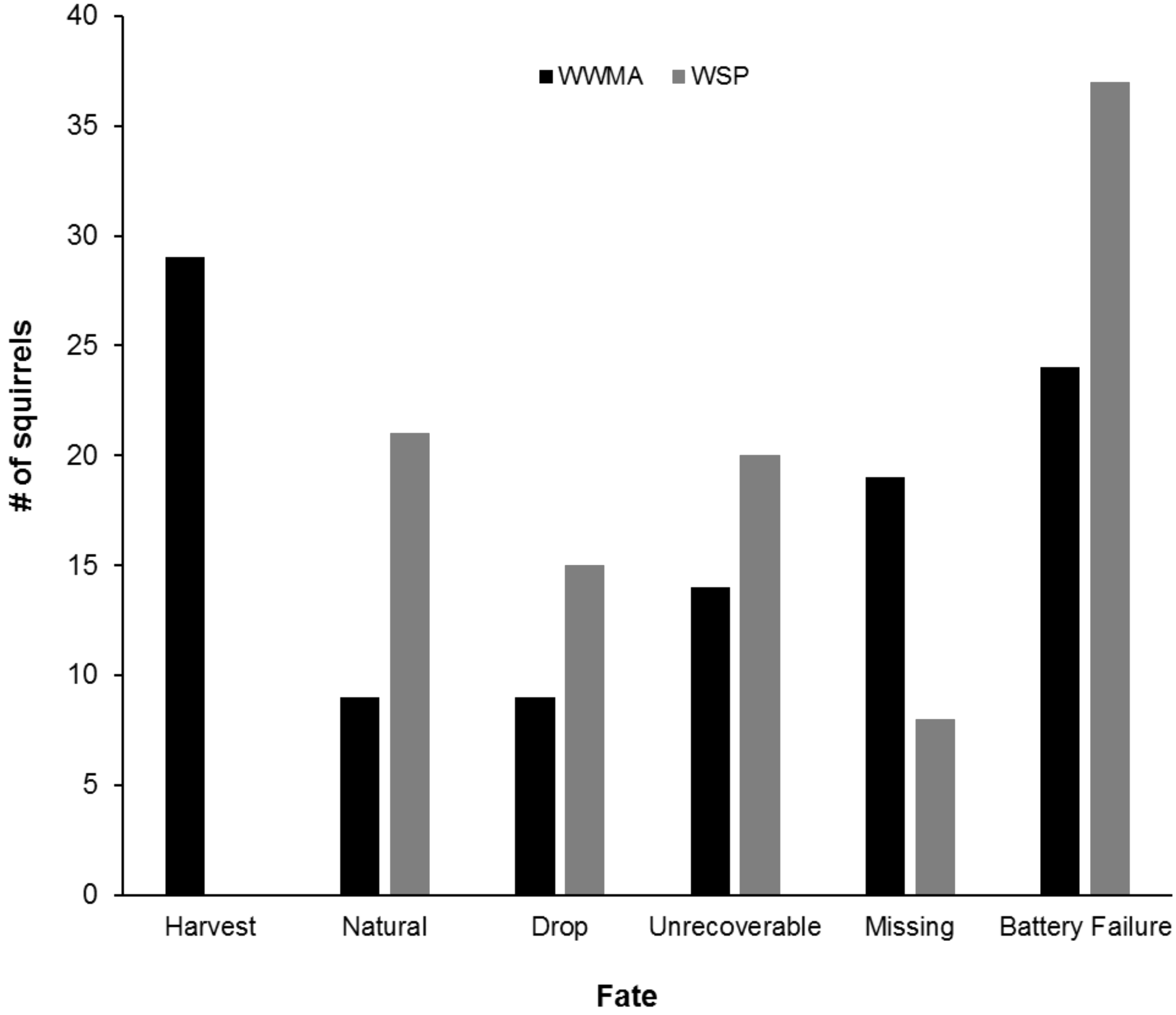

Figure 3. Fates of radiocollared gray and fox squirrels on Whitewater Wildlife Management Area, Minnesota (WWMA) and Whitewater State Park, Minnesota (WSP) 11 July 2015 – 25 May 2017.

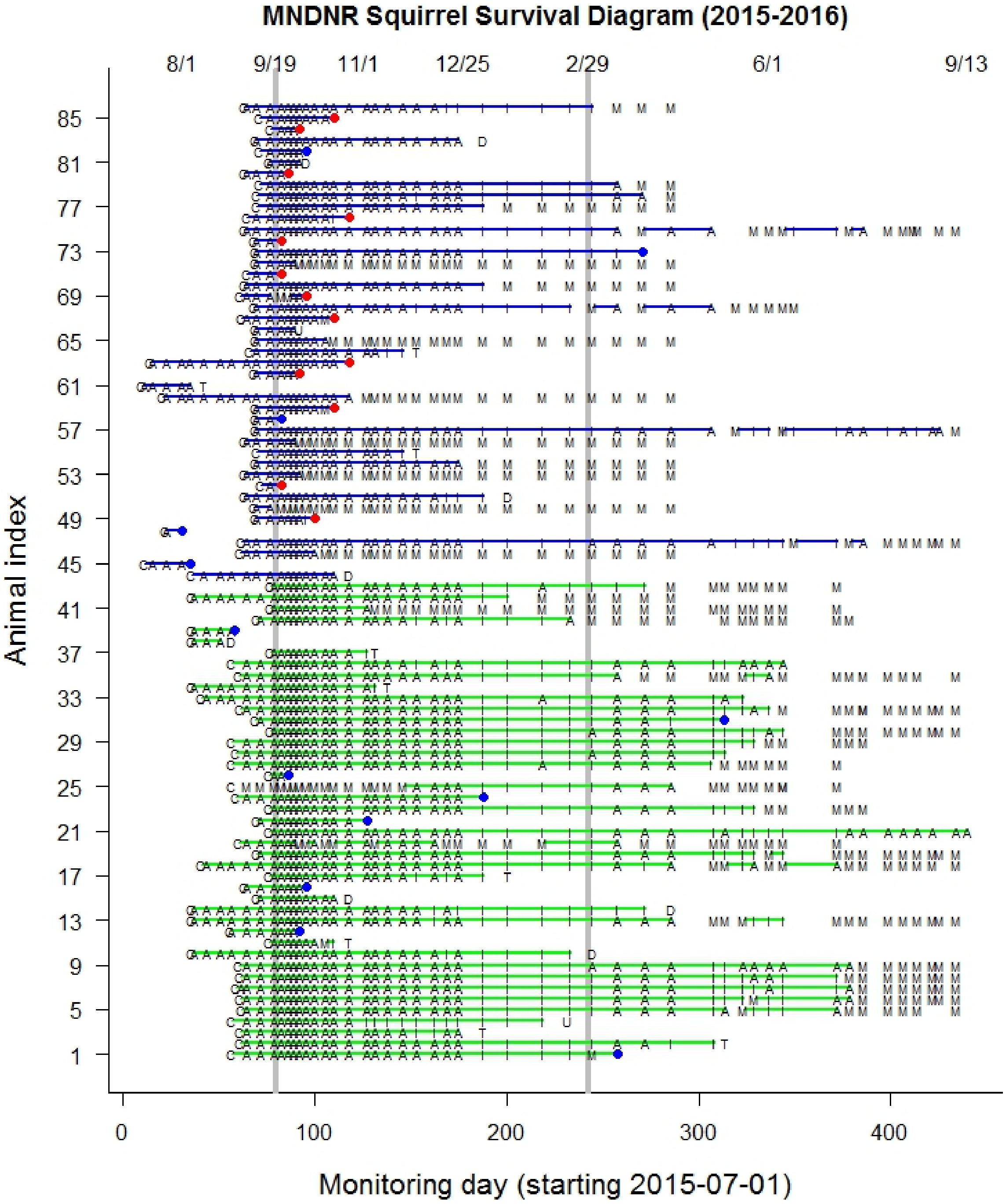

Figure 4. Life history diagram of radiocollared gray and fox squirrels captured in 2015 at Whitewater Wildlife Management Area, Minnesota (blue lines, n = 43) and Whitewater State Park, Minnesota (green lines, n = 43). Solid lines indicate continued survival across monitoring intervals. Fates of squirrels during the 2015-2016 hunting season are found within the gray vertical lines. Fate codes are as follows: red dot = harvest, blue dot = natural mortality, C = date of capture, A = active alive, I = inactive presumed alive, T = mortality in tree, D = dropped collar, U = undetermined collar loss, M = missing/signal not heard.

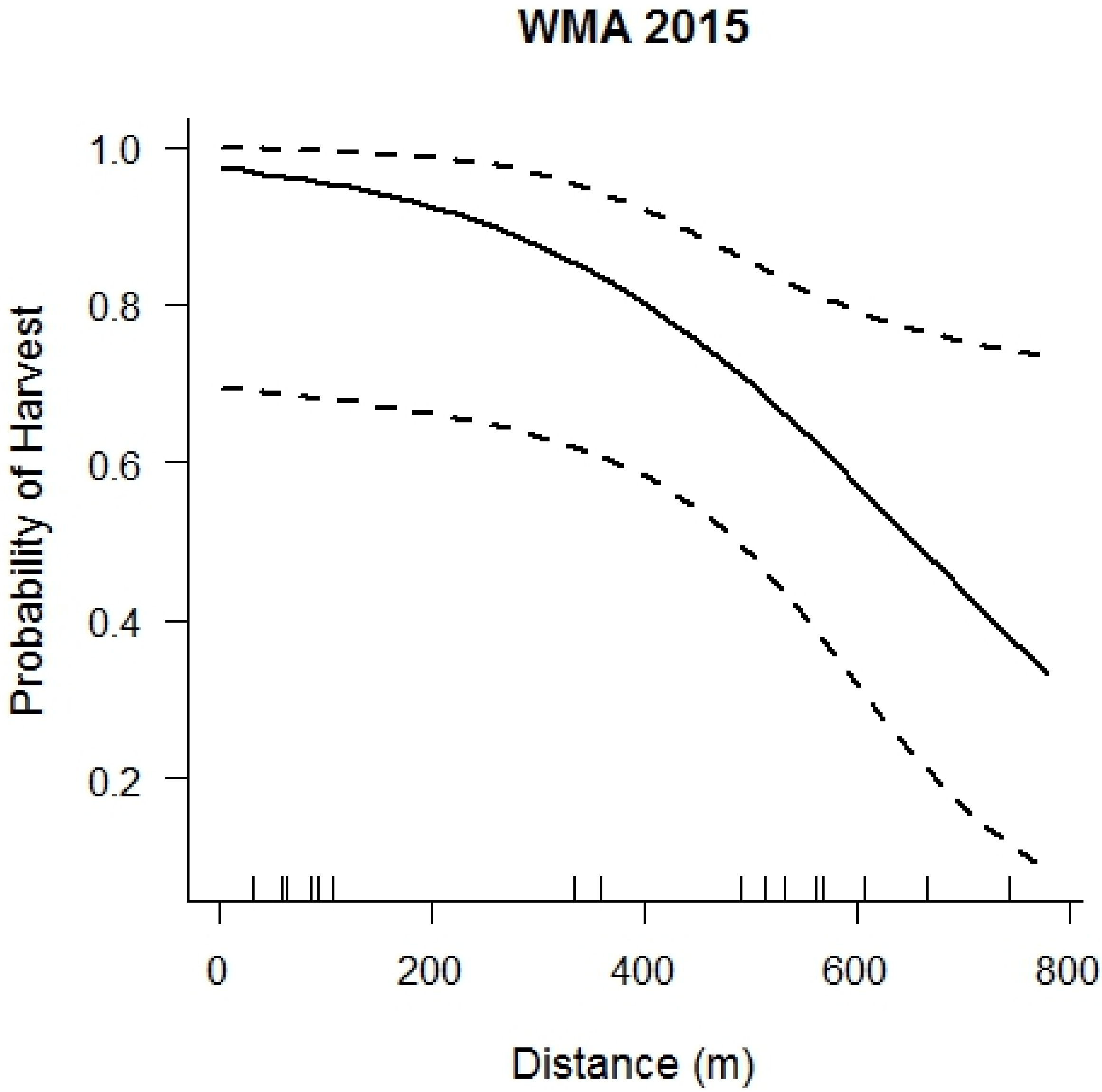

Figure 5. Probability of harvest of gray and fox squirrels (n = 44) in relation to distance from capture site to nearest hunter access point in Whitewater Wildlife Management Area (WWMA), Minnesota during 19 September 2015 – 29 February 2016.

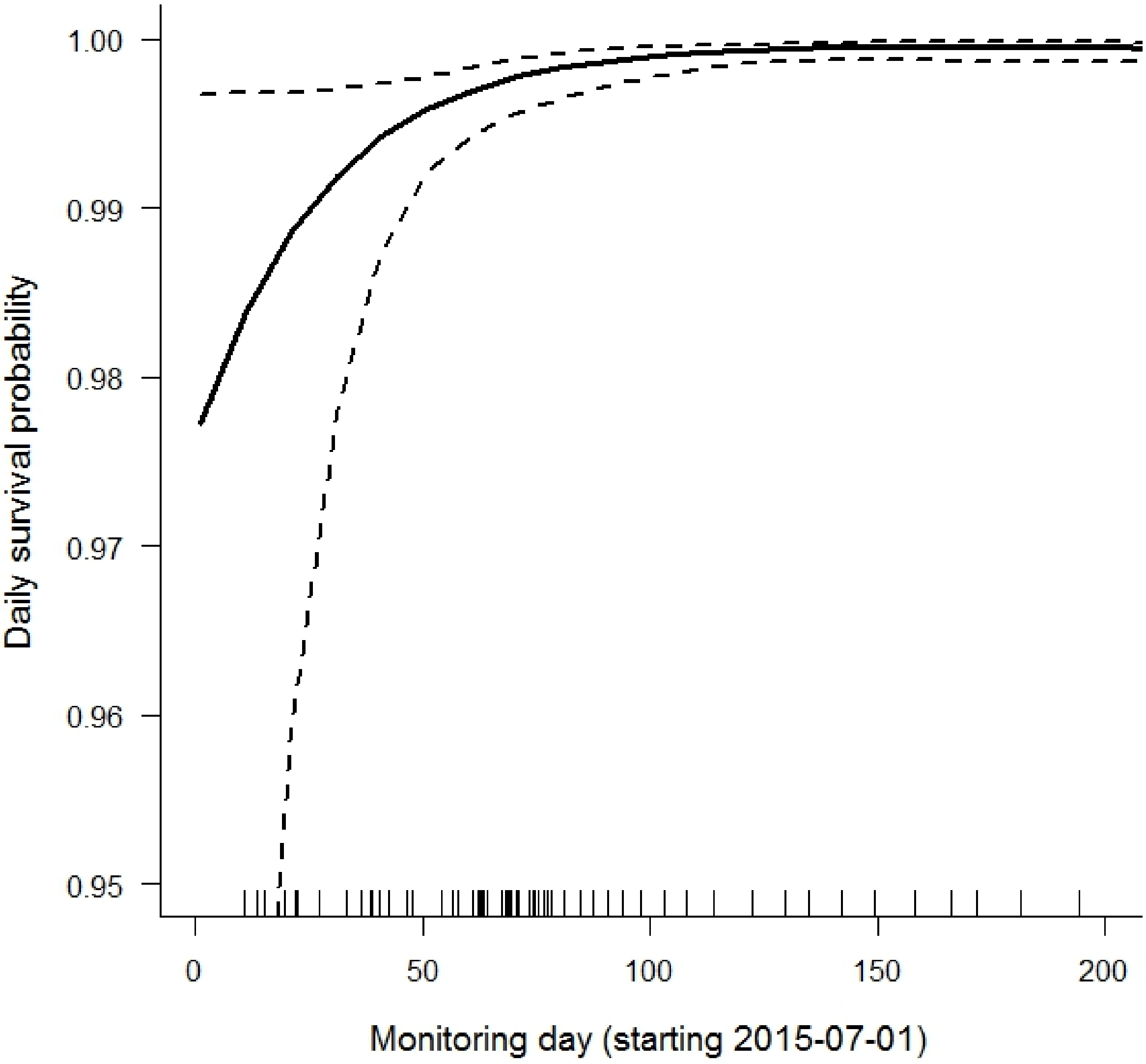

Figure 6. Daily survival probability (excluding the first 6 weeks of hunting season) of gray and fox squirrels in Whitewater Wildlife Management Area, Minnesota (n = 44) and Whitewater State Park, Minnesota (n = 43) during late summer, fall, winter, and early spring 2015-2016.

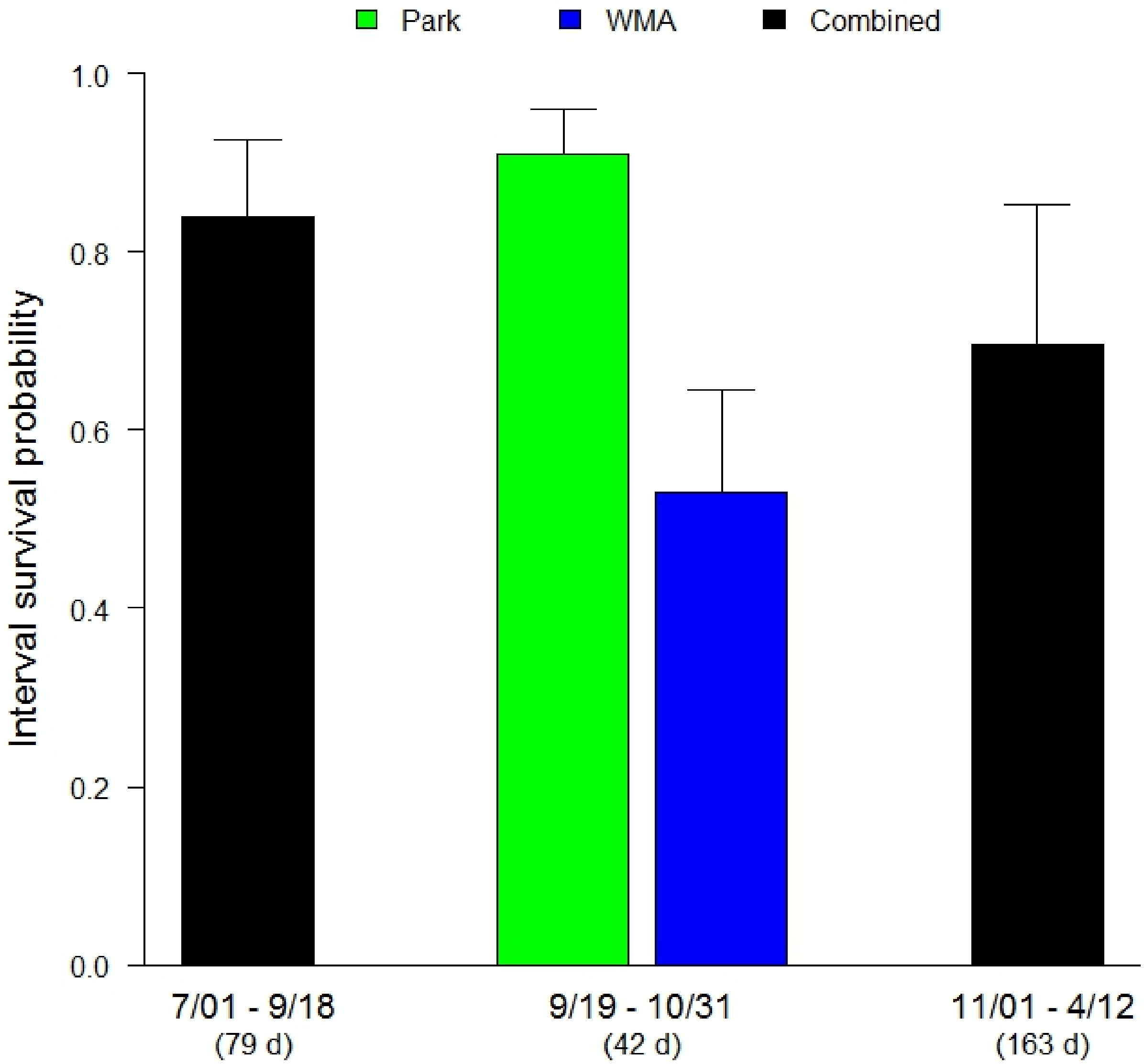

Figure 7. Estimates of combined squirrel survival (black, n = 87) and by site (green = Whitewater State Park, Minnesota (WSP, n = 43); blue = Whitewater Wildlife Management Area, Minnesota (WWMA, n = 44) during the pre-hunt time period (1 July 2015 – 18 September 2015), first 6 weeks of hunting season (19 September 2015 – 31 October 2015), and winter-spring time period (1 November 2015 – 12 April 2016).

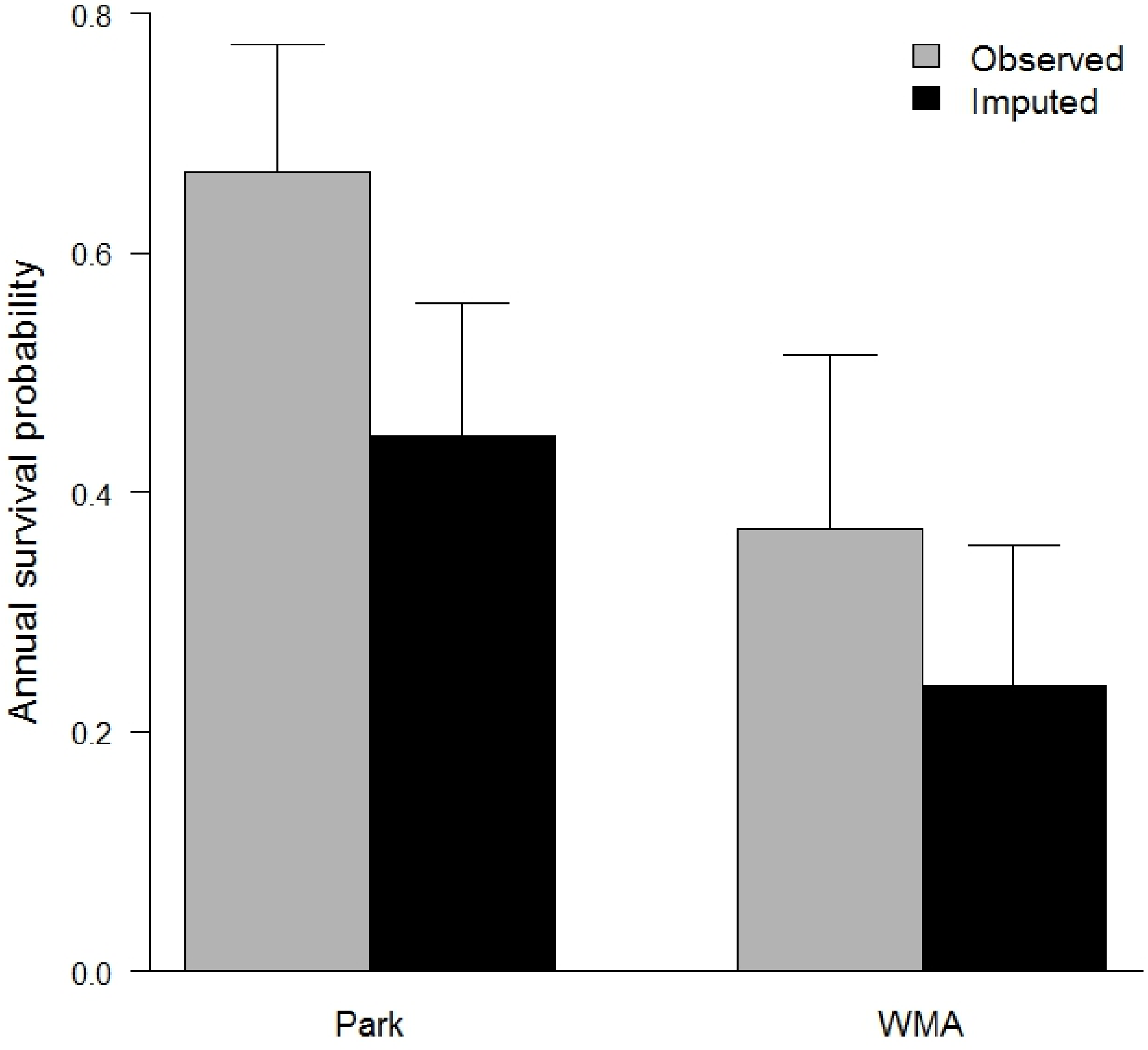

Figure 8. Imputation analysis indicating positive bias of observed annual survival probability due to non-independence of censoring and fate caused by high proportion of gray and fox squirrels with unknown fates. Sixteen of 43 and 22 of 44 squirrels had unknown fates on Whitewater State Park (WSP), Minnesota and Whitewater Wildlife Management Area (WWMA), Minnesota, respectively, during 2015-2016.

Human Dimensions Summaries

MINNESOTA HUNTER OPINIONS ABOUT DEER POPULATIONS AND MANAGEMENT

Leslie E. McInenly, Louis Cornicelli, and Eric Walberg

SUMMARY OF FINDINGS

The Minnesota Department of Natural Resources (MNDNR) Section of Wildlife conducted a survey of firearm white-tailed deer (*Odocoileus virginianus*) hunters to assess hunter preferences for deer population management, harvest regulations, and agency decision making. Results indicate hunter support for increased deer numbers, relative to 2014-2016, in many areas; however, factors identified by hunters as most important to consider in population management provide mixed direction for MNDNR. Although reported satisfaction with deer numbers and quality was generally low, satisfaction with the general hunting experience was relatively high; potentially influenced by non-consumptive motivations. Responses to questions regarding regulatory changes suggest that hunters, for the most part, prefer current regulations over commonly suggested alternatives and that hunting traditions have an important influence on regulatory preferences. Notably, however, establishment of a statewide youth season received relatively high levels of support statewide, suggesting further MNDNR consideration is warranted. Finally, results related to public participation in deer management suggest that opportunities to enhance relationships between staff and hunters should be explored. Given reported preferences for direct, rather than representative, means to provide input, MNDNR could revisit current public engagement methods to enhance support for management.

INTRODUCTION

MNDNR periodically conducts stakeholder surveys to collect information about public desires and opinions regarding specific natural resource management issues. Survey recipients are selected randomly and provide a statistically representative sample of stakeholder opinions. Over the past decade, MNDNR has conducted over a dozen deer hunter surveys to evaluate regulatory preferences and hunter satisfaction (Minnesota DNR 2016).

The 2015-2017 Minnesota deer hunting survey was conducted to assess hunters' season participation and activities, deer population perceptions and preferences, satisfaction, attitudes about deer management, regulatory preferences, relationship with MNDNR, and involvement in agency decision-making. During this time, MNDNR was coordinating a public process to revisit deer population goals for most of the deer permit areas (DPAs) in the state and public attention to deer management was high. Survey timing after the 2014 and 2015 seasons was coincident with the 2 lowest annual harvests in over a decade, a management response to population declines following 2 consecutive years (2013 and 2014) of moderate-to-severe winter conditions.

OBJECTIVES

The purpose of this study was to gather information at levels that adequately represent regional stakeholder attitudes (e.g., northeastern Minnesota). Specific survey objectives were to:

1) Continue to assess hunter perspectives on regional deer population trends and management,
2) Evaluate support for potential regulatory changes commonly raised by stakeholders, as well as the influence of deer population management decisions on regulatory preferences, and,
3) Better understand stakeholder relationships with MNDNR and preferences for communication/input in agency decisions to improve engagement processes and hunter satisfaction.

METHODS

Sampling

The 2015-2017 deer management study was divided into 5 strata covering all but the southeastern and southwestern portions of the state (Figure 1). Deer hunter attitude surveys were previously conducted in southeastern (Pradhananga et al. 2013) and southwestern (D'Angelo & Grund 2014) Minnesota. For this study, surveys were sent to 25,319 hunters in 5 different regions between winter 2015 and spring 2017, reflecting hunters' experiences and opinions after the 2014, 2015, or 2016 deer seasons. Because this survey was coincident with the deer population goal setting process in parts of the state, survey blocks H1, H3 and H4 were further stratified by sub-regions; the goal setting process in H2 and H5 was already complete. The target response size for each sub-region was 900; in former goal setting blocks, the target response size was 1,200. For all surveys, our error rate at the survey block level was approximately +/-3%.

For each survey block, random samples were drawn from the MNDNR electronic licensing system (ELS), selecting for adult hunters that declared intent to hunt a deer permit area (DPA) within that region during the most recent deer season[1]. Within each survey block, hunters were randomly assigned to 10 subsample groups. Each subsample group received 1 of 10 survey versions; all surveys were identical except for the order and set of regulatory choice options which were unique to each of the 10 survey versions. This design provided the ability to conduct a discrete choice experiment within each of the survey blocks (Louviere, Hensher & Swait 2000).

Data Collection

Surveys were presented online or as a 12-page paper booklet, including a cover page with photo. Online and paper surveys presented the same series of questions, tailored to the survey block of interest. Each survey contained 2 sections; a section focused on deer population observations and preferences and a section focused more broadly on hunting regulations, involvement with hunting, hunter satisfaction, hunter relationships with MNDNR, preferences related to MNDNR management and decision-making, and hunter demographics.

Data were collected using a web-first, mixed mode design that included a combination of online and mail surveys following the process outlined by Dillman and others (Dillman, Smyth, & Christian, 2014). The first 2 waves of letters requested survey completion online through the

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[1] At the time of license purchase, hunters 'declare' an area they intend to hunt. However, they are not legally required to stay in that area and although there is high site fidelity, some movement across the state occurs.

internet survey platform (Qualtrics, Provo, UT); each online survey code was unique and could be used only once. The third and fourth waves included a cover letter, a self-administered mail back survey booklet, and a business reply envelope. Because the fourth wave only increased the overall response rate by a small percent for surveys H1 – H4 (range = 8% - 9%), we opted to employ a 3-wave survey (i.e., 2 letters requesting online survey response followed by 1 mail-back paper survey booklet) for the H5 study area.

Contact letters were sent approximately 2 weeks apart; potential survey respondents were contacted up to 4 times between February and May of 2015 (H1 and H2), November 2015 and April 2016 (H3 and H4), or January and February 2017 (H5). Personalized cover letters explained the purpose of the study and made an appeal for respondents to complete the survey online; however, for survey recipients that do not have internet access, letters indicated that a paper survey would be mailed at a later date. Data were collected through July 2015 for the H1 and H2 surveys; through June 2016 for the H3 and H4 surveys; and through April 2017 for the H5 survey.

Discrete Choice Experiment

The survey also included a discrete choice experiment (DCE) designed to help MNDNR better understand individuals' preferences for different potential combinations of deer seasons and regulations in Minnesota. Discrete choice surveys present hypothetical scenarios and force respondents to choose an alternative among a suite of options (Adamowicz et al. 1994; Oh et al. 2005). By using an experimental design, scenarios selected by respondents can be used to identify the relative importance, or influence, of each attribute on regulatory and season combinations. In addition, by analyzing individuals' preferences for different levels of each attribute, we can estimate the utility, or relative desirability, of each level among respondents. The experiment in this survey focused on a combination of (1) management strategies that are often suggested by hunting stakeholders and (2) management designations that reflect both hunter opportunity and management toward a specific population goal. Survey respondents were presented with 8 deer season choice scenarios and asked to choose one option. Each scenario included 2 season structure choices plus a "none" (i.e., I would not hunt deer in Minnesota with these options).

Data Entry and Analysis

Online survey data were downloaded as .csv files using Qualtrics software (Qualtrics, 2015), converted to Excel 2013 spreadsheets, and provided the basic data entry template for hard-copy mail surveys. Data from mail surveys were manually entered in Excel 2013. A subsample of paper surveys (50 per survey) were double-entered to assess data entry error rates. Data entry error rates for each survey area ranged from 0.39% to 1.44%.

Basic descriptive summaries and statistical analyses were conducted using the Statistical Program for the Social Sciences (SPSS Statistics for Windows, version 24). Responses across survey blocks were compared using chi-squared tests and one-way analysis of variance (ANOVA). Independent samples t-tests or Mann-Whitney U tests were used to test differences in responses between groups. We measured effect size for chi-squared tests, ANOVA, and independent samples t-tests using Cramer's V, eta, and Cohen's *d*, respectively. Commonly accepted values (Cohen 1988, Vaske 2008) were used to interpret effect sizes as small, medium, and large (Cramer's $V \geq 0.1, 0.3, 0.5$; eta $\geq 0.1, 0.24, 0.37$; $d \geq 0.2, 0.5, 0.8$). The DCE portion of the survey was analyzed using Lighthouse Studio and hierarchical Bayes analysis.

State-level data were analyzed for all respondents, weighted by DPA to account for the proportion of hunters within the H1-H5 that purchased a 2014 license[2]. Region-level analyses were conducted by comparing responses across surveys and responses were similarly weighted by DPA to reflect the hunting population.

RESULTS

Overall, there were 973 undeliverable surveys; 10,894 completed hunter surveys were returned, yielding a 45% adjusted response rate (Table 1). Age and gender of non-responding survey recipients, from the MNDNR ELS, was compared with that of survey respondents to assess potential nonresponse bias. Median age of respondents was greater than that of non-respondents (52 versus 41) and Mann-Whitney U tests between these groups in each survey area indicate a substantial age difference (U = 1073047.5 – 4874450.0, Z = -14.388 – -20.450, $p < 0.001$, r = -0.238 – -0.265). No gender differences were detected.

Differences in attitudes and demographics between early respondents (mailing waves 1-3) and late respondents (mailing wave 4) were also explored to assess potential nonresponse bias. In general, no practical significance (effect size) was evident for most attitude responses. However, smaller proportions of late respondents in east central (H2) and northeastern (H3) Minnesota indicated preferences for population increases than did early respondents (V = 0.112 and 0.129, respectively). Median age of wave 4 respondents did not differ from earlier survey respondents.

Respondent Experience, Background, and Participation in Deer Hunting

On average, survey respondents were about 50 years old and nearly 90% of respondents were male. Most respondents (>60%) were not members of a hunting or conservation organization; reported membership was highest for local sporting clubs (14%) with smaller proportions of hunters indicating affiliation with organized deer hunting groups.

Respondents have hunted deer in Minnesota an average of 29 years overall and 20 years in the DPA they hunted most often. Almost all respondents (>98%) hunted during the previous deer season; less than 1% indicated they hadn’t hunted during the three previous years. Overall, 98% of hunters in all survey areas hunted during the firearm season; far fewer hunters participated in the archery (17%) or muzzleloader (13%) seasons. Of the estimated days spent scouting and hunting, only days spent afield during the firearm season substantially differed across survey areas, likely a result of the 16-day firearm season in the 100-series zone (northeastern, north central, and east central Minnesota; Table 2). As expected, fidelity to deer permit area was high; most respondents (>90%) reported they hunt the same area every year. The percentage of time spent hunting private vs. public land varied considerably by public land availability (Table 3). Overall, more than half of hunters did at least some of their hunting on private land.

Respondents were asked to indicate agreement, on a scale of 1 (“strongly disagree”) to 5 (“strongly agree”), with statements regarding their involvement (Kyle et al. 2007) with deer hunting in Minnesota (Table 4). Hunters indicated greatest agreement with items related to social relationships (e.g., opportunity to be with friends) and pleasure derived from the activity (e.g., one of the most enjoyable things I do). Notably, items associated with external perceptions (e.g., you can tell a lot about a person when you see them hunting) had some of the lowest levels of agreement. Similarly, respondents were also asked to rate, on a scale of 1 (“not at all”) to 5 (“extremely”), the importance of experiences to their deer hunting satisfaction during the

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[2] In Minnesota, hunters are required to designate the DPA they are most likely to hunt within during the hunting season; this information is used to estimate hunting pressure and can be assumed to reflect distribution of the hunting population.

previous deer season (Table 5). Factors respondents reported as most important to deer hunting satisfaction were also primarily experiential and social, including enjoying nature and the outdoors, hunting with family, enjoying a preferred pastime, being with hunting companions, and hunting with friends. Items associated with harvest success, and particularly buck harvest success, were rated among the least important.

Hunting techniques, personal harvest restrictions, and hunting approaches differed slightly across the areas. Most respondents reported using an elevated stand for hunting with smaller percentages of respondents indicating use of a ground stand, stalking, or participation in deer drives (Figure 2). Although a majority of hunters reported that they focus at least a portion of the firearm season on harvesting a large buck (44%) or any antlered buck (17%), most (83%) indicated they would shoot an antlerless deer if given the opportunity.

Population Trends and Perceptions about Deer Populations

A majority of hunters (67%) indicated there were fewer deer in the DPA they hunt most often than 5 years ago. Substantial differences in perceptions were observed among survey areas; in northeastern Minnesota, 82% of respondents indicated deer populations had declined whereas only 52% reported a decline in south central Minnesota. Statewide, 62% of respondents believed the population was too low. Again, differences were observed across all 5 regions. Respondents in northeastern Minnesota were most likely to indicate that populations were too low (80%) whereas nearly half of the respondents in south central and north central Minnesota reported that they felt the deer population had not changed (44% and 44% respectively) or was too high (5% and 4% respectively).

More than two-thirds of respondents wanted to see an increase in deer densities at some level (Figure 3). Across areas, preferences for future deer population management also varied depending on the type of land hunted, with greater proportions of hunters who primarily hunt public land supporting deer population increases (>80%) than those who primarily hunt private land (65% - 81%, depending on the type of land hunted).

Population Management Considerations

To better understand the factors hunters believe are most important to consider when setting deer population goals, MNDNR asked respondents to rate the importance of 12 items that would lead to management for either higher or lower deer populations (Figure 4). Respondents rated severe winter mortality, deer hunting heritage, and hunter satisfaction as the 3 most important items. Respondents were asked about their level of agreement with steps in setting deer population goals. On a scale of 1 (“strongly disagree”) to 5 (“strongly agree”), strongest agreement was with the importance of having decision makers explain the different options considered when deer population goals are set and why the final option was selected ($\bar{x}$ = 4.4), followed by opportunities for hunters and landowners to provide input ($\bar{x}$ = 4.3). With respect to input opportunities, more respondents felt it was important that hunters (93%) and landowners (91%) have opportunities to provide input regarding deer population goals than did those that felt it was important for Minnesotans, in general (67%), to have input opportunities. A majority of respondents also agreed that it is important to use the best available science (77%) and follow consistent decision-making procedures (73%). Less than half (48%) of hunters agreed that it is important to consider diverse interests in setting deer population goals.

Hunter Success and Satisfaction

Deer season regulations from 2014 to 2016 were conservative (i.e., designed to limit harvest and increase populations) in most deer permit areas statewide. As a result, harvest was biased toward legal bucks and antlerless permits were unavailable or limited in many areas. Roughly twice as many hunters reported they killed and tagged a legal buck (22%) as compared to those who reported killing an antlerless deer (12%). Overall, 27% to 44% of hunters reported harvesting a deer for themselves or another hunter, depending on the survey area.

Reported hunter satisfaction with deer numbers and quality was low. When asked about current (2014, 2015, or 2016) deer numbers in the DPA they hunt, most respondents in northeastern (76%), east central (63%), and northwestern (53%) Minnesota reported they were dissatisfied; just under half of hunters in south central (46%) and north central (49%) Minnesota reported dissatisfaction. Similar to reports of satisfaction with deer numbers in DPAs, a majority of hunters in northeastern (69%) and east central (60%) Minnesota indicated dissatisfaction with the number of deer seen while hunting; smaller proportions of hunters in northwestern (47%), north central (42%), and south central (42%) Minnesota indicated dissatisfaction with the number of deer seen. While the importance of seeing a lot of bucks for personal hunting satisfaction received only moderate ratings ($\bar{x}$ = 3.0; on a scale of 1 to 5 where 3 = "somewhat important"), most hunters reported dissatisfaction with the number of legal bucks (55%) and reported satisfaction was negatively correlated with the relative importance individual hunters placed on seeing bucks (r = -0.157, $p < .05$). Statewide, more hunters reported dissatisfaction (53%) than satisfaction (29%) with the quality of legal bucks. Reported satisfaction with the number of antlerless deer varied across the state (X^2=572.652, $p < 0.001$, V = 0.116), with hunters indicating greater satisfaction in south central (56%), north central (54%), and northwestern (49%) Minnesota than those in northeastern (30%) or east central (40%) Minnesota. Contrary to responses regarding deer numbers and quality, a majority of hunters (71%) indicated satisfaction with their general deer hunting experience during the recent season and this didn't substantially differ by area (X^2=287.957, $p < 0.001$, V = 0.083).

Overall satisfaction with the most recent deer hunt, a rating that likely included aspects of the deer population (numbers and quality) and the individual experience, varied across survey areas (Table 6). Higher overall satisfaction levels were reported in northwestern, south central, and north central Minnesota than in northeastern or east central Minnesota. Of the hunters reporting overall satisfaction with their deer season, satisfaction ratings were significantly higher for those who reported killing a deer than for those who did not, and this trend was evident within all survey areas (Figure 5).

Regulatory Preferences for Deer Management

Hunters were asked about their preferences regarding the scale of regulation implementation, season options, and various potential regulatory changes. Across all survey areas, a preference for more local (DPA; 44%) or regional (zone; 40%), rather than statewide, application was evident. Regardless of survey area, a majority of hunters supported the establishment of a statewide youth season in mid-October (Figure 6). In contrast, hunter preference regarding firearm season length varied across survey areas (X^2=878.222, $p < 0.001$, V = 0.291), with the majority of hunters in northwestern (60%) and south central (58%) Minnesota indicating a preference for a 9-day season and hunters in northeastern (75%), east central (66%), and north central (58%) Minnesota indicating a preference for a 16-day season, consistent with the prevalent season length offered in the respective survey areas. Across all areas, hunters indicated general support for a regulation that would increase the proportion of antlered bucks in the deer permit area they hunted most often. Consistent with previous surveys of Minnesota deer hunters, support for specific regulatory alternatives was lower than that expressed for an unspecified regulation (Figure 7).

Discrete Choice Experiment: Regulatory Combinations

Alternative hunting season packages presented in the DCE consisted of 5 attributes concerning different potential combinations of deer seasons and regulations in Minnesota: (1) cross-tagging of harvested deer, (2) whether or not antler point restrictions are in place, (3) timing of the firearm opener during or out of the rut, (4) deer population level, and (5) deer harvest limit. Across all survey areas, timing of the opener - either in early or in late November (in or out of the rut) - had the most influence on scenario choice followed closely by deer numbers in all but north central Minnesota (Table 7). The third most important attribute was cross-tagging in the majority of survey areas. Implementation of antler point restrictions had the least influence on scenario choice in northwestern and east central Minnesota whereas harvest limit was least important in northeastern, south central, and north central Minnesota.

Across all survey areas, a hunting opener in early November had the highest utility and was preferred over a late-November opener, legal cross-tagging for either sex was preferred over antlerless-only cross-tagging or no cross-tagging, no antler point restriction was preferred over an antler point restriction regulation, deer numbers higher than 2014-2016 levels were preferred over levels experienced during that time period or lower population levels, and, the preferred seasonal harvest (bag) limit was a 1-deer, either sex regulation (Hunter Choice) rather than a 1-deer limit with an antlerless lottery (Lottery) or a 2-deer limit (Managed) (Table 8).

Results of the DCE allow comparison of various regulatory packages via market simulation to estimate the proportion of respondents that would be expected to choose a particular scenario. For example, a simulation comparing a regulatory package representing existing regulatory structures with 2014-2016 population levels compared to similar packages with a higher deer population suggest that hunters would prefer scenarios with higher deer populations (68%) and, of those, most would prefer regulations requiring a 1-deer limit (40%). A second simulation was conducted to examine preferences related to 5 regulatory packages that could increase the proportion of antlered bucks in the population. In this simulation, the option describing existing regulations was preferred (31%). Notably, not hunting (13%) was predicted to be preferred over a package including a late-November hunt at 2014-2016 population levels (10%). If the same package were offered but with higher deer population levels, the existing regulations were predicted to receive an even greater share (33%) of hunter preference and a smaller percentage (9%) of hunters were predicted to indicate they would not hunt given the options provided.

Public Participation in Deer Management

With respect to statements about the approach MNDNR uses to set deer population goals (e.g., provides enough opportunities for input, provides adequate information), responses indicated neutral to slight disagreement across all areas. Statewide, the greatest proportion of respondents disagreed that MNDNR provides enough opportunities for hunters to provide input (40%) and do not trust MNDNR to establish appropriate deer goals (38%). Respondents were undecided – or not sure – about their level of agreement with most other statements related to agency decision making about deer population goals, including consideration of science (53%), consistency of decision-making processes (51%), input opportunities for Minnesotans (47%) and landowners (45%), explanation of decision alternatives (42%), and the adequacy of information provided by MNDNR (41%). Hunters were similarly undecided regarding their agreement with statements about the MNDNR approach to setting deer hunting rules, including opportunities for hunters to provide input (46%).

Overall, fewer respondents were neutral about their relationship and communication with MNDNR than they were with statements about agency decision-making procedures. Hunter agreement was neutral to negative regarding having adequate opportunities to communicate with MNDNR staff ($\bar{x}$ = 2.9; on a scale of 1 to 5 where 1 = “strongly disagree” and 5 = “strongly agree”). In contrast, hunter agreement was neutral to positive regarding knowing who to contact if they have questions or comments about deer management ($\bar{x}$ = 3.1).

Responses indicated greater ties to local conservation officers than with local wildlife managers or deer management staff (Figure 8). Across all areas, a majority of those familiar with their local area manager felt that they had adequate opportunities to communicate with MNDNR whereas only about a quarter of those who did not know their local area manager felt they had adequate opportunities to communicate with MNDNR (Table 9).

Hunters indicated a preference for direct rather than representative input to MNDNR, with preferences for online questionnaires (42%), written questionnaires (17%), and general public meetings (14%). The least preferred option to provide input was via advisory teams (3%), followed by informal communication (4%) and input through a representative organization (4%). Notably, providing no input (8%) rated higher than all but the top three options (Figure 9). Statewide, greater proportions of hunters over the age of 50 indicated a preference to provide input via general and issue-based public meetings (22%) and written questionnaires (19%) than younger hunters (16% and 12%, respectively), whereas a greater proportion of younger hunters reported a preference to provide input via online questionnaires (49% versus 33% for older hunters) (X^2=321.886, $p < 0.001$, $V = 0.178$).

Hunter agreement was neutral to negative with statements that MNDNR can be trusted to make decisions that are good for the resource ($\bar{x}$ = 3.0), will be open and honest in the things they do and say ($\bar{x}$ = 2.9), or will listen to the concerns of hunters ($\bar{x}$ = 2.9). In contrast, hunter agreement was neutral to positive with statements that MNDNR will make decisions about deer management in a way that is fair ($\bar{x}$ = 3.1) and that MNDNR has deer managers and biologists who are well trained for their jobs ($\bar{x}$ = 3.3). Age was weakly but negatively correlated (r = -0.052, $p < .001$) with trust that MNDNR will establish appropriate deer population goals. Members of organized deer groups (MDHA, QDMA, MBI, and MWA[3]) also reported significantly lower levels of trust than those who were not members of an organized deer group ($\bar{x}$ = 2.6 and 2.9, respectively; t = 9.004, p <0.001, d = 0.429).

DISCUSSION

Although differences were observed by region, the majority of hunters reported recent declines in deer populations, felt deer populations were too low, and desired management to increase deer densities in their area. Factors identified by hunters as most important to consider in setting population goals provide mixed direction for management because concerns about deer mortality would suggest management for lower populations whereas concerns about deer hunting heritage and hunter satisfaction might suggest management for higher populations. Most respondents also felt that hunter and landowner input, as well as the best available science, should be considered in setting deer population goals; however, less than half agreed it was important to consider diverse interests in setting goals. This finding is counter to the recommendation made by the Minnesota Office of the Legislative Auditor for MNDNR to enhance human dimension surveys in order to consider more diverse perspectives (Minnesota OLA 2016). Although the state manages wildlife for public benefit, broadly, continued tension relative to the weight given to various stakeholder perspectives should be anticipated.

Measures of hunter satisfaction can be difficult to interpret because a number of variables may influence a satisfaction rating (see also Cornicelli & McInenly 2016). Contributing factors include personal motivations and expectations (many of which are non-consumptive), the context of the experience, and harvest success. Notably, hunters in areas with the lowest estimated deer densities (D’Angelo & Giudice 2015) reported both the lowest (northeastern Minnesota) and

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[3] MDHA = Minnesota Deer Hunters Association, QDMA = Quality Deer Management Association, MBI = Minnesota Bowhunters Inc., MWA = Minnesota Whitetail Alliance (Facebook group)

highest (south central Minnesota) levels of satisfaction with deer numbers. Of note, larger proportions of hunters in each survey area reported satisfaction with the number of deer *seen while hunting* than reported satisfaction with deer numbers *in the DPA they hunt most often*, suggesting greater satisfaction with deer numbers observed at more local levels. Contrary to responses regarding deer numbers and quality, a majority of hunters indicated satisfaction with their general deer hunting experience during the recent season, reinforcing earlier results that suggest non-consumptive motivations can have a greater influence on satisfaction with the deer hunting experience than do consumptive motivations.

Results from this survey suggest that hunters, for the most part, prefer current regulations over commonly suggested alternatives (e.g., prohibition of cross-tagging) and that hunting traditions (e.g., early November firearm opener and current season length) have an important influence on regulatory preferences. Notably, however, this is the first time MNDNR included a question about support for a statewide youth season. Results of this survey suggest further MNDNR consideration regarding establishment of a statewide youth season is warranted.

Market simulation results, based on the DCE, suggest that bag limit preferences are somewhat insensitive to population levels, i.e., the preference for a higher population is not driven by a desire to harvest more than deer based on current statewide hunter preferences. Additionally, simulation results suggest that, statewide, commonly proposed DNR regulatory packages that could increase the proportion of antlered bucks in the population are currently less attractive than existing DNR regulations even at higher population levels. Future work exploring the influence of hunter heterogeneity on preferences could refine these findings.

Finally, results related to public participation in deer management suggest that opportunities to enhance relationships between staff and hunters should be explored. Although >90% of respondents indicated it was important to provide opportunities for hunter input in decision making, nearly half were unsure about MNDNR decision-making processes and opportunities for input. Most hunters also reported that they have not communicated with or did not know area wildlife managers or deer management staff. Given reported preferences for direct, rather than representative, means to provide input, MNDNR could revisit current public engagement methods to enhance support for management.

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Table 1. Overall sample size, returns, adjusted response rates, and survey timing for Minnesota deer hunter surveys, 2015 - 2017. Youth respondents (reported ages <18 years) removed from analysis.

Survey block	Region	*N*	Undeliverable	Returned	Response	Survey timing
H1	Northwestern MN	7,801	333	3,095	41.4%	Spring 2015
H2	East Central MN	3,616	138	1,553	44.7%	Spring 2015
H3	Northeastern MN	5,202	222	2,544	51.1%	Fall/Winter 2015-16
H4	South Central MN	5,201	152	2,313	45.8%	Fall/Winter 2015-16
H5	North Central MN	3,499	128	1,389	41.2%	Fall/Winter 2016-17
Total		25,319	973	10,894	44.8%	

Table 2. Average number of days spent scouting or hunting reported by Minnesota deer hunters, 2015-2017, by season.

		Area								
Days scouting	*n*	1 (NW)	2 (EC)	3 (NE)	4 (SC)	5 (NC)	TOTAL	*F*	*p*	η^2
Archery	666	14.1	12.1	10.8	15.1	10.0	11.3	2.526	0.040	0.015
Firearm	3,870	5.5	7.0	7.7	7.3	6.1	6.4	3.617	0.006	0.004
Muzzleloader	14	8.0	7.2	5.9	7.8	4.7	6.1	1.028	0.392	0.008
Days hunting										
Archery	1,763	16.8	18.4	14.6	16.8	13.2	16.1	5.726	.000	0.013
Firearm	9,629	4.9	6.1	7.5	4.6	5.7	5.7	336.512	.000	0.123
Muzzleloader	1,368	5.8	6.2	5.8	6.3	6.1	6.0	1.301	.268	0.004

Table 3. Type of land hunted during most recent deer hunting season, reported by Minnesota deer hunters, 2015-2017.

		Area						
Type of land hunted		1 (NW)	2 (EC)	3 (NE)	4 (SC)	5 (NC)	TOTAL	Significance
Private land that I own	None	36.0%	38.6%	40.8%	43.2%	40.7%	39.5%	χ2=157.957*** V = 0.077
	Some	10.0%	10.5%	18.0%	10.7%	15.2%	13.0%	
	Most	18.8%	15.7%	17.2%	16.2%	15.7%	16.8%	
	All	35.1%	35.2%	24.0%	29.8%	28.4%	30.7%	
Private land that I lease for hunting	None	92.0%	94.6%	89.5%	91.1%	92.2%	92.0%	χ2=26.472** V = 0.035
	Some	3.4%	2.7%	4.1%	3.8%	2.8%	3.4%	
	Most	2.2%	1.6%	2.8%	2.5%	2.6%	2.3%	
	All	2.4%	1.1%	3.6%	2.7%	2.3%	2.3%	
Private land that I do not own or lease	None	32.1%	35.9%	49.4%	20.2%	41.8%	37.0%	χ2=541.189*** V = 0.143
	Some	18.4%	15.5%	21.5%	17.0%	19.1%	18.4%	
	Most	18.8%	14.7%	12.8%	22.1%	13.6%	16.0%	
	All	30.8%	33.9%	16.4%	40.7%	25.4%	28.6%	
Public land	None	59.0%	57.3%	22.8%	54.0%	28.5%	42.9%	χ2=1398.245*** V = 0.235
	Some	29.2%	22.7%	27.5%	31.7%	25.3%	26.7%	
	Most	6.8%	9.0%	22.2%	7.9%	18.3%	13.7%	
	All	5.0%	11.0%	27.4%	6.4%	27.9%	16.6%	

** $p < 0.01$, *** $p < 0.001$

Table 4. Involvement in deer hunting in Minnesota Level of agreement, reported by Minnesota deer hunters, 2015-2017.

Statement	*n*	Mean[1]
Deer hunting provides me with the opportunity to be with friends	10415	4.3
Deer hunting is one of the most enjoyable things I do	10441	4.3
I enjoy discussing deer hunting with my friends	10395	4.3
I contribute to deer management through hunting	10405	4.2
Deer hunting is very important to me	10413	4.1
To change my preference from deer hunting to another activity would require major thinking	10419	4.0
Deer hunting is one of the most satisfying thing I do	10421	3.9
I can really be by myself	10413	3.8
I identify with people and images associated with deer hunting	10409	3.8
When I am deer hunting, others see me the way I want them to see me	10411	3.8
Most of my friends are in some way connected with deer hunting	10425	3.7
Participating in deer hunting says a lot about who I am	10405	3.6
You can tell a lot about a person when you see them deer hunting	10392	3.5
When I am deer hunting, I don't have to be concerned about what other people think of me	10409	3.4
Deer hunting has a central role in my life	10392	3.4
A lot of my life is organized around deer hunting	10436	3.4

[1] *Mean is based on the scale: 1 = strongly disagree, 2 = disagree, 3 = neutral, 4= agree, 5 = strongly agree.*

Note: Means reflect weighted averages for a statewide response.

Table 5. Average importance rating of experiences to deer hunting satisfaction during the recent hunting season, reported by Minnesota deer hunters, 2015-2017.

Experience	*n*	Mean[1]
Enjoying nature and the outdoors	10308	4.5
Hunting with family	10307	4.2
Enjoying a preferred pastime	10300	4.1
Being with hunting companions	10353	3.9
Hunting with friends	10326	3.9
Seeing a lot of deer	10309	3.6
Becoming a better deer hunter	10340	3.5
Improving my knowledge	10309	3.4
Helping manage deer populations	10291	3.4
Developing skills and abilities	10341	3.3
Harvesting at least one deer	10287	3.2
Getting food for my family	10331	3.1
Proving my hunting skills and knowledge	10272	3.0
Challenges of harvesting a trophy	10296	3.0
Seeing a lot bucks	10298	3.0
Harvesting any deer for meat	10305	2.9
Influencing deer sex ratios or age structure	10265	2.9
Harvesting a large buck	10277	2.7
Harvesting any buck	10295	2.5
Selectively harvesting a large buck	10300	2.5
Getting a buck every year	10305	1.9

[1] *Mean is based on the scale: 1 = not at all important, 2 = slightly important, 3 = somewhat important, 4= very important, 5 = extremely important. Note: Means reflect weighted averages for a statewide response.*

Table 6. Overall satisfaction with most recent deer hunt, reported by Minnesota deer hunters, 2015-2017.

Area	*n*	Year	Very dissatisfied	Slightly dissatisfied	Neither	Slightly satisfied	Very satisfied	Mean[1]
1 (NW)	2919	2014	13.2%	23.3%	14.9%	28.4%	20.3%	3.2
2 (EC)	1455	2014	18.9%	28.0%	13.5%	26.6%	12.9%	2.9
3 (NE)	2416	2015	20.8%	28.1%	13.7%	25.3%	12.1%	2.8
4 (SC)	2222	2015	10.1%	21.8%	13.3%	33.4%	21.3%	3.3
5 (NC)	1322	2016	10.0%	19.6%	13.8%	32.7%	23.9%	3.4
TOTAL	10302		15.2%	24.5%	13.9%	28.8%	17.7%	3.1
					χ2=330.621*** V = 0.089			F=81.621*** η^2 = 0.031

*Mean is based on the scale: 1 = very dissatisfied, 2 = slightly dissatisfied, 3 = neither dissatisfied nor satisfied, 4 = slightly satisfied, 5 = very satisfied. *** p < 0.001*

Table 7. Relative importance of season choice attributes derived from hierarchical Bayes estimation of utilities of Minnesota deer hunters surveyed 2015-2017.

	Importances (SD)					
Season choice attribute	NW *n = 1,234*	EC *n = 958*	NE *n = 1,098*	SC *n = 1,597*	NC *n = 869*	Statewide *n = 2,757*
Cross-tagging	18.7 (9.4)	19.0 (9.7)	15.1 (8.4)	19.1 (11.1)	21.9 (10.6)	18.5 (9.8)
Antler Point Restrictions	15.7 (10.9)	15.1 (10.3)	15.8 (11.0)	18.6 (13.0)	15.6 (11.2)	15.9 (11.1)
Timing of opener	26.5 (14.9)	26.5 (14.5)	30.2 (15.4)	25.0 (15.5)	27.8 (15.1)	28.0 (15.6)
Deer numbers	22.0 (12.3)	23.2 (11.9)	25.0 (14.1)	22.1 (12.5)	21.0 (13.2)	22.0 (13.1)
Harvest limit	17.1 (10.9)	16.3 (9.9)	13.9 (9.0)	15.2 (9.4)	13.6 (9.4)	15.5 (10.3)

Table 8. Statewide results of the hierarchical Bayes model for regulatory choice for Minnesota deer hunters showing utilities of different levels of season attributes of Minnesota deer hunters surveyed 2015-2017.

Choice attribute - level	Average utilities	*SD*
Cross-tagging		
- Cross-tagging legal for antlerless only	8.4	21.1
- Cross-tagging illegal for both sexes	-40.8	39.8
- Cross-tagging legal for either sex	32.3	33.9
Antler Point Restrictions		
- *No antler point restrictions*	17.4	45.3
- Antler point restrictions	-17.4	45.3
Timing of opener		
- *Early November*	59.2	54.1
- Late November	-59.2	54.1
Deer numbers		
- Deer numbers lower than current levels	-55.7	40.2
- Deer numbers at current levels	11.3	14.1
- *Deer numbers higher than current levels*	44.5	39.1
Harvest limits		
- One deer limit, antlerless by permit only (lottery)	-14.0	35.9
- *One deer limit, either sex (hunter choice)*	23.8	26.3
- Two deer limit (managed)	-9.8	43.5
None	-99.9	219.3

Notes: n=2,757, attribute level with highest utility italicized

Table 9. Agreement with statement… I have adequate opportunities to communicate with MNDNR, based on reported familiarity with area wildlife manager, from Minnesota deer hunters surveyed 2015-2017.

Know area manager	n	Strongly disagree	Disagree	Not sure	Agree	Strongly agree	Significance	Effect size
				Area 1 (NW)				
No	2585	8.5%	30.3%	36.6%	22.9%	1.7%	χ2=170.144***	*V* = 0.242
Yes	317	2.5%	17.4%	22.1%	50.5%	7.6%		
				Area 2 (EC)				
No	1319	8.3%	26.2%	38.4%	25.6%	1.4%	χ2=73.278***	*V* = 0.225
Yes	124	8.9%	11.3%	19.4%	53.2%	7.3%		
				Area 3 (NE)				
No	2196	8.5%	27.6%	38.9%	24.2%	0.8%	χ2=203.866***	*V* = 0.291
Yes	208	3.4%	19.7%	16.3%	49.5%	11.1%		
				Area 4 (SC)				
No	1994	8.2%	25.7%	40.1%	24.4%	1.5%	χ2=176.833***	*V* = 0.284
Yes	198	3.0%	11.1%	19.2%	55.6%	11.1%		
				Area 5 (NC)				
No	1168	6.4%	24.7%	40.8%	25.9%	2.1%	χ2=90.438***	*V* = 0.264
Yes	132	3.8%	10.6%	20.5%	53.8%	11.4%		
				STATE				
No	9237	8.0%	27.1%	38.7%	24.7%	1.5%	χ2=638.559***	*V* = 0.250
Yes	973	4.3%	14.5%	19.8%	52.0%	9.4%		

*** *p< 0.001*

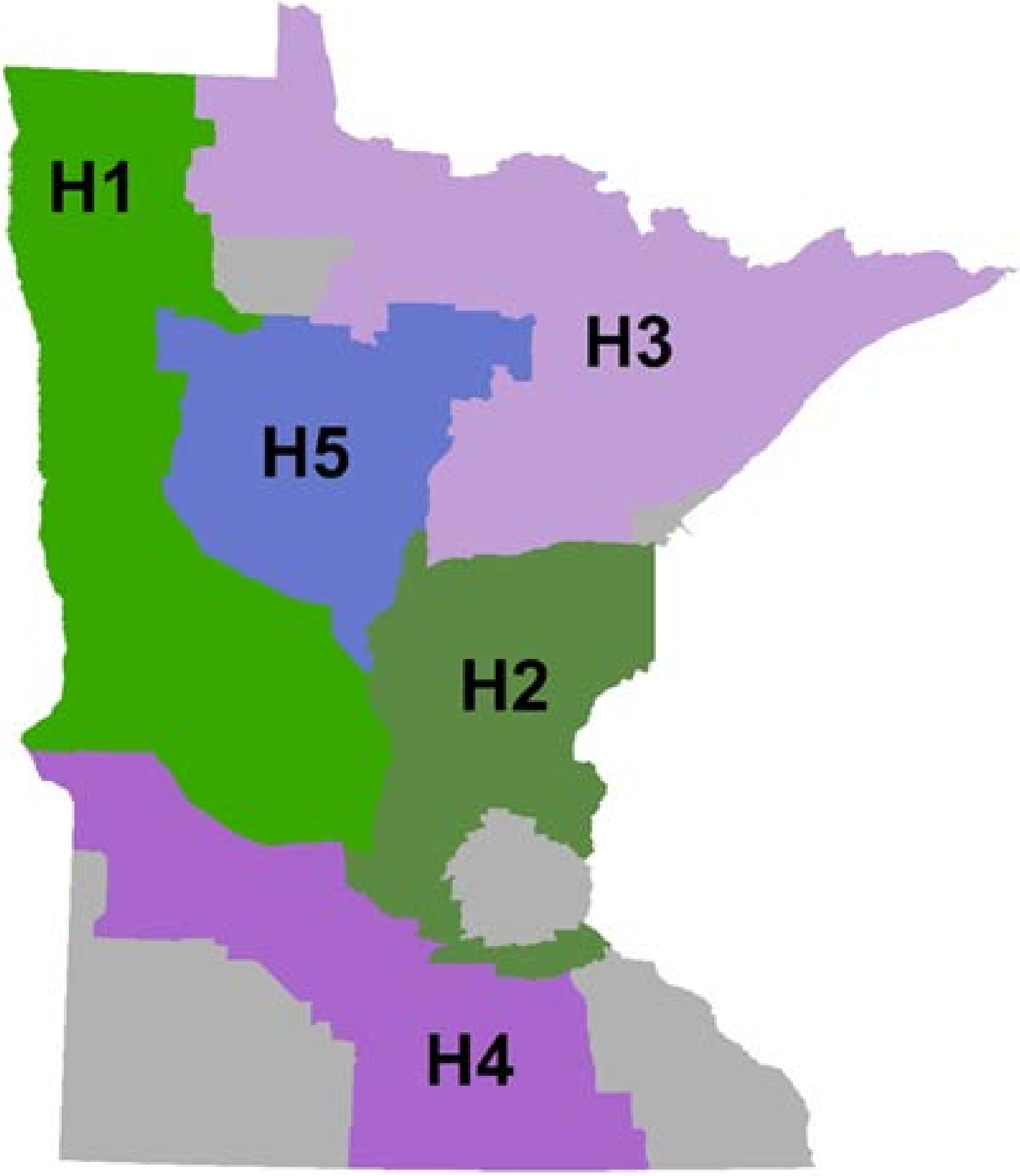


Figure 1. Hunting regions (survey blocks) surveyed by Minnesota Department of Natural Resources between 2015 and 2017 to evaluate hunter preferences for managing white-tailed deer in the region.

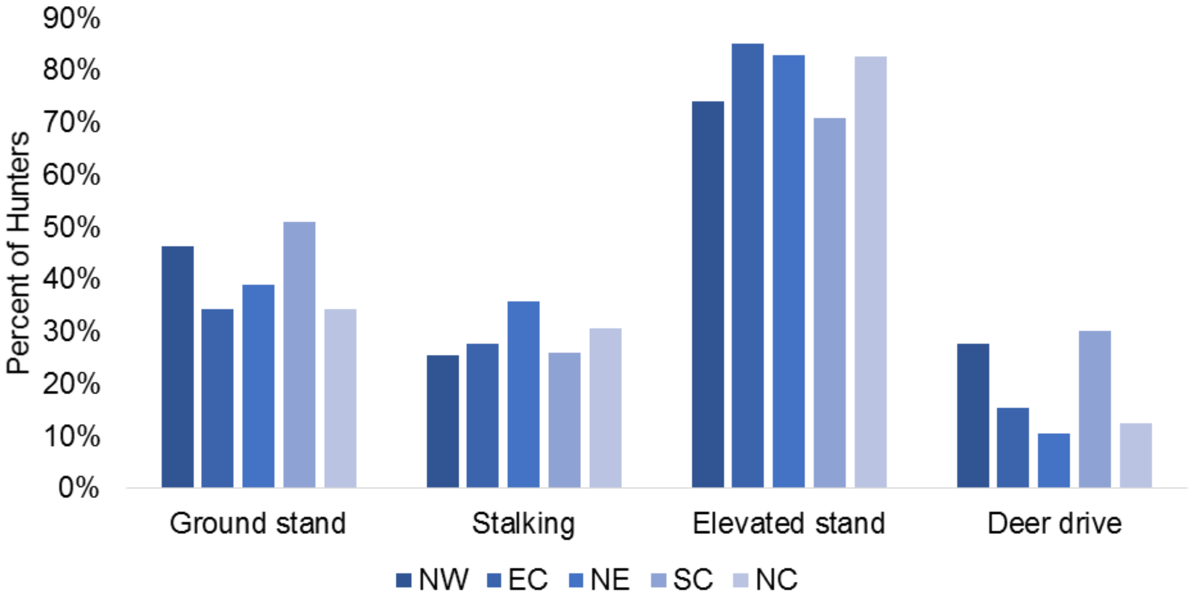


Figure 2. Hunting techniques used during most recent year hunted, by Minnesota deer hunter survey area, 2015-2017.

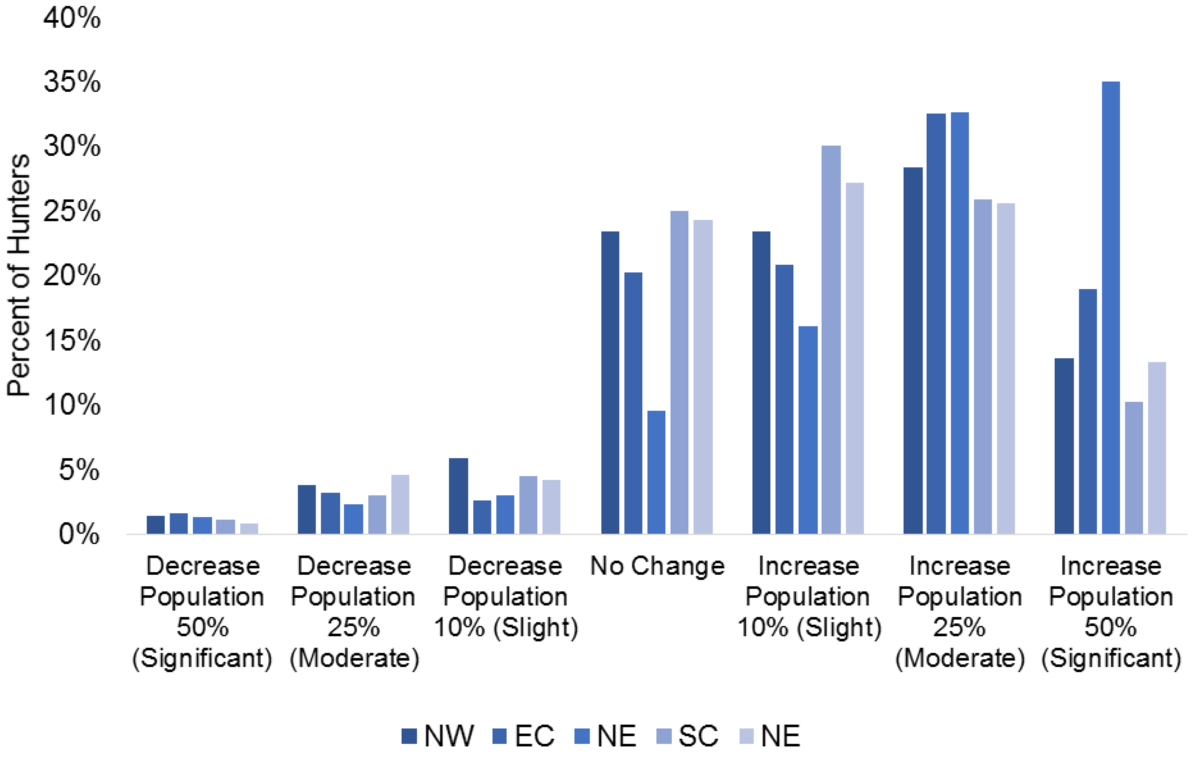


Figure 3. Future Minnesota deer management preferences, relative to 2014, 2015, or 2016 levels, by area.

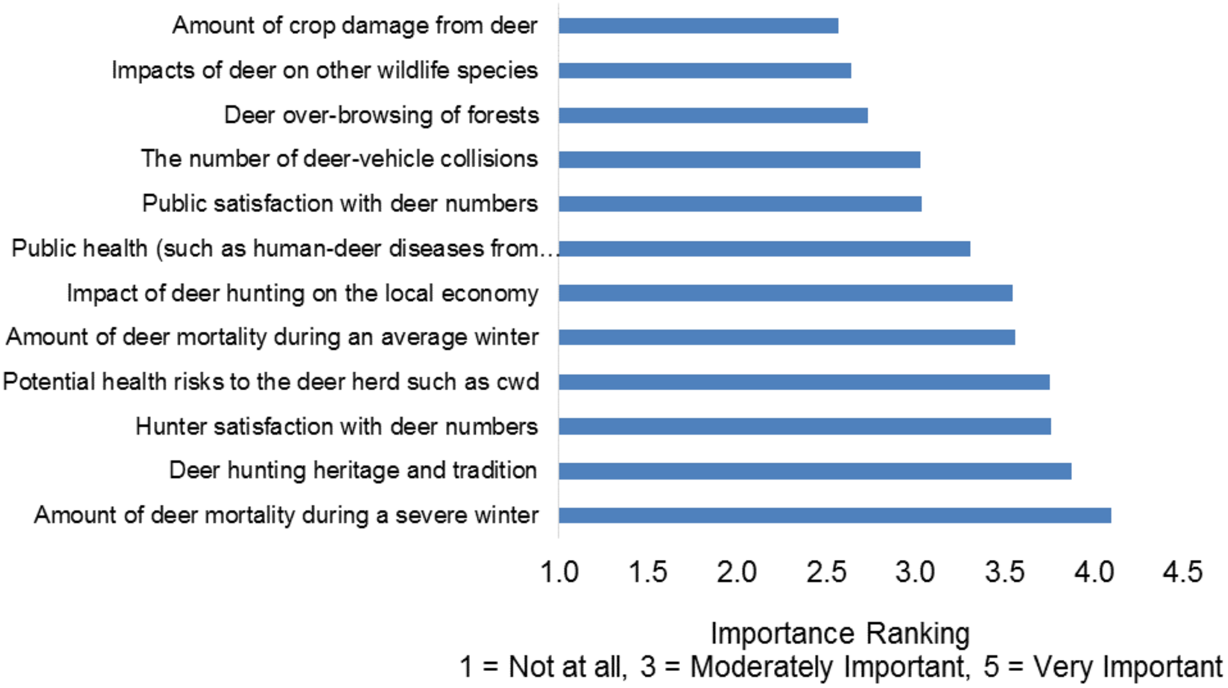
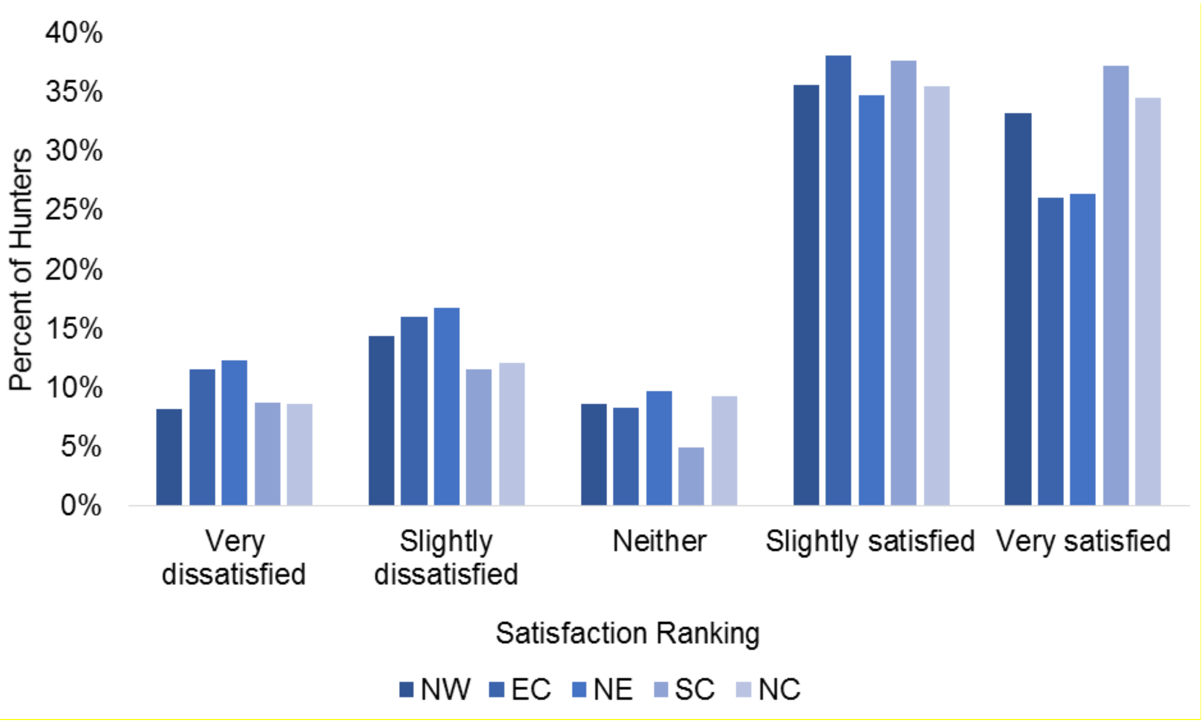
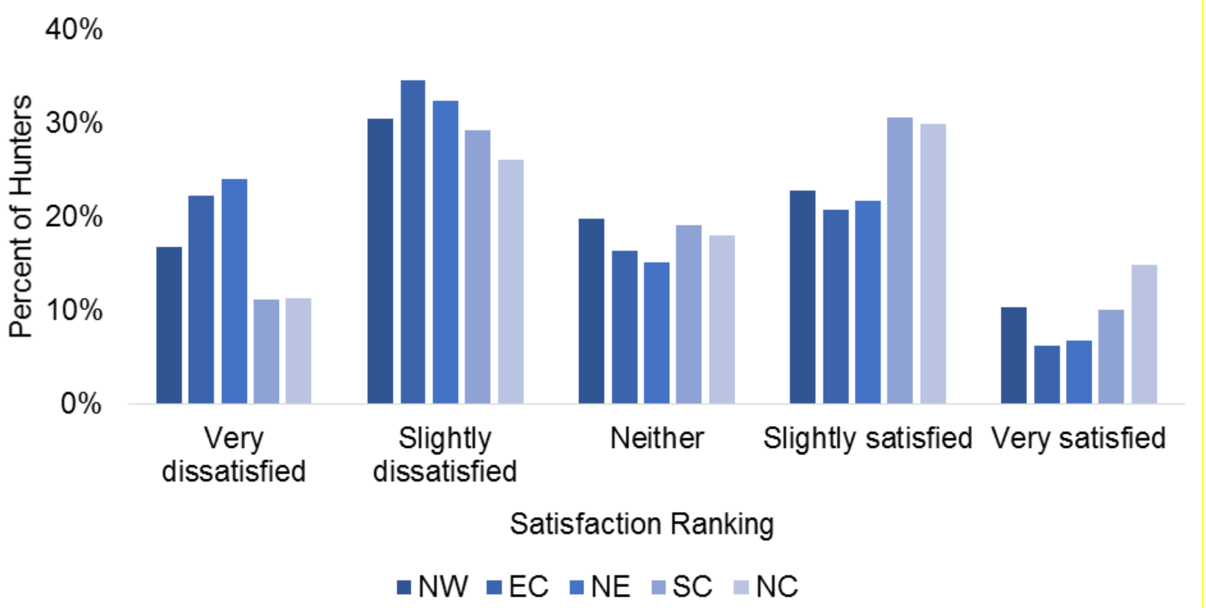


Figure 4. Mean hunter rankings for factors to consider when setting Minnesota deer population goals, 2015-2017. Means reflect weighted averages for all deer permit areas.



(a) Killed a deer for myself or another



(b) Did not kill a deer

Figure 5. Overall Minnesota deer hunt satisfaction based on harvest success, by survey area, 2015-2017. Responses reflect satisfaction ratings from hunters who killed (a) or did not kill (b) a deer during the most recent deer season.

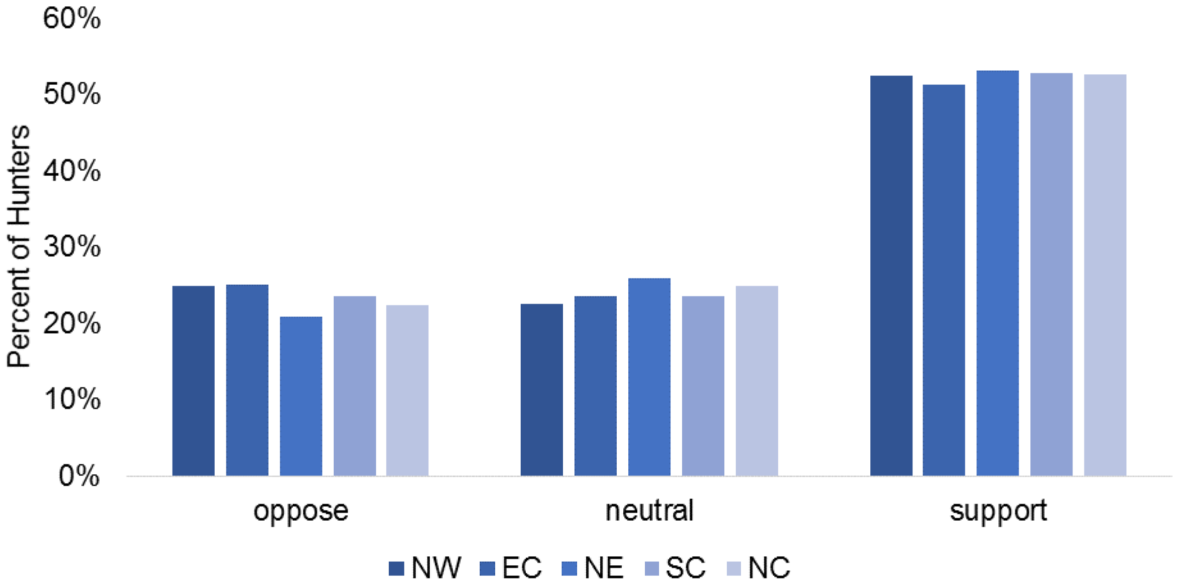


Figure 6. Support for a Minnesota statewide youth season in mid-October, by area, 2015-2017.

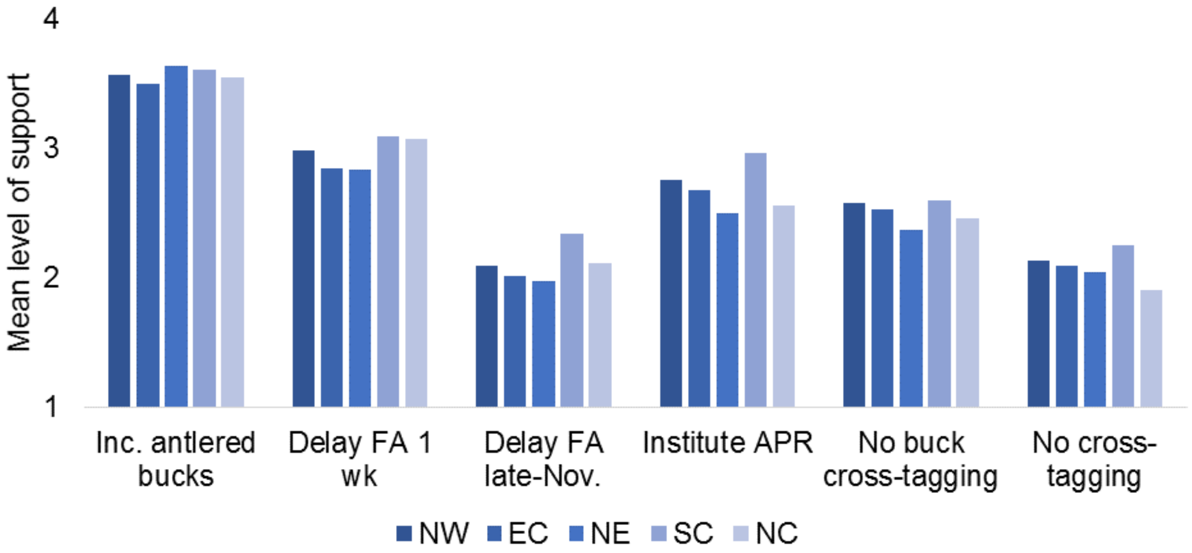


Figure 7. Support for specific Minnesota regulatory alternatives, by area, 2015-2017. Mean is based on the scale: 1 = strongly oppose, 2 = slightly oppose, 3 = neither, 4 = slightly support, 5 = strongly support.

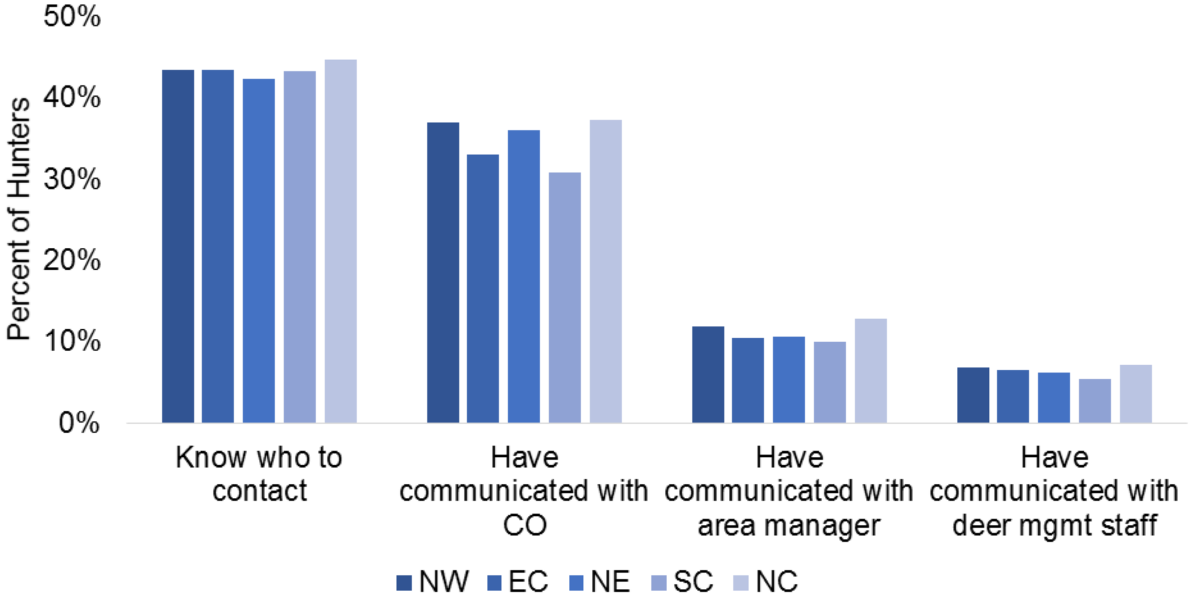


Figure 8. Communication with MNDNR as it relates to deer management, from a survey of Minnesota deer hunters, 2015-2017.

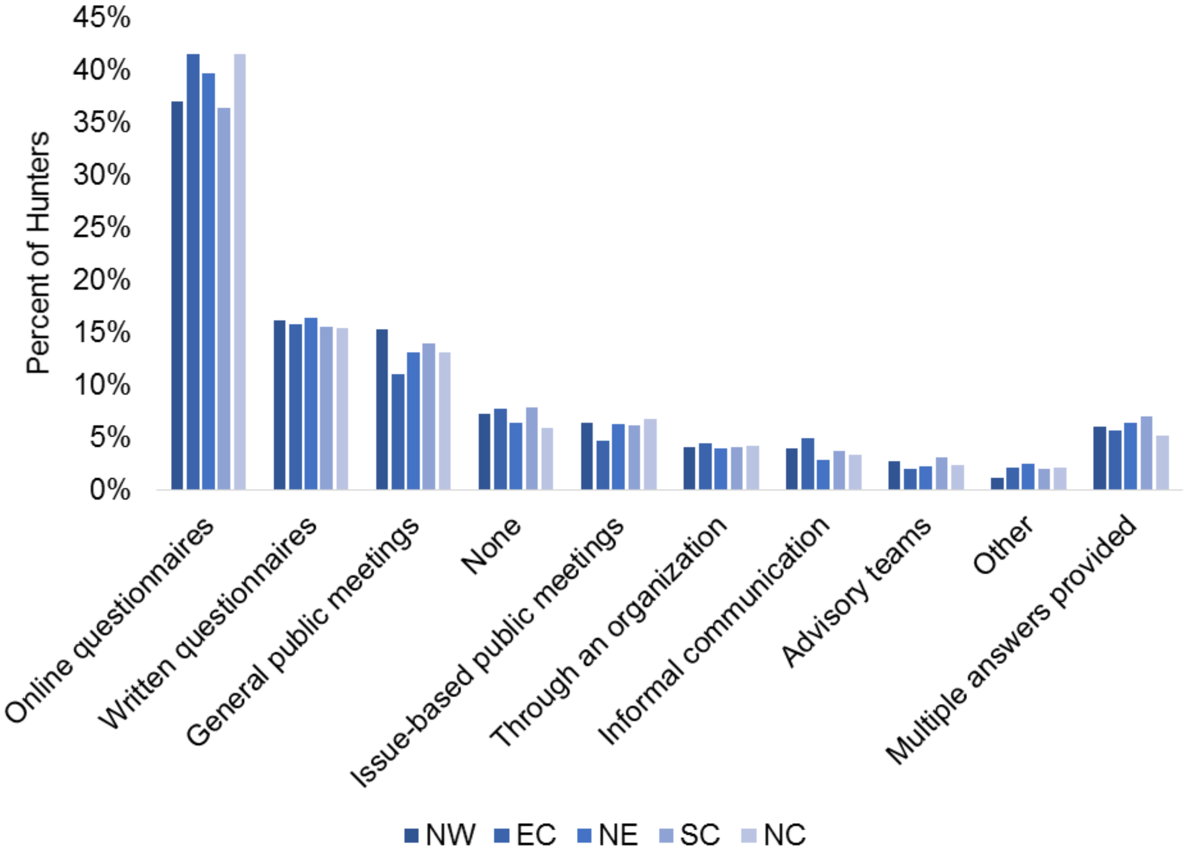


Figure 9. Preferred means to provide input to MNDNR, from a survey of Minnesota deer hunters, 2015-2017.

LANDOWNER ATTITUDES TOWARD ELK IN NORTHWEST MINNESOTA

Eric Walberg, Gino D’Angelo, Lou Cornicelli, & Leslie McInenly

SUMMARY OF FINDINGS

Managing the elk (*Cervus elaphus*) population in northwest Minnesota provides a multitude of challenges ranging from whether or not they should be present at all to how both private and public lands should be managed to benefit elk. This divisiveness has led to strong opinions about both elk and their management. Historically, there has been a lack of information about public attitudes toward elk in northwest Minnesota, which further complicates the issue. Long-term viability of the elk population in northwest Minnesota is largely dependent on public support and tolerance by private landowners. In collaboration with the University of Minnesota, we conducted a self-administered mail back questionnaire of landowners in northwest Minnesota to determine their attitudes and preferences for future elk management. We surveyed 3,000 private landowners in northwest Minnesota within the current elk range and the surrounding area to describe landowner attitudes toward elk and preferences for future elk population management. This report is a summary of activities to date. A more complete analysis and report will be provided in the future.

INTRODUCTION

Minnesota’s natural resources provide many benefits to the state’s residents, including outdoor recreational activities such as hunting, fishing, and wildlife viewing. Elk are valued for aesthetic and intrinsic reasons, recreation including hunting and viewing, and for revenues derived from recreation associated with elk (Wisdom and Cook 2000). Prior to the 1900s, elk ranged over most of the state but were functionally extirpated due to overharvest and habitat loss (Hazard 1982). Restoration efforts near Grygla in the early 20th century and natural immigration from Manitoba and North Dakota into Kittson County have allowed the northwest elk population to increase to approximately 150 elk; however, this estimate fluctuates due to population movement across the International border (Minnesota DNR [MNDNR] 2016). Elk currently exist in 4 localized herds in 2 areas of northwest Minnesota, but the population is managed at low levels to reduce human-wildlife conflicts (MNDNR 2016). The long-term vision is to increase the population size and range of the elk population in Minnesota (MNDNR 2016).

We examined results from a 2016 survey of private landowners in northwest Minnesota to assess attitudes toward elk, preferences for elk population size, and tolerance of elk in northwest Minnesota. The objective was to determine the attitudes of private landowners toward elk and their preferences for future elk management.

STUDY AREA

The overall population of interest were private landowners in parts of Beltrami, Kittson, Marshall, Pennington, and Roseau Counties. Our overall study area included, 1) current elk range (within), and 2) potential elk range (outside) (Figure 1).

METHODS

Within our study area we defined 2 strata: 1) within the current elk range, and 2) outside the elk range. We identified the current elk range by using Global Positioning System (GPS) locations from radiocollared elk (Freeman et al. 2017), visual sightings by local MNDNR staff, and previous elk range estimates that were generated from aerial population surveys. We obtained GPS locations for 20 female elk in northwest Minnesota using location data obtained from February to August 2016. The GPS collars were placed on the female elk as part of a parallel research project conducted in northwest Minnesota (Freeman et al. 2017). We created Minimum Convex Polygons (MCP) within ArcGIS 10.4.1 (ESRI 2015) using Geospatial Modeling Environment (GME Version 0.7.4.0; Beyer 2015). We identified 2 main elk ranges in northwest Minnesota by merging MCPs of GPS locations and visual sightings of elk with estimated elk ranges based on past MNDNR surveys.

We delineated the outside elk range strata as land that contained habitat likely suitable for sustaining a future elk population, although elk are not currently present. We used major roads and the Minnesota border with Canada to delineate the area representing potential elk range. As expected, the potential elk range strata was larger than the current elk range (3,661 sq. miles vs 517 sq. miles).

For both strata, we surveyed individuals who owned at least 0.5 acres. We ultimately conducted a census of landowners within elk range (N = 768) and selected a random sample of landowners outside of elk range (n = 2,232) for a total sample size of 3,000. We obtained addresses through publicly available property tax identification lists. We surveyed landowners using a self-administered mail-back questionnaire based on an adapted Tailored Design Method (Dillman, Smyth, and Christian 2009). We contacted recipients 3 times between October 2016 and January 2017 using a full-length questionnaire with a cover letter. We mailed a shortened version of the survey questionnaire to non-respondents in February 2017, which served as a non-response check.

The survey design was a 12-page questionnaire that was divided into the following categories: (1) land ownership, (2) elk interactions, (3) elk population preferences, (4) attitudes toward elk, (5) tolerance of elk, and (6) recreation. We separated data from the 2 study groups due to differences in past interactions with elk. We did not include questionnaires returned after April 2017 in our analyses.

RESULTS

Between October 2016 and April 2017, 134 surveys were returned due to incorrect addresses or deceased individuals; we received a total of 1,178 completed questionnaires for an adjusted response rate is 41%. Of the 1,178 completed surveys, 390 responses were completed by recipients within elk range (53% response rate) and 788 responses by recipients outside elk range (37%). A non-response check indicated that respondents were slightly older (Mean = 60 years old) on average than non-respondents (Mean = 58 years old) and more likely to be male (87%) than non-respondents (75%).

Attitudes Toward Elk

Overall, a majority of landowners in northwest Minnesota had positive attitudes toward elk (66%; Figure 2). Landowners outside of the current elk range had slightly more favorable attitudes toward elk than landowners within elk range (67% vs. 64%). Landowners outside of elk range (72%) were slightly more supportive of having a wild, free-roaming elk population in northwest Minnesota than landowners within elk range (69%). Both groups were equally supportive of having wild, free-roaming elk living within 5 miles of their property (65% and 65%, respectively) or on their property (56% and 57%, respectively).

Population Size

A majority of landowners (58%) believed that the elk population in northwest Minnesota was too small, 32% believed the population was about right, and 10% believed the population was too large. Less than half of landowners within elk range (46%) believed the elk population was too small, 31% believed the population was about right, and 24% believed the population was too large. A majority of landowners outside of elk range (58%) preferred increasing the elk herd size in northwest Minnesota over the next 10 years, 31% preferred no change, and 11% preferred decreasing the elk population. Less than half of landowners within elk range (49%) preferred increasing the elk population, 28% preferred no change, and 23% preferred decreasing the elk population.

Recreation

A majority of landowners within elk range (53%) and outside elk range (61%) indicated they would be more likely to make a special trip to view elk in northwest Minnesota if the elk population increased. Many landowners both within (68%) and outside elk range (61%) indicated they would allow people other than family members to view elk on their land. Few landowners indicated they (7% and 6%, respectively) would charge people to view elk on their land.

As expected, a small number of landowners both within and outside elk range have hunted elk in Minnesota (6% and 0%, respectively); more have hunted elk elsewhere in North America (23% and 21%, respectively). Among landowners who have not hunted elk in Minnesota, less than one-third have applied for an elk permit in Minnesota (24% and 10%, respectively) or plan to do so in the future (32% and 28%, respectively). A majority of landowners were supportive of legal, regulated hunting in general (86% and 85%, respectively) and elk hunting specifically (83% and 80%, respectively).

Most landowners within (86%) and outside elk range (79%) indicated that they would likely hunt or allow immediate family members to hunt elk if there were elk on or within 5 miles of their land. A majority of landowners within elk range (55%) and less than half of landowners outside elk range (39%) indicated they would allow individuals other than immediate family members to hunt on their land. Overall, 25% of landowners within and 22% outside elk range indicated they would likely lease their land for elk hunting.

Depredation Within Elk Range

Approximately one-third of landowners within elk range (31%) experienced damage from elk during the last year, with row crops (20%) and small grains (19%) reported most often as being damaged. Less than half of landowners (38%) indicated that they had ever experienced damage from elk. Similarly, row crops (22%) and small grains (24%) were the main types of damage the landowner had ever experienced. Landowners experienced relatively minor damage to non-agricultural land uses; however, fences were most frequently damaged during the last year (14%) or during previous years (19%).

ACKNOWLEDGMENTS

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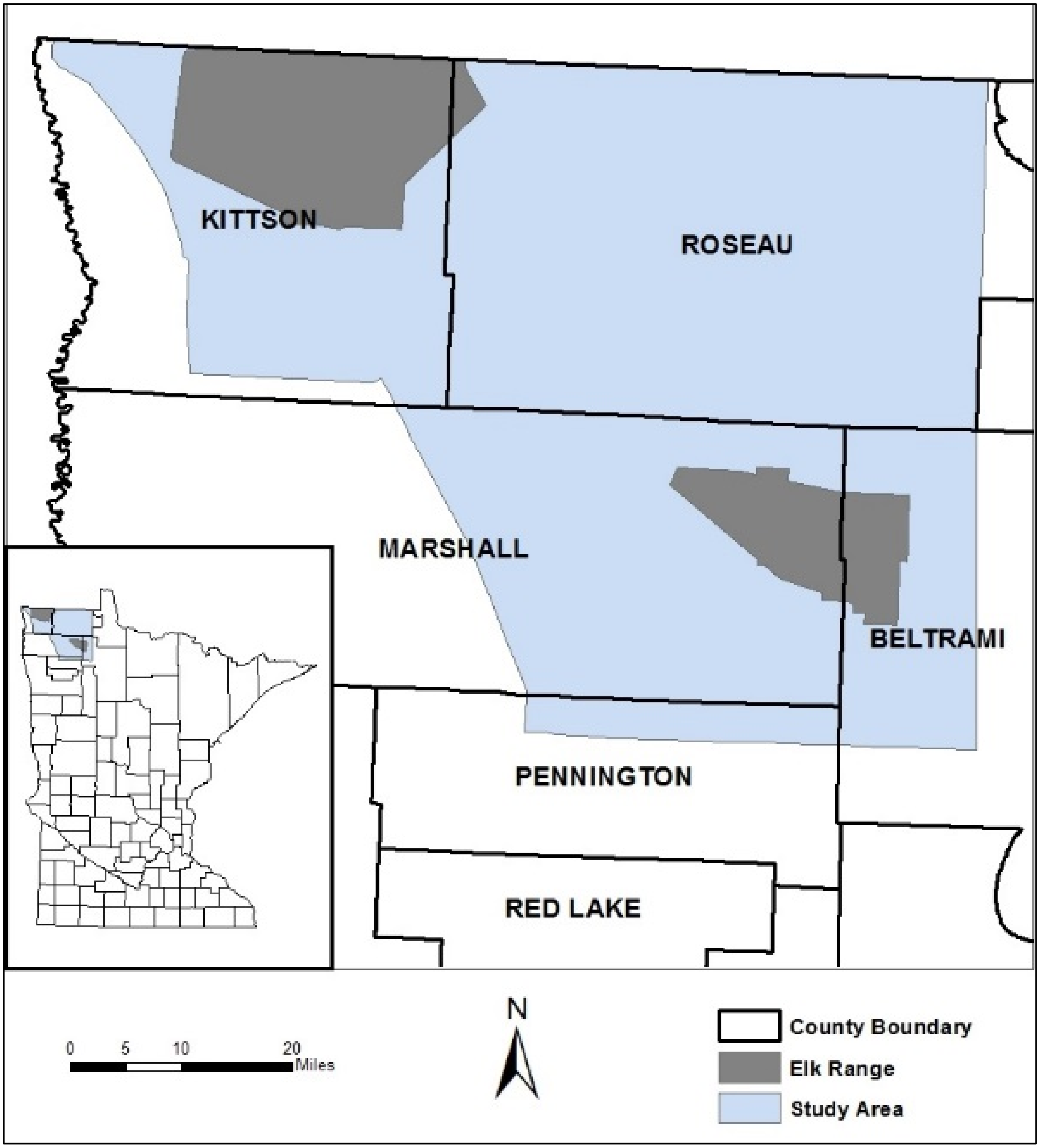

Figure 1. Location of study area in northwest Minnesota. Surveyed landowners to evaluate landowner attitudes toward elk and elk management in northwest Minnesota. Mail survey of 3,000 landowners in northwest Minnesota between October 2016 and April 2017.

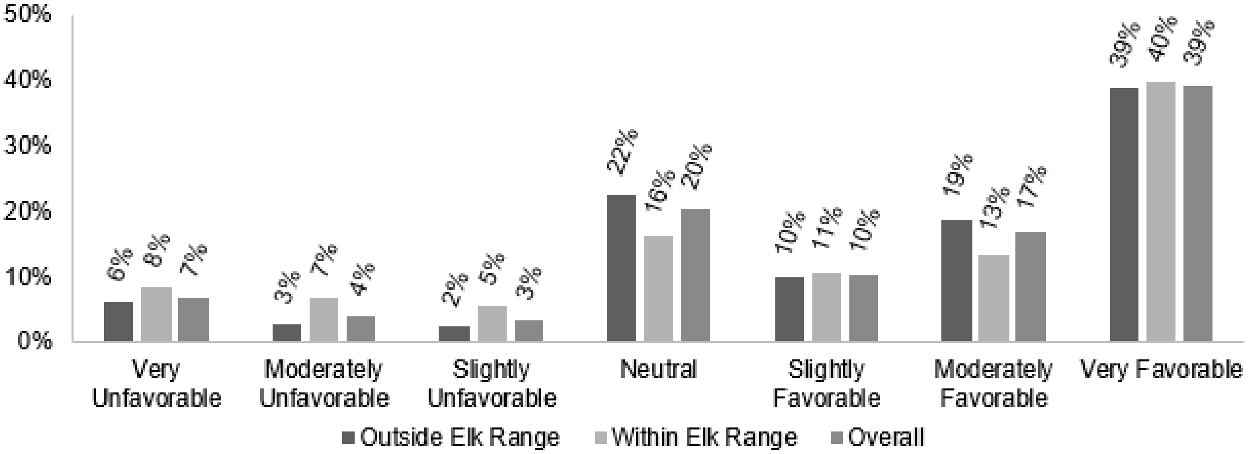

Figure 2. Landowner attitudes toward elk in northwest Minnesota. Mail survey of 3,000 landowners in northwest Minnesota between October 2016 and April 2017.

UNDERSTANDING USER PREFERENCES AND VISITOR NUMBERS AT MINNESOTA WILDLIFE MANAGEMENT AREAS[1]

Kelsie LaSharr, Louis Cornicelli, and John Giudice

SUMMARY OF FINDINGS

The Wildlife Management Area (WMA) land classification was created as part of the Outdoor Recreation Act of 1975 (Minnesota Statutes MS 86A). They were established “to protect lands and waters that have a high potential for wildlife production, to develop and manage these lands and waters for the production of wildlife, public hunting, fishing and trapping, and other compatible outdoor recreational uses”. The WMA system is administered by the Minnesota Department of Natural Resources (MNDNR), Section of Wildlife and is currently comprised of about 1,440 units totaling over 1.3 million acres. Given the breadth of the WMA system, DNR staff were interested in understanding how people use these lands, which activities they enjoy pursuing, and the number of individuals recreating during peak hunting seasons. WMAs are used most frequently during fall hunting seasons for both big and small game. To achieve our research goals, we 1) intercepted hunters in the field during the 2015-2016 fall hunting season, and 2) solicited respondents through small game license and pheasant stamp sales. This research summary provides background information, methods, and some preliminary study results.

INTRODUCTION

Wildlife management agencies are tasked through the public trust doctrine to protect, conserve, allocate, and control wildlife and their habitat for the benefit of the public (Jacobson et al. 2010). Managers must understand ecology and anthropogenic interests, and be willing to integrate both in order to achieve socially desirable benefits associated with wildlife (Forstchen and Smith 2014; Organ et al. 2014). One important public benefit is wildlife-based recreation opportunities, such as hunting, fishing, and viewing (Driver 1985). Spending time outdoors, through wildlife-related activities, has been shown to have many economic, environmental, and social benefits, as well as providing satisfaction on multiple levels for the individual participant. Specifically, being in nature has well-documented short-term and long-term effects on both the psychological and physiological aspects of human life (Wolf et al. 2014; Mayer et al. 2009, Haluza et al. 2014). While demand and interest in some recreational activities has reached all-time highs (e.g., birdwatching; Cordell 2008, Outdoor Foundation 2014), hunter numbers nationally are declining (U.S. Department of the Interior 2012, Vrtiska et al. 2013). Hunting has been shown to be an important beneficial activity sought by the public (Hammitt et al. 1990; Decker et al. 1980; Brown et al. 1977; Hendee 1974; Driver 1985), and it is important to understand which barriers, such as lack of access to land, may be pressuring hunters to drop-out of the activity.

Publicly accessible properties provide a crucial resource for protecting wildlife habitat and providing recreational opportunities. Hunting opportunities on publicly-owned state land are

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[1] Several all-day “correction factor” shifts were conducted at a limited number of sites to determine the difference in visitation between morning and afternoons at single WMAs.

especially important in the east and Midwest United States, where the majority of lands are in private ownership. Publicly restricted hunting, such as opportunities on private lands, continues to decline as these properties are parceled, sold, and otherwise fragmented (Larson et al. 2014). Land that is owned in a checkerboard pattern also becomes problematic for hunters when barriers to quality habitat are formed by unpassable private property. As such, it is important to maintain public land for hunters to use, especially when private land is otherwise not available. Wildlife management agencies can ensure the public obtains benefits from publicly managed, wildlife-producing lands by better understanding desired outcomes and motivations of hunters (Schroeder et al. 2006, Hayslette et al. 2001; Decker et al. 1980; Hammit et al. 1990). In turn, these agencies can help ensure hunters are recruited, retained, and reactivated for generations to come (Larson et al. 2014).

OBJECTIVES

The overall goal of this research was to improve the understanding of visitor use at MNDNR WMAs. Specifically, our objectives were to:

1. Characterize WMA users through an increased understanding of beliefs, values, and satisfactions they associate with using WMAs.
2. Determine participation levels by estimating visitor usage during fall hunting seasons (September through December).

METHODS

Study Area and WMA Selection

Our study area covered 43 counties located in the prairie pothole region of western Minnesota and contained 1,061 WMAs. We divided the study area into 2 regions (northwest and southwest) based on the abundance of WMAs found in these areas (Figure 1). The northwest study area is best characterized by larger counties, and fewer but larger WMAs. Conversely, the southwest study area has smaller counties and more, albeit smaller WMAs (Table 1). Using ArcMap 10.2, we created a sampling grid for each region, with grid size being a function of average county size within each region (Figure 1). Within each of the 21 grid blocks we randomly selected 1 WMA and the 9 nearest neighbors to create a cluster of WMAs for sampling visitor usage. Cluster size was modified in some cases because of access issues and to ensure equal sampling effort. The final sample consisted of 228 WMAs organized into 21 driving routes (clusters of WMAs).

Field Observations

To estimate visitor use, we used methods adopted from Fulton and Anderson (2003), modified from techniques recommended by Gregoire and Buyoff (1999) and Watson et al. (2000). Sampling occurred on weekend days (Saturday and Sunday) over an 11-week period from September 26, 2015 (waterfowl opener) to December 6, 2015. Observers drove a specified route over a set time period of four hours, with each sample WMA being surveyed once per weekend day. This resulted in a point-in-time sample of observed WMA user groups. We also surveyed a subset of WMAs intensively (repeated visits from sunrise to sunset) to estimate probability of intercept, which we defined as the average proportion of total user groups per site-day that we intercept at a random point in time. We then used the probability of intercept to convert observed counts (point-in-time) to expected total user groups per site-day.

Visual observations of vehicles (parties) at each WMA were recorded using unique identifying information (license plate numbers), and in addition, field surveyors left intercept letters on all intercepted vehicles. The letters briefly explained the project, a future mail survey, and invited WMA visitors to record their party information (ages of visitors, time spent at individual sites, date), as well as contact information for that future survey work. Accompanying the intercept letters were self-addressed stamped envelopes for visitors to return their invitations to the researchers. Returned intercept letters were used to create a database of WMA visitors who were later sent surveys in spring 2016.

Because sample WMAs were organized into driving routes, efforts were taken to ensure the same WMAs were not sampled at the same time of day each week. Technicians alternated driving routes forwards and backwards from week to week, as well as the order that assigned routes were visited each weekend. This ensured the same WMAs were not repeatedly observed early in the morning or later in the afternoon when traffic was likely to be reduced. A survey protocol was developed to record vehicle counts and license plate information in order to count unique visitors. Each technician (n = 18) surveyed 2 routes per weekend (1 route per day) for a minimum of 4 hours per route.

Visitor Estimate Analyses

For each of the 1,061 WMAs within our study area, we determined a series of site attributes using GIS data layers obtained from MN Geospatial Commons, with data processing accomplished using ArcGIS 10.3. We obtained information on WMA name, county locality, area, nearest town, species present (including deer, small game, forest upland birds, sharptailed grouse, pheasants, waterfowl, turkey, and doves), managed parking areas, dominant cover types, perimeter length, and easting/northing vectors. We also determined how far each WMA was from various points of interest via Euclidian distance: US Fish and Wildlife Service Wildlife Production Areas (WPAs), other WMAs, major roads, and towns of various densities.

We used a linear mixed-effects to explore the relationship between average car counts/WMA/day and WMA attributes, with the goal of predicting expected mean user groups per weekend day for all WMAs in our sampling frame.

Mail Surveys

Data were collected using mail-back surveys following a process adapted by Dillman (2008) to increase response rates. Respondents were sent questionnaire-booklets with personalized cover letters, and included a business-reply envelope to return their responses. Potential respondents were sent multiple contacts 4 times between March 2016 and July 2016. The cover letter explained the purpose of the study and requested respondents to complete and return the survey. The back of the personalized cover letter contained a map of all of the counties in Minnesota, to help respondents address questions in regards to where they specifically hunted during the 2015-2016 hunting season. About 5 weeks after the first mailing, a second mailing (identical to the first) was sent to individuals with valid addresses who had not yet returned their survey-booklets. A third mailing was sent approximately 4 weeks after the first mailing, containing identical information as a final attempt to elicit responses from those who had not participated in the study yet. Any surveys returned after July 1st were not included in the results. Surveys were collected and double-entered into an Excel template. At the end of July 2016, a shortened 1 page, two-sided survey and a business reply envelope was sent to individuals who had not responded by July 1st to identify any non-response bias within the sample. Any respondents who returned their non-response surveys after August 31st were not included in the results.

Data were double-entered into Excel 2010 and comparisons between databases were completed in Excel to look for discrepancies. Further data cleaning and manipulation was completed in Program R (version 3.2.5). Statistical analyses and tables were completed in Program R. Mail survey responses were analyzed in the aggregate. Analysis of the mail survey focused on descriptive statistics, including reporting frequencies, average responses, chi-square test statistics, and F-statistics from an analysis of variance, when appropriate.

RESULTS

Field Observations

Technicians stopped 2,493 times over the field season, where they observed 2,093 vehicles at our selected WMAs. This resulted in an average 0.83 cars seen at each stop (averaged over all sites throughout the entire field season). The average size of an observed WMA was 274.9 acres, which is comparable to the average size for all WMAs in our study area, 278.8 acres. Returned intercept letters from hunters indicated that the average party size for each car was 1.9 people, and they stayed for 3.9 hours, on average. We utilized wildlife managers' experience and knowledge about WMA users to investigate potential trends in visitor use based on site attributes. Our best-supported predictive model for mean user groups/day included positively correlated fixed effects for the presence of pheasants, distance to a major road, and WMA size.

Analysis is on-going; final results will be presented in a future report.

Mail Surveys

We distributed 2,046 invitations during our field season and 405 were returned (20% invitation return rate), which yielded 443 individuals who provided information to receive a WMA visitor survey. Given the small sample population (n = 443), we opted to recruit additional respondents into the study using individuals who purchased both a small game license and pheasant stamp. We randomly selected 5,000 people from the MNDNR ELS database and sent them an invitation letter that explained the study, asked if they hunted WMAs, and if they would they be willing to participate in the WMA user study. Each mailed intercept letter included a postcard that could be returned with an affirmation they had hunted a WMA during the past hunting season and were willing to participate. In total, 88 were undeliverable and 932 were returned, which brought our effective sample population to 1,375.

Of the 1,375 full-length surveys that were sent out, 11 were undeliverable and 1 requested not to participate due to his guardian's concerns for his status as a minor. This resulted in 1,363 viable surveys, of which 593 were returned during the first wave, 267 were returned after the second wave of mailing, and 95 were returned after the third and final wave of mailing (Cut-off date was July 1st, 2016). This resulted in 949 surveys returned, a 70% return rate. This can be further broken down by respondent type. For respondents who were contacted in the field, 288 out of 443 respondents completed the original survey (65%). For respondents who were contacted with a mailed letter due to their status as a pheasant stamp and small game license holder, 661 out of 932 completed the original survey (71%). Non-response surveys were sent to 418 respondents, of which 4 were undeliverable. Of the 414 viable surveys, 141 were returned on or before August 31st, a 34% return rate (Table 2).

Only 2 demographic questions demonstrated a statistical difference between the field-intercepted and the postcard-recruited respondents, and these were Education ($p = 0.045$) and Income ($p = 0.011$) (Table 3). Satisfaction and participation in regards to individual species/seasons was similar between the 2 groups. Overall, 58% of field-intercepted respondents indicated they hunted pheasants, as compared to 70% of postcard-recruited respondents. This outcome was expected given we recruited individuals who purchased a small game license and a pheasant stamp; however, it is unlikely this difference influenced overall survey results. Results of the shortened non-response survey indicate that respondents who did not respond to the original mailing followed similar trends in terms of hunting on a WMA during the 2015-2016 hunting season.

The average user in this study was a 51 year old, white male. We found respondents had the highest interest in pheasant hunting (80%), followed by duck hunting (37%) and firearm deer hunting (31%). Over half of respondents (55%) recorded being moderately to extremely satisfied with their WMA overall experiences and 64% were "Extremely Likely" to return to a WMA. However, 63% of WMA hunters did not use WMAs for any activity outside the hunting season. Strikingly, 38% of users indicated that they use Private Land "None" of the time, demonstrating the importance of having public land available for hunting use in Minnesota.

We found a majority of respondents agreed that the number of WMAs should be increased (86%). More than half of the respondents agreed WMAs provide high quality hunting experiences (63%); however, they feel that WMAs are too crowded (62%) and there are not enough WMAs located near them (61%). We found the biggest constraints to hunting WMAs fell into the following categories: 1) Not enough game, 2) Lack of time, and 3) Family/relationship responsibilities. Two sources of information were found to be "Moderately to Extremely Important" for more than half of respondents 1) spotting WMA signs in the field (60%) and using WMA boundary maps (58%). Finally, about half of respondents indicated that they never use lead shot on WMA properties.

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Table 1. Study Area Differences from a survey of wildlife management area users in Minnesota, 2015-2016.

	Northwest region	Southwest region
Number of counties	14	29
Total Area (mi^2)	13,836	19,931
Average county size (mi^2)	988	687
Total WMAs per region	315	809
Average WMA size (mi^2)	0.58	0.33
Proportion of WMAs with RNPH[1]	0.345	0.929

[1] Ring-necked Pheasants

Table 2. Survey Response Rates from a survey of wildlife management area users in Minnesota, 2015-2016.

Sample type	Surveys administered (n)	Surveys returned (n)	Survey response rate (%)	Non-response surveys administered (n)	Non-response surveys returned (n)	Non-response rate (%)
Field intercept	443	288	65%	149	69	46%
Postcard	932	661	71%	269	72	27%
Total	1,375	949	70%	418	141	34%

Table 3. Demographic differences between sample populations of wildlife management area users in Minnesota, 2015-2016.

Demographic variable	Field-intercept mean	Postcard-recruited mean	p-value
Average age	51.5 years	50.5 years	0.289
Age at first hunt	13.7 years	14.4 years	0.500
Income[1]	$72,654	$82,228	0.011
Miles driven	98.3 miles	101.0 miles	0.710
Male	96%	97%	0.336
Took a dependent	45%	46%	0.874
Took a spouse/partner	18%	18%	0.860
Proportion of pheasant hunters	58%	70%	<.001
Percent duck hunters	37%	29%	0.012
Percent firearm deer hunters	38%	22%	<.001
Percent archery deer hunters	15%	11%	0.09
Percent muzzleloader deer hunters	12%	7%	0.016
Education	5.7 (out of 10)	6.1 (out of 10)	0.006

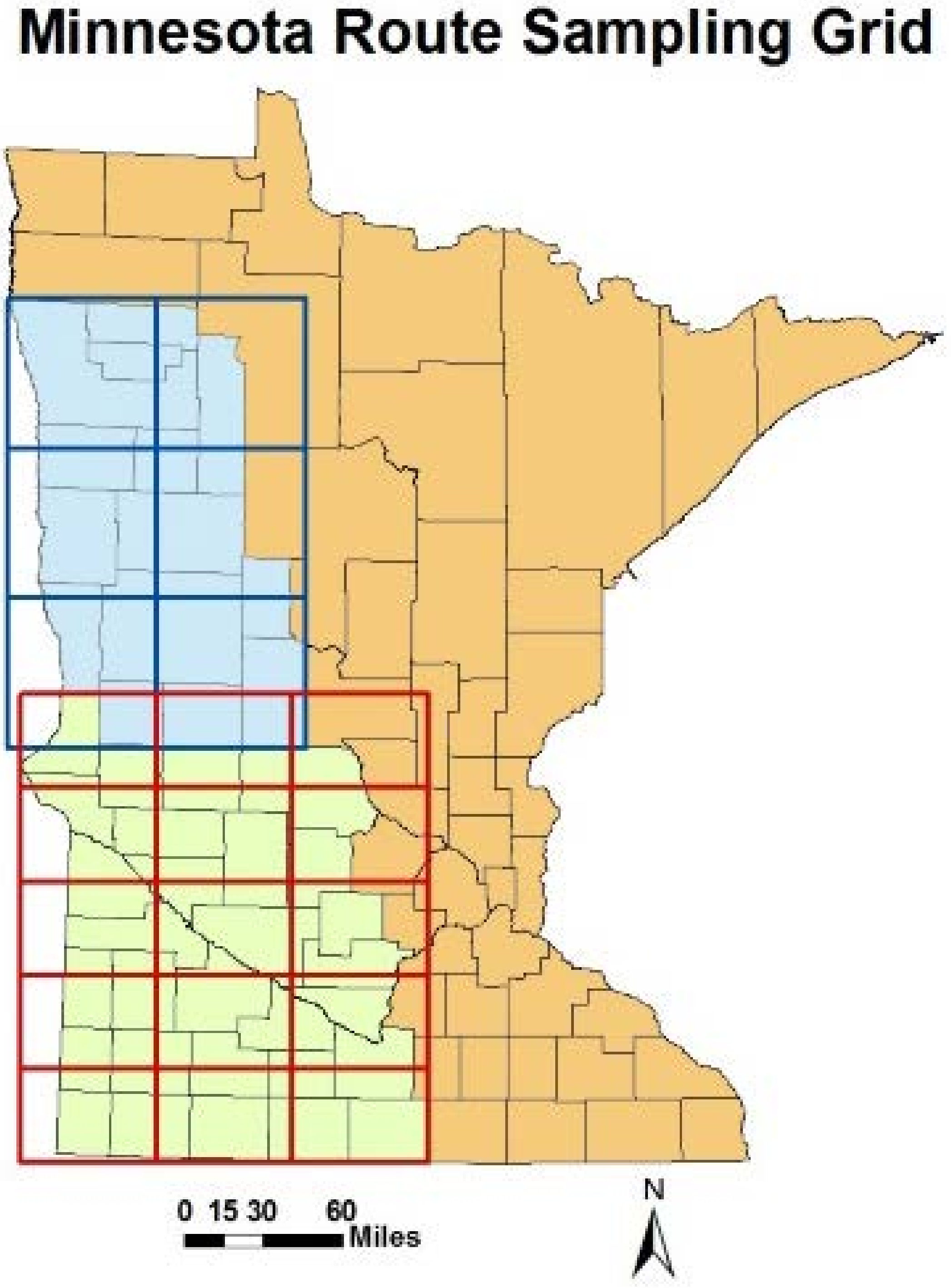

Figure 1. Overlay of northwest and southwest grid blocks on corresponding Minnesota counties, 2015-2016. Note the larger counties in the northwest were sampled with larger, fewer grids, while the smaller counties in the southwest were sampled using a higher number of smaller grids.

Publications Lists

Forest Group Publications

Berg, S. S., **J. D. Erb**, V. T. Spaid, **P. L. Coy**, **B. A. Sampson**, J. R. Fieberg, T. W. Arnold, and J. D. Forester. *In revision*. The role of nearby large-diameter cavity trees in the selection of den sites by female fishers (*Martes pennanti*) in northern Minnesota.

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