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## Bat Use of Afforested and Encroached Patches and Their Role in Extending Bat Habitat into the Nebraska Sandhills

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Bat Use of Afforested and Encroached Patches and Their Role in Extending Bat Habitat  
into the Nebraska Sandhills

by

Jacob L. Wagner

A THESIS

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Bat Use of Afforested and Encroached Patches and Their Role in Extending Bat Habitat  
into the Nebraska Sandhills

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University of Nebraska, 2023

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The Nebraska Sandhills are currently undergoing a state shift to a redcedar dominated woodland due to anthropologic planting of eastern redcedar (*Juniperus virginiana*) and woody encroachment from the periphery of the Sandhills. To better understand this novel ecosystem and how bats are utilizing it I collected data at Barta Brothers Ranch with acoustic sensor grids consisting of 24 100m spaced acoustic sensors placed adjacent to planted windbreaks. Supplemental data from the Nebraska North American Bat Monitoring Program was used for data analysis at larger spatial scales. I used linear regressions and kriging interpolation maps to see how bats used windbreaks during nightly activity. *Eptesicus fuscus*, *Lasiurus borealis*, and *Nycticeius humeralis* showed close usage activity around windbreaks while *Lasiurus cinereus* and *Lasionycteris noctivagans* showed even usage activity throughout the sensing area, while still showing a relationship with trees at a larger scale. I used multi model inference and model averaging to find the best models to explain bat species richness and call count. I found that distance to trees, with a preference for deciduous trees, and time through the summer to be the best predictors of bat species richness and call count. I used linear regressions of first instance calls in my grids to determine if bats are using windbreaks to roost or travel from roosts to feeding grounds. Across all species, *Eptesicus fuscus* and

*Lasionycteris noctivagans* showed greater numbers of first instances closer to the windbreak, which indicates that, in general, bats are using windbreaks for roosting or for pathways between roosts and feeding grounds. *Lasiurus cinereus* first instances were not related to windbreaks hinting that this species may travel across open grassland from roost to feeding grounds. Bat presence and use of trees in the Sandhills presents a dilemma of ecosystem service tradeoffs, in which management aimed at controlling the spread of woody plants for grassland diversity and forage quality and quantity may reduce habitat for bat species.

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## CHAPTER 1 - INTRODUCTION

The Nebraska Sandhills ecoregion is a semi-arid mixed grass prairie composed of rolling grass-stabilized sand dunes. The Sandhills is part of the larger Great Plains region with sporadic grasslands stretching from Canada to Texas (Augustine et al. 2021). Like other Great Plains ecoregions, the Nebraska Sandhills is transitioning from grassland to a cedar dominated woodland, as a consequence of afforestation through planting of eastern redcedar (*Juniperus virginiana*) in windbreaks and woody encroachment from those windbreaks and the periphery of the Sandhills, creating a novel Sandhills ecosystem (Donovan et al. 2018; Fogarty et al. 2022a, 2022b; Scholtz & Twidwell 2022). The spread of eastern redcedar in grasslands lowers grassland biodiversity and herbaceous cover, as herbaceous cover under dense redcedar canopies can decrease by as much as 99% (Briggs et al. 2002). The negative effects redcedar has on grasslands make cedar dominated woodlands undesirable by most parties (Roberts and Allen 2023). There are efforts to control redcedar spread in Nebraska with fire, chemical and mechanical removal (Bidwell & Weir 2017), but removal rates do not match the rates of spread with 40,000 acres of grasslands being lost each year to redcedar invasion. Tree cover in Nebraska's grasslands has doubled since 2000 and is now approaching one million acres (Fogarty 2022). Eastern redcedar trees are still being planted and are valued in Nebraska as a windbreak species that protect homes and livestock from wind and inclement weather. Some Nebraska agencies distribute eastern redcedar as a 'conservation species' with around 850,000 seedlings distributed annually (Ganguli et al. 2008), while other governmental organizations incentivize their removal (Roberts et al. 2018).



Bat population globally and in the United States are declining in part due to habitat loss/decline, white nose syndrome, and problems migrating bats have with wind turbines. White nose syndrome is a fungal infection that is especially harmful to northern long-eared bat (*Myotis septentrionalis*) (Reynolds et al. 2016), which is a federally listed endangered species. White nose syndrome disrupts hibernation, increasing activity in bats and lowering survival rates. White nose easily spreads through hibernacula, infecting whole colonies, causing high mortality and, in some cases, local extirpation (Frick et al. 2015, Warnecke et al. 2012). Migrating populations are also threatened by mortality from wind turbines, especially during months of migration (Arnett et al. 2008, Cryan et al. 2014). North American forests compositions have changed since pre-European settlement (Nemec et al. 2011). The spread of species such as eastern redcedar (*Juniperus virginiana*), russian olive (*Elaeagnus angustifolia*), and autumn olive (*Elaeagnus umbellata*), in forests causes midstories to become cluttered and limits the use of these habitats by larger bat species (Sleep & Brigham 2003).

Bat population declines and habitat degradation by invasive and problem species make it important to understand bat utilization of the emerging, novel ecosystems of cedar woodlands in the Nebraska Sandhills. There is minimal data on bat habitat use in the Sandhills and assumptions about habitat are likely to be based upon the Sandhills being a mostly tree free area, which is becoming less of a reality each year. In this thesis I examined bat use of the Nebraska Sandhills using acoustic sensor grids and data from the North American bat monitoring program (North American Bat Monitoring Program [NABat] [usgs.gov]). I examined how bats are spatially utilizing cedar windbreaks, what features and variable are driving bat activity and species richness, and if redcedar

windbreaks are being utilized for roosting or travel from roost to feeding grounds. The use of this novel ecosystem by bats would create a dilemma for land managers and stakeholders as to what ecosystem services, species, or habitats are more valuable and which to protect when the presence of one seems to be at odds with another. The presence of the federally listed northern long eared bat could take priority to tree removal, as habitat suitability for this species includes wooded fencerows and live trees  $\geq$  3" diameter at breast height (DBH) (Endangered and Threatened Species: Northern Long-eared Bat, 2023), which includes most mature redcedar trees.

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## CHAPTER 2 – BAT UTILIZATION OF NOVEL AFFORESTED ECOSYSTEM IN TRANSITIONING GRASSLANDS

### Introduction

The Nebraska Sandhills is a semi-arid region in Nebraska comprised of rolling grass-stabilized sand dunes, vegetated with mixed grass prairie. The Nebraska Sandhills are part of the larger Great Plains grasslands, which extend from Canada into Texas and which have analogs globally; all these grasslands are threatened by afforestation and/or woody plant encroachment (Scholtz & Twidwell 2022). In the past century, portions of the Sandhills have become cedar-dominated, creating a novel Sandhills ecosystem (Fogarty et al. 2022a, 2022b). The current management practice is to cut, burn or chemically treat cedars to control their spread (Bidwell & Weir 2017), but rates of control are below rates of spread. Furthermore, cedar windbreaks are still valued and planted. The annual average distribution in Nebraska of eastern redcedar as a ‘conservation species’ by the state is 850,000 seedlings (Ganguli et al. 2008). At the same time, other governmental agencies incentivize cedar removal (Roberts et al. 2018).

Most woody species in Nebraska, including many Oak (*Quercus* sp.) species, can resprout after a fire. This is not the case for eastern redcedar, whose spread has been historically suppressed by fire (Briggs et al. 2002). According to early European settlers, eastern redcedar were contained to streams, ravines and lowlands and were not present in upland grasslands (Blewett 1986). Fire has been a part of Great Plains grasslands, either from wildfire or from indigenous land management practices, for thousands of years (Courtwright 2011). Most grassland species are fire tolerant and have little negative impact from fire or benefit directly. Eastern redcedar is susceptible to fire; fire

suppression practices along with human planting has caused cedar to invade grasslands where it was not historically present (Streit Krug et al. 2017). Eastern redcedar is drought tolerant and a habitat generalist, thriving in many different soil, climate and topographic settings (Van Haverbeke 1976). Eastern redcedar is invading grasslands from the perimeter and more insidiously, from windbreak plantings throughout the Great Plains (Donovan et al. 2018; Fogarty et al. 2022b). Approximately 40,000 acres/year of grassland is lost to cedar invasion in Nebraska each year. Eastern redcedar changes the composition of grassland ecosystems; herbaceous production can be reduced as much as 99% in closed canopy cedar forests (Briggs et al. 2002), making cedar forest and woodland a largely undesired alternative stable state of Great Plains grasslands.

In grasslands where eastern redcedar was introduced or invaded, this novel woody structure could provide new habitats for bats. Eastern redcedar structure, for individual trees, patches and forests, is very dense, and while bat species are not believed to forage under the canopy of cedar like they do in deciduous forests, bats could use the three-dimensional structures above and around eastern redcedar canopies. If bats do utilize this novel afforested grassland habitat, it creates a management conundrum. Humans planted cedar windbreaks for protection from the wind for their homes and cattle. If bats are utilizing these afforested lands, these novel habitats create a novel challenge, as current efforts are underway to eliminate or reduce these forests because of their negative impacts on rangeland productivity, and because of the proclivity for cedar to invade beyond their plantings. The presence of federally listed bat species could preclude tree removals. The current habitat suitability model for the federally listed northern long-



eared bat includes wooded fencerows and live trees  $\geq 3''$  DBH (Endangered and Threatened Species: Northern Long-eared Bat, 2023).

Bat populations in Nebraska and globally face habitat degradation and change. Forests that were present before European colonization have changed in composition (Nemec et al. 2011), and tree planting along with biological invasions have increased the geographic extent, and abundance, of forest cover. Tree cover in Nebraska's rangelands has doubled since 2000 and is now approaching one million acres (Fogarty 2022). These forests include introduced and invasive species like Russian olive (*Elaeagnus angustifolia*), Autumn olive (*Elaeagnus umbellate*) and eastern redcedar (*Juniperus virginiana*) which cause the mid-story of forests to become cluttered, and lead to woody encroachment of former grasslands. Most afforestation in the Sandhills proper is due to monocultures of cedar which has a very dense, cluttered, sub-canopy structure. Cluttered mid-stories create blockages and cause physical limits for large bat species that attempt to forage within them (Sleep & Brigham 2003).

There are thirteen species of bats in Nebraska, including the federally threatened northern long-eared bat (*Myotis septentrionalis*). All thirteen bats are insectivorous, and their diets primarily consist of flying insects. These bats provide ecosystem services to farmers and the public by consuming many invertebrate pests. Bat species such as little brown bat (*Myotis lucifugus*) and big brown bat (*Eptesicus fuscus*) hunt in open habitats and edge habitats for flying insects (Jones et al. 2016). The estimated cost that the U.S. farming industry saves due to insect pest consumption by bats is estimated to vary between 3.7 billion to 53 billion US dollars per year (Boyles et al. 2011), although the assumptions used in that estimate have been questioned (Fill et al. 2022) because bat

activity in agriculture, including rangelands, is not uniformly distributed. Bat insect predation has positive impacts for humans as well. An individual little brown bat (*Myotis lucifugus*) can eat more than a thousand mosquito-sized insects in one hour, though many bats will target larger higher calorie insects such as moths (Ducummon 2000). Thus, bats reduce populations of insects that can vector diseases and reduce crop yields.

Bat species, in many locations, including much of North America, have been in decline, especially in those species susceptible to white-nose syndrome such as the northern long-eared bat (Reynolds et al. 2016). White-nose syndrome interrupts hibernation forcing bats to burn stored fats. This causes individuals to have a much-reduced survival rate, spreading the fungus throughout winter colonies leading to population declines and, in some cases, local extirpation (Frick et al. 2015, Warnecke et al. 2012). Additionally, there is evidence that wind turbines may be negatively affecting bat abundance. Mortality around wind turbines increases during periods of bat migration and species that migrate are at higher risk (Arnett et al. 2008, Cryan et al. 2014). Given population declines in many species of bats, and declines in the quality of many habitats for bats due to habitat loss and degradation of forest understories by invasive species, it is important to understand bat use of novel anthropogenic habitats, such as windbreaks in grasslands.

I examined the spatial utilization of afforested grasslands by Nebraska bat species in the Nebraska Sandhills, using acoustic sensor grids. The objective for this study is to determine if Nebraska bats are using planted eastern redcedar windbreaks as habitat extensions and novel habitats into Nebraska grasslands. Research reports low utilization of grasslands by bats, and utilization is reduced with distance from forested features

(Treitler et al. 2016). I hypothesize that bats will use windbreaks as a feeding and roosting habitat and utilize the windbreak at a higher rate than the grassland, enlarging bat habitat in the Sandhills, increasing bat habitat overall, but introducing tradeoffs in management for ecosystem services, as redcedar reduction in this ecosystem is a state and regional priority.

## **Methods**

### **Study Area and Site Selection**

I conducted this study at Barta Brothers Ranch in the Nebraska Sandhills (Figure 1a). Barta Brothers Ranch is a University of Nebraska–Lincoln field station in Rock and Brown Counties in the Northeastern Sandhills (42.226381, -99.635297). Barta has an area of approximately 2,000 hectares representative of the Eastern Sandhills with rolling sand dunes covered in cool and warm season grasses, sedges, forbs, planted cedar tree lines, and patches of invading cedar (Figure 1b,1c). The ranch is used by University of Nebraska students, staff, and partners for agricultural, range and wildlife research and demonstrations. I selected study sites using areal imagery in ArcGIS Pro, ground truthed for accuracy. The study sites consist of planted cedar tree lines or stands adjacent to open rangeland. Sites were free of livestock during the data collection period and stock tanks were dry. Supplemental data was also used, for analysis at a scale beyond the study site and sensor extent, from data collected for the Nebraska North American Bat Monitoring Program from 2016-2019, 2021. (Figure 1a; North American Bat Monitoring Program [NABat] [usgs.gov]). The NABat program was implemented in Nebraska to answer baseline bat habitat questions and ensure a continuation of monitoring in Nebraska in accordance with federal standards (Seguin 2019). Collection was done using Titley

Scientific Anabat Express acoustic sensors placed singly, elevated about 2m to clear ground interference. Sensors were left out for 4 nights for data collection. Much of the setting up and taking down of sensors was done using citizen science with volunteers doing much of the work. Species identification was done using Kaleidoscope Pro. Locations were chosen randomly using the generalized random tessellation stratified survey design algorithm across the state (Seguin 2019).

### **Materials and Equipment**

I recorded bat calls using Titley Scientific Anabat Express acoustic sensors with an effective range of 50 meters in a grid of 24 sensors 100 meters apart from one another resulting in a sensor area of 400 meters by 600 meters (Figure 1d). The acoustic sensors were mounted on extendable painter's poles customized to hold the sensors approximately 4 meters off the ground to improve detection. The poles were held up using metal stakes attached to a piece of PVC pipe to ensure sensors are kept in place during the data collection period. I created the sensor grid in ArcGIS Pro, with 24 fishnet points 100 meters away from each other in a 4 x 6 formation. These points were converted into GPX points using ArcGIS Pro tools and transferred to a Garmin GPSMAP 64, these points were used to place acoustic sensors. The sensor grid was placed adjacent to or around the cedar tree feature with sensors in range of cedar habitat or open grassland habitat (Figure 1d). Acoustic sensors were set to night mode turning on 30 minutes before sunset and turning off 30 minutes after sunrise. The data collection period is 4 nights of non-inclement weather to ensure bat activity. Inclement weather, in this study, included nightly temperatures below freezing, high winds Beaufort 5 or above, and precipitation. If any of these events occurred during data collection the sensors were left

up another night to reach the target of 4 collection nights. Calls from all data collection nights at a site were combined for total counts.

### **Bat Identification**

Detected bat calls were stored on an SD card inside of the acoustic sensor as raw nightly acoustic files. These raw files were uploaded into AnalookW (version 4.6c) which separates the raw file into individual bat passes or individual sound sequences. These files are output from AnalookW as ZCA files can be bat passes, insect noise, wind, or any other sound within the range of the acoustic sensor. For species identification the ZCA files are uploaded to Wildlife Acoustics Kaleidoscope Pro (Version 5.4.2). Kaleidoscope uses signal parameter settings selected by the user to identify bat calls. These parameters include frequency range, length of detected pulses, maximum inter syllable gap and minimum number of bat pulses. For this study, frequency range was set to 8-120 kHz, length was set to 2-500 ms, inter-syllable gap was set to 500 ms, and minimum number of pulses was set to 3. Species included for consideration for identification in Kaleidoscope included big brown bat, eastern red bat, hoary bat, silver-haired bat, western small-footed myotis, little brown bat, northern long-eared bat, long legged myotis, evening bat, and tri-colored bat (Table 4). Kaleidoscope identifies characteristics of the sound waves and compares them with their database of confirmed calls to give a species identification. Kaleidoscope categories each ZCA file either as noise, a known bat call or an unidentified bat call. Species IDs are given as a percentage of matched pulses. For example, a bat may fly by a sensor and emit 10 pulses inside of the sensor range, if 8 out of 10 of the pulses match a given species Kaleidoscope will display 80% match for that species. Kaleidoscope will, on some occasions, give a second and third alternative

identification in decreasing percent match, when characteristics of a pulse match more than one species. The highest percentage match was taken and used for data analysis.

Unidentified bat calls were not used in statistical analysis or data interpolation.

Kaleidoscope also outputs presence probability (Britzke et al. 2002), which I collected at the site level.

Data censoring was implemented when a sensor malfunctioned. The only instance of data censoring occurred at site Barta.D for sensor 8, the sensor had blown over from wind and the microphone was bent. No data was recorded from this sensor throughout the collection period.

### **Statistical Analysis and Data Interpolation**

Statistical analysis was completed using R (R Core Team 2021). Excel files for each acoustic sensor exported from Kaleidoscope were loaded, combined, and organized in R at the site/grid level. The data organization allowed for the data to be viewed by species, time or location and sums to be given for specific parameters. Log transformations were utilized to achieve normality in the data. Linear regressions were calculated between the number of bat calls recorded by species and distance from the cedar tree features in kilometers. Distance measurements were collected manually using the measure tool in ArcGIS Pro to the nearest meter. Analysis was conducted on species with  $\geq 30$  calls per site. Presence probability was compared to species that reached the  $\geq 30$  call per site cutoff (Table 1).

Myotis species recorded included northern long-eared bat (*Myotis septentrionalis*), little brown myotis (*Myotis lucifugus*), western small-footed myotis (*Myotis ciliolabrum*), and long-legged myotis (*Myotis volans*). Myotis species call

recordings were rare but when combined at site Barta.G reached the  $\geq 30$  call threshold, so analysis was conducted on this suite of species together.

Supplemental NABat data were compiled and combined by site for available collection years (Table 2). This data was not normally distributed, so log transformations were once again used to achieve normality. Linear regressions were calculated between the number of bat calls of each species and distance to the closest tree at each collection site. NABat data was used in concert with Barta Brothers Ranch data to look at multiple scales and at the spatial extent past the relatively small sensor grid to see the bigger picture for the whole Sandhills region.

Data interpolation and map creation was accomplished in ArcGIS Pro (Version 2.6.0). Data interpolation was done using the kriging tool in the geostatistical wizard in ArcGIS Pro using the simple prediction kriging method. Data interpolation in this case takes the data from the individual points and estimates the number of bat calls in between the points of the grid to provide a continuous estimate of relative abundance throughout the grid. Maps were created using the output data to show usage intensity along the tree line and a usage gradient out into the grassland if it existed. Interpolation maps were made for bat species that met the analysis requirements described above.

## **Results**

### **Bat Presence**

Across all sites at Barta Brothers Ranch the species with recorded calls over number of calls threshold ( $n \geq 30$ ) were big brown bat, eastern red bat, hoary bat, silver-haired bat, and evening bat (Table 1). Presence probability showed a significant p-value

for all but one instance of calls for species/site combinations meeting the threshold of 30 calls. Silver-haired bat had 62 recorded calls at site Barta.G, crossing the  $\geq 30$  call threshold but not showing a significant presence probability p-value (Table 1).

Hoary bat was the most common bat species recorded ranging from 162-664 calls at each site (Table 1). Silver-haired bat was also recorded at each site but with fewer calls, ranging from 3-70 calls. Presence probability p-values only showed a significant p-value at site Barta.E with 70 calls and a p-value of 0.0006 (Table 1).

### **Bat call – Tree Distance Regressions**

Big brown bat at site Barta.G showed a significant negative relationship with distance to cedar windbreak with a distance estimate of -2.41 and a p-value of 0.01 (SE 0.83, adjusted  $R^2$  0.24) (Table 3). Significant negative relationships between distance from the planted cedar line and number of calls also occurred at site Barta.G for eastern red bat (estimate -2.45, SE 0.98, p-value 0.02, adjusted  $R^2$  0.19) and evening bat (estimate -2.26, SE 0.93, p-value 0.02, adjusted  $R^2$  0.18) (Table 3). Combined *Myotis* species also showed a significant negative relationship at site Barta.G (estimate -2.41, SE 0.73, p-value 0.00, adjusted  $R^2$  0.30) (Table 3); for these species, occurrence increased with decreasing distance from cedar features. Hoary bat in all linear regression analyses at Barta showed a mixture of positive and negative distance estimates and non-significant p-values indicating no significant relationship between distance from the planted windbreaks and number of bat calls (Table 3). Silver-haired bat analysis showed similar results with non-significant p-values, indicating no significant relationship between distance from planted windbreaks and the number of bat calls (Table 3).



Regression analyses from statewide bat monitoring (NABat) showed significant negative relationships between distance from tree cover and number of bat calls for almost every species (Table 3). These analyses showed remarkably strong negative estimates and significant p-values indicating call occurrence increasing with decreasing distance from tree features. Western small-footed myotis, northern long-eared bat and tri-colored bat did not show a significant relationship between tree cover and number of bat calls or a high number of calls in general (Table 2, Table 3).

### **Interpolation Maps**

The kriging interpolation map for big brown bat at site Barta.G shows higher numbers along the planted cedar line and lower numbers as you move out into the pasture showing an association with the cedar feature and use (Figure 2a). The same association is shown in the interpolation maps for eastern red bat and evening bat (Figures 2b,2c). The combined Myotis interpolation map also shows this same association with high calls next to the cedar line and lower numbers out into the pasture (Figure 2d).

Kriging interpolation maps for hoary bat and silver-haired bat were similar to each other with pockets of high and low call frequency across the grids with no clear feature associated with higher call numbers (Figures 3a-4d) at the spatial scale of my analysis. The one exception to this is silver-haired bat at Barta.E with high call numbers around the present tree stand (Figure 4a).

### **Discussion**

Insectivorous bats are utilizing planted eastern redcedar tree lines in the Nebraska Sandhills, a finding that is possibly robust across the Great Plains. Big brown bat, eastern

red bat and evening bat have small usage areas closer to tree lines during nightly activity while hoary bat and silver-haired bat utilized larger areas, these results were reflected in linear regressions and interpolation maps. When looking at just the analyses from Barta Brothers Ranch one might conclude that these two species are using grasslands evenly as they are at the limited 400m x 600m grid. Data at a larger spatial scale from statewide bat monitoring shows that both hoary and silver-haired bats have a negative relationship between distance from trees and number of calls at these larger scales. A larger scale interpolation map would likely show a similar utilization pattern for all species but at different distances depending on the species. Combined *Myotis* species have a short-distance utilization pattern similar to that of big brown bat. When combining all *Myotis* species recorded there were just enough calls to meet the threshold of 30 calls for analysis. Statewide data also showed that western small-footed myotis and northern long-eared bat did not have a relationship to trees, but the calls recorded were also exceptionally low compared to other species (Table 2). There currently is insufficient data to support whether *Myotis* species are using cedar structure or of their presence in the Sandhills.

Human planted eastern redcedar windbreaks have spread and created the novel Sandhills ecosystems. Bat utilization of these novel ecosystems creates a dilemma for current cedar management recommendations. The negative impacts of eastern redcedar are well known and degrade native biodiversity and cattle forage quality (Twidwell et al. 2013, Morford et al. 2022). The dilemma arises when the current management recommendation to eradicate cedar trees from grassland ecosystems conflicts with bat management, especially if endangered or threatened bat species are using these new

anthropogenic habitats. Humans planted cedar windbreaks to protect their homes and cattle and consequently changed the composition of the Sandhills grasslands. Now with the realized negative consequences that these afforested and encroached grasslands have on cattle production and biodiversity humans seek to remove trees and restore grasslands. In the years between planting and management bats have started to use this novel ecosystem creating potential roadblocks for land managers. Additionally, cedar woodlands are an emerging alternative state of Great Plains grasslands, and are not simple to restore, as the transition to a wooded state exhibits strong hysteresis.

Big brown bat, silver-haired bat and evening bat are habitat generalists, using open or cluttered areas for foraging (Norberg & Raynor 1987; Jones et al., 2016). Hoary bat and eastern red bat tend to favor more open areas as well as high altitudes which favor their aerial hawking foraging style (Norberg & Raynor 1987; Jones et al., 2016). Planted cedar lines do not allow for a cluttered habitat, a cedar line creates a straight line of edge habitat next to large areas of open habitat. All species recorded at significant amounts have been shown to utilize open habitats.

White-nose syndrome continues to impact bat populations in North America with northern long-eared bat and tri-colored bat being at the highest susceptibility of Nebraska bats (Langwig et al. 2012). White-nose syndrome was first recorded in Nebraska in 2016 and in the far East, highly forested areas of Nebraska (Lorch et al. 2016). These two species showed low call abundance in the Barta Brothers Ranch study and in state-wide bat monitoring. The tri-colored bat had a relatively large call number at only a single site, which lies at the periphery of the Sandhills near Burwell Nebraska in a cedar dominated canyon. Northern long-eared bat numbers were low or zero at all sites for both studies.

Northern long-eared bats are species adapted for highly cluttered areas such as closed canopy deciduous forests (Ratcliffe & Dawson 2003). This would explain why their detection presence was low, it may be that cedar dominated habitats could not support the needs of northern long-eared bats.

Current cedar management efforts may reduce the area of this novel ecosystem. Bats, which are already pressured by disease and habitat decline, could be reduced in abundance by cedar removal. Although support for removing invasive cedar, which often originates from human plantings, is high, support for removing windbreaks is considerably lower, because there are limited alternatives available. Removing easily identifiable seed-bearing females from windbreaks could eliminate propagule sources while preserving critical windbreak services for humans and cattle at the same time preserving foraging and possibly roosting habitat for bats. New windbreaks planted in offset rows 4 deep are robust to the removal of 50% of individuals when identified as female; simple binomial probability suggests that this approach would maintain, on average, 95% of windbreak functionality, having at least 1 tree along the windbreak. Currently there are conflicting activities regarding cedar management, whereby cedars are still distributed and planted by some government agencies while those and other are actively incentivizing their removal (Roberts et al. 2018). Bats will be collaterally affected by management regardless of what management action or policy is chosen. On an increasingly crowded and non-stationary planet, humanity will be forced to make increasingly difficult decisions regarding which ecosystems services to support and which to sacrifice.

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**Tables**

Table (1) Number of bat calls recorded for each species at each bat monitoring location on Barta Brothers Ranch, as well as the presence probability p-values from Kaleidoscope Pro, with significant p-values bolded. For presence probabilities, the lower the p-value the more confident that the given bat species was present at each site with a value of 0.05 being the confidence cutoff.

Bat Calls										
Site	EPTFUS	LASBOR	LASCIN	LASNOC	MYOCIL	MYOLUC	MYOSEP	MYOVOL	NYCHUM	PERSUB
Barta.B			294	6						
Barta.C	3		411	3						
Barta.D	7		664	14						
Barta.E	11		184	70						
Barta.F	7		162	3	1					
Barta.G	111	190	580	62	13	10	3	4	129	16

Presence probability										
Site	EPTFUS	LASBOR	LASCIN	LASNOC	MYOCIL	MYOLUC	MYOSEP	MYOVOL	NYCHUM	PERSUB
Barta.B	1	1	<b>0</b>	1	1	1	1	1	1	1
Barta.C	1	1	<b>0</b>	1	1	1	1	1	1	1
Barta.D	1	1	<b>0</b>	1	1	1	1	1	1	1
Barta.E	1	1	<b>0</b>	<b>0.0006</b>	1	1	1	1	1	1
Barta.F	1	1	<b>0</b>	1	0.1	1	1	1	1	1
Barta.G	<b>0</b>	<b>0</b>	<b>0</b>	1	0.66	1	0.07	<b>0.02</b>	<b>0</b>	1

Table (2) Number of bat calls recorded for each species of bat at each monitoring location for the Nebraska North American Bat Monitoring Program. The table also indicates the distance from trees the acoustic sensor was located and whether the closest tree was an eastern redcedar tree or a deciduous tree.

Location	LASBOR	LASCIN	LASNOC	MYOCIL	EPTFUS	NYCHUM	PERSUB	MYOLUC	MYOSEP	Tree Distance (m)	Tree Type
161821	2	0	0	0	0	0	0	0	0	739	Cedar
161822	0	1	0	0	0	0	0	0	0	475	Cedar
161823	0	1	0	0	0	0	0	0	0	1420	Deciduous
161831	1	1	0	0	0	0	0	0	0	579	Deciduous
161891	1	4	1	2	0	0	0	0	0	1666	Deciduous
193841	470	445	100	0	296	137	32	137	2	0	Cedar
193881	62	79	18	0	80	35	1	8	1	0	Cedar
195421	1	4	1	2	4	1	0	0	0	743	Cedar
195431	0	1	0	1	2	2	0	1	0	131	Deciduous
195432	0	2	0	1	2	2	0	0	0	1392	Deciduous
195491	1	7	0	1	4	0	0	0	0	1276	Deciduous
259411	1	24	7	0	8	0	0	0	0	337	Deciduous
259421	49	439	397	0	421	10	2	5	0	50	Deciduous
259422	19	60	34	0	28	3	0	0	1	0	Cedar
259431	2	77	31	0	92	0	0	1	0	341	Deciduous
259432	2	71	46	0	166	2	0	1	0	57	Deciduous
264211	10	28	7	6	10	7	2	0	0	143	Cedar
264221	30	88	9	46	20	12	3	0	0	0	Cedar
270611	8	37	19	0	50	3	0	3	0	0	Deciduous
270641	3	32	18	0	33	9	1	1	0	0	Deciduous

Table (3) Linear regression outputs from data collected for the Nebraska North American Bat Monitoring Program as well as from Barta Brothers Ranch. Regressions here were conducted by comparing the number of calls for each species and distance from the closest tree in km at all sites. Bolded lines indicate species with significant estimates.

Location	Species	Intercept Estimate	Intercept SE	Intercept p-value	Distance Estimate	Distance SE	Distance p-value	Adjusted R <sup>2</sup>
<b>Barta.G</b>	<b>Myotis</b>	<b>1.08</b>	<b>0.22</b>	<b>0.00</b>	<b>-2.41</b>	<b>0.73</b>	<b>0.00</b>	<b>0.30</b>
<b>Barta.G</b>	<b>EPTFUS</b>	<b>2.04</b>	<b>0.25</b>	<b>0.00</b>	<b>-2.41</b>	<b>0.83</b>	<b>0.01</b>	<b>0.24</b>
Barta.G	LASCIN	2.94	0.31	0.00	-0.61	1.01	0.55	-0.03
<b>Barta.G</b>	<b>LASBOR</b>	<b>2.44</b>	<b>0.30</b>	<b>0.00</b>	<b>-2.45</b>	<b>0.98</b>	<b>0.02</b>	<b>0.19</b>
Barta.G	LASNOC	1.25	0.21	0.00	-0.50	0.71	0.49	-0.02
<b>Barta.G</b>	<b>NYCHUM</b>	<b>2.10</b>	<b>0.28</b>	<b>0.00</b>	<b>-2.26</b>	<b>0.93</b>	<b>0.02</b>	<b>0.18</b>
Barta.F	LASCIN	1.86	0.36	0.00	-0.28	1.98	0.89	-0.04
Barta.E	LASCIN	1.70	0.41	0.00	-1.22	1.37	0.38	-0.01
Barta.E	LASNOC	0.66	0.33	0.06	-1.73	1.12	0.14	0.06
Barta.D	LASCIN	2.26	0.65	0.00	0.50	2.74	0.86	-0.05
Barta.C	LASCIN	1.94	0.49	0.00	-1.45	2.58	0.58	-0.03
Barta.B	LASCIN	1.21	0.42	0.01	1.78	1.40	0.22	0.03
<b>Statewide</b>	<b>ALL</b>	<b>4.87</b>	<b>0.51</b>	<b>0.00</b>	<b>-2.78</b>	<b>0.71</b>	<b>0.00</b>	<b>0.43</b>
<b>Statewide</b>	<b>EPTFUS</b>	<b>3.67</b>	<b>0.44</b>	<b>0.00</b>	<b>-2.55</b>	<b>0.61</b>	<b>0.00</b>	<b>0.46</b>
<b>Statewide</b>	<b>LASBOR</b>	<b>2.51</b>	<b>0.41</b>	<b>0.00</b>	<b>-1.80</b>	<b>0.57</b>	<b>0.01</b>	<b>0.32</b>
<b>Statewide</b>	<b>LASCIN</b>	<b>3.86</b>	<b>0.43</b>	<b>0.00</b>	<b>-2.20</b>	<b>0.60</b>	<b>0.00</b>	<b>0.40</b>
<b>Statewide</b>	<b>LASNOC</b>	<b>2.98</b>	<b>0.41</b>	<b>0.00</b>	<b>-2.28</b>	<b>0.57</b>	<b>0.00</b>	<b>0.44</b>
Statewide	MYOCIL	0.49	0.29	0.11	0.03	0.40	0.95	-0.06
<b>Statewide</b>	<b>MYOLUC</b>	<b>1.10</b>	<b>0.33</b>	<b>0.00</b>	<b>-0.96</b>	<b>0.46</b>	<b>0.05</b>	<b>0.15</b>
Statewide	MYOSEP	0.22	0.09	0.03	-0.20	0.12	0.13	0.07
<b>Statewide</b>	<b>NYCHUM</b>	<b>1.93</b>	<b>0.34</b>	<b>0.00</b>	<b>-1.49</b>	<b>0.47</b>	<b>0.01</b>	<b>0.32</b>
Statewide	PERSUB	0.72	0.24	0.01	-0.63	0.33	0.07	0.12

Table (4) A list of bats that were sampled at Barta Brothers Ranch with corresponding scientific names and species codes.

common name	Scientific name	Species code
big brown bat	<i>Eptesicus fuscus</i>	EPTFUS
eastern red bat	<i>Lasiurus borealis</i>	LASBOR
hoary bat	<i>Lasiurus cinereus</i>	LASCIN
silver-haired bat	<i>Lasionycteris noctivagans</i>	LASNOC
western small footed myotis	<i>Myotis ciliolabrum</i>	MYOCIL
little brown bat	<i>Myotis lucifugus</i>	MYOLUC
northern long-eared bat	<i>Myotis septentrionalis</i>	MYOSEP
long-legged myotis	<i>Myotis volans</i>	MYOVOL
evening bat	<i>Nycticeius humeralis</i>	NYCHUM
tri-colored bat	<i>Perimyotis subflavus</i>	PERSUB

# Figures

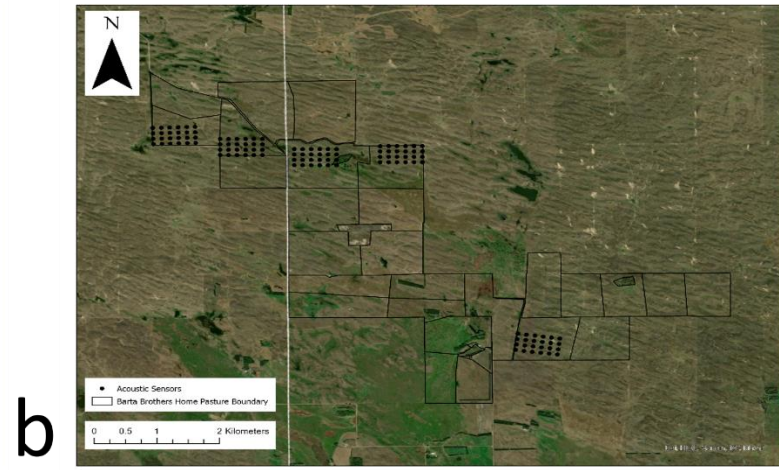
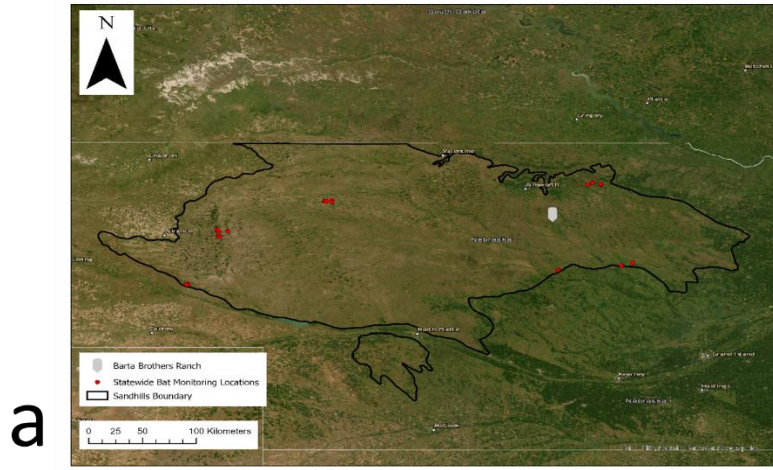


Figure 1(a) Satellite imagery showing the boundary of the Nebraska Sandhills with markers denoting the location of Barta Brothers Ranch and statewide bat monitoring locations.

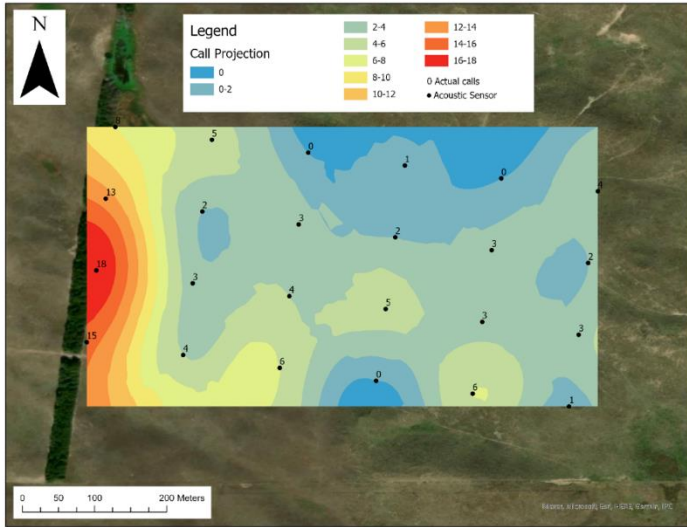
(b) Satellite imagery showing the boundary of Barta Brothers Ranch home pasture with markers showing the locations of acoustic sensor grids.

(c) Satellite imagery showing an example of planted eastern redcedar windbreak complexes at Barta Brothers Ranch in the Nebraska Sandhills.

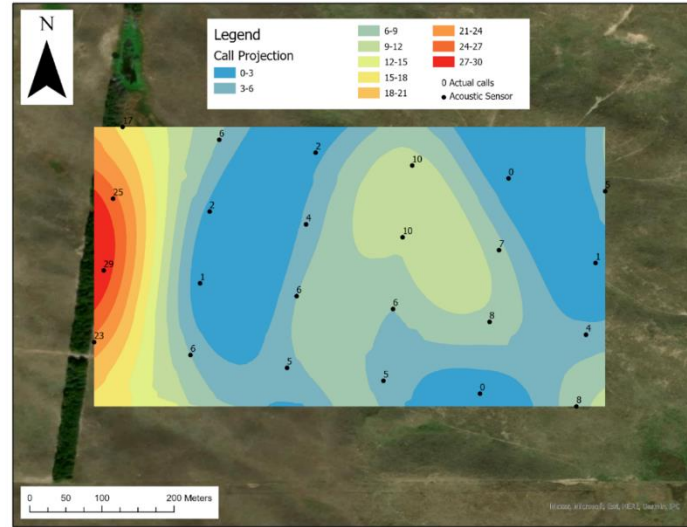
(d) Satellite imagery showing an example of how acoustic sensors were set up in relation to planted eastern redcedar windbreaks. This example is the location for Barta.B and Barta.G at Barta Brothers Ranch.



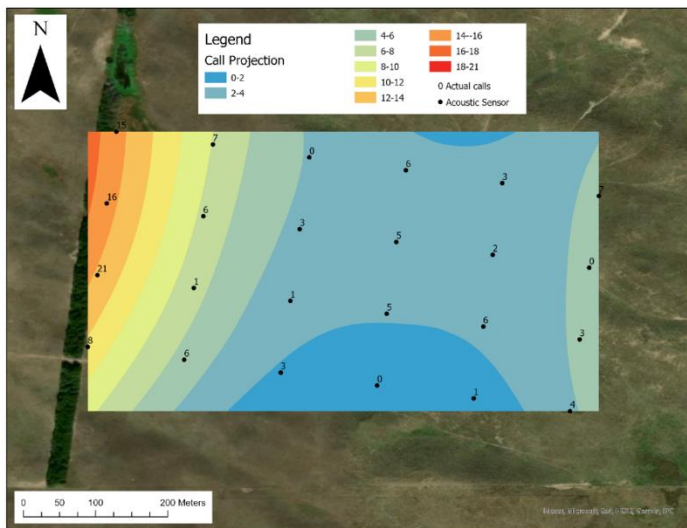
a



b



c



d

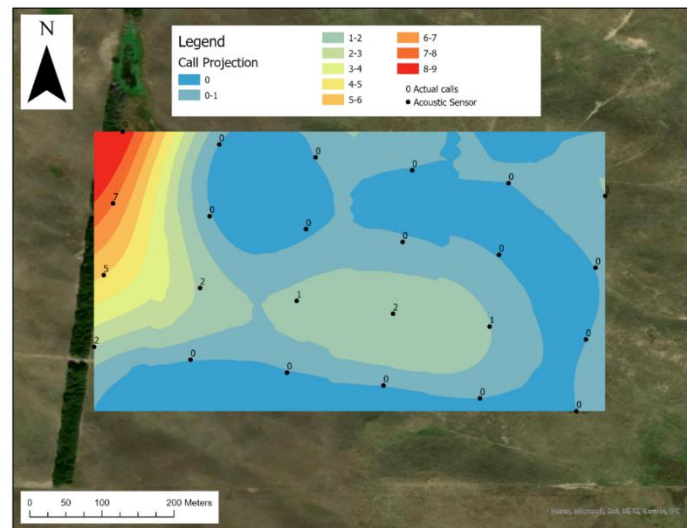


Figure 2(a) Interpolation map showing the kriging projection for big brown bat (*Eptesicus fuscus*) at location Barta.G on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

(b) Interpolation map showing the kriging projection for eastern red bat (*Lasiurus borealis*) at location Barta.G on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

(c) Interpolation map showing the kriging projection for evening bat (*Nycticeius humeralis*) at location Barta.G on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

(d) Interpolation map showing the kriging projection for combined myotis bat species including: northern long-eared bat (*Myotis septentrionalis*), little brown myotis (*Myotis lucifugus*), western small-footed myotis (*Myotis ciliolabrum*), and long-legged myotis (*Myotis volans*) at location Barta.G on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

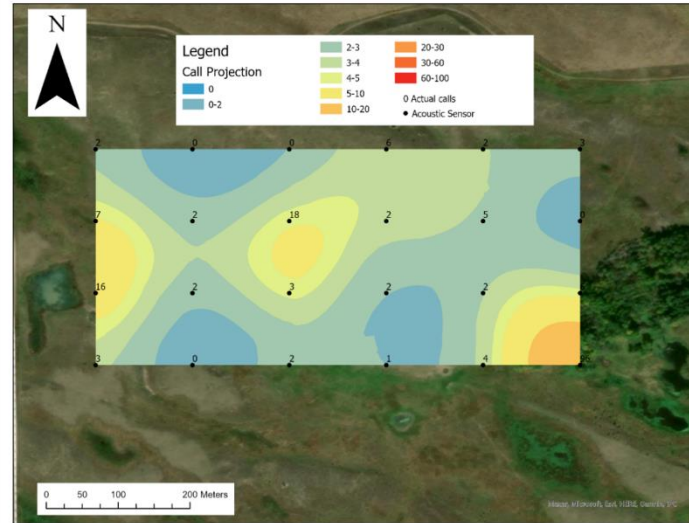
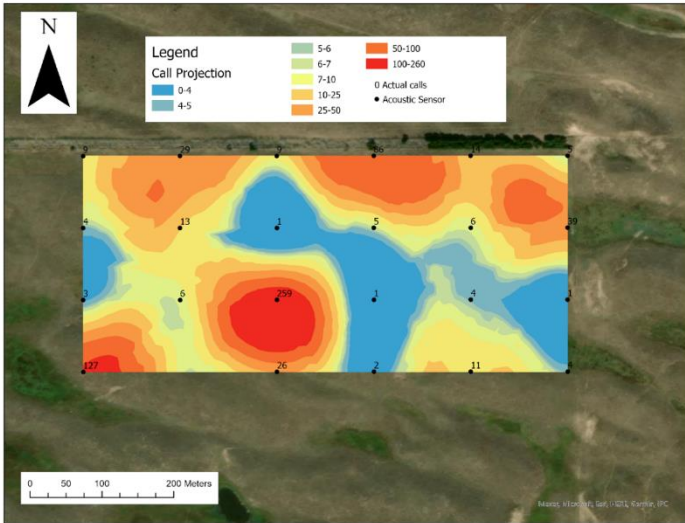
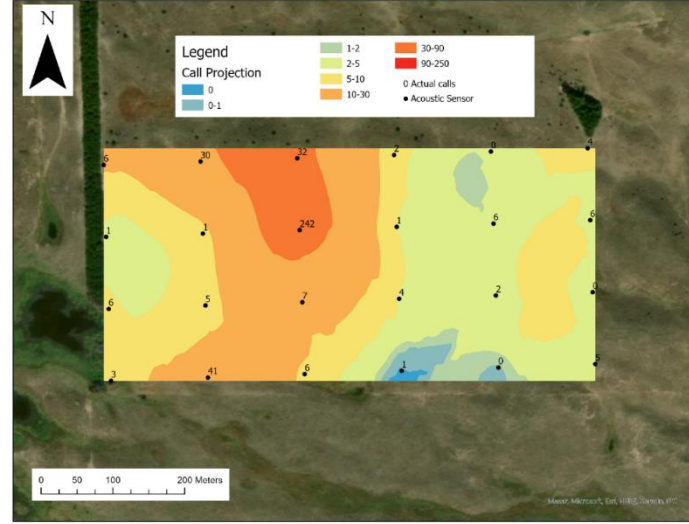
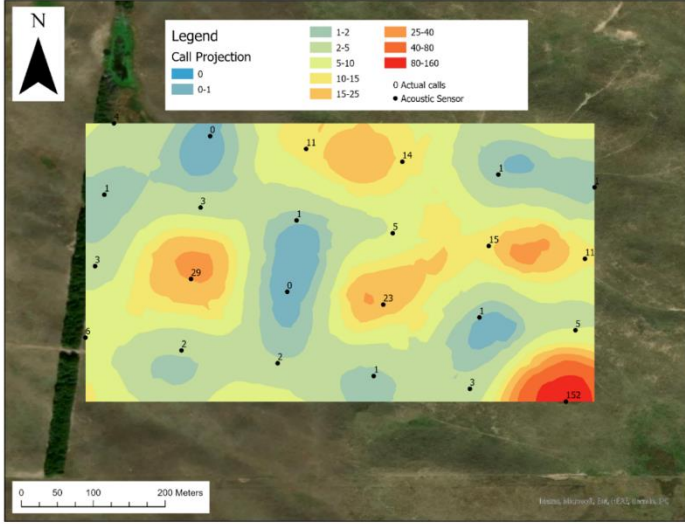
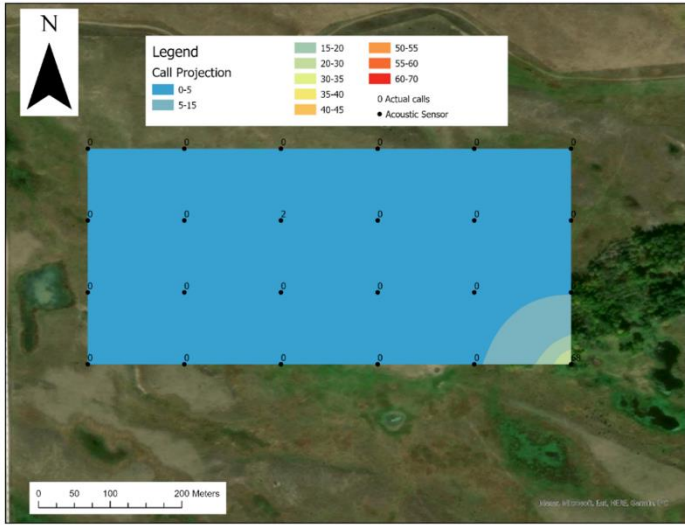


Figure 3(a) Interpolation map showing the kriging projection for hoary bat (*Lasiurus cinereus*) at location Barta.B on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

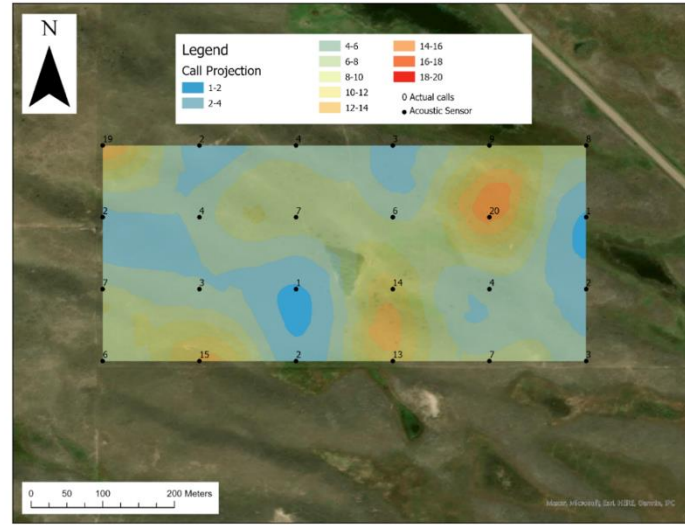
(b) Interpolation map showing the kriging projection for hoary bat (*Lasiurus cinereus*) at location Barta.C on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

(c) Interpolation map showing the kriging projection for hoary bat (*Lasiurus cinereus*) at location Barta.D on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

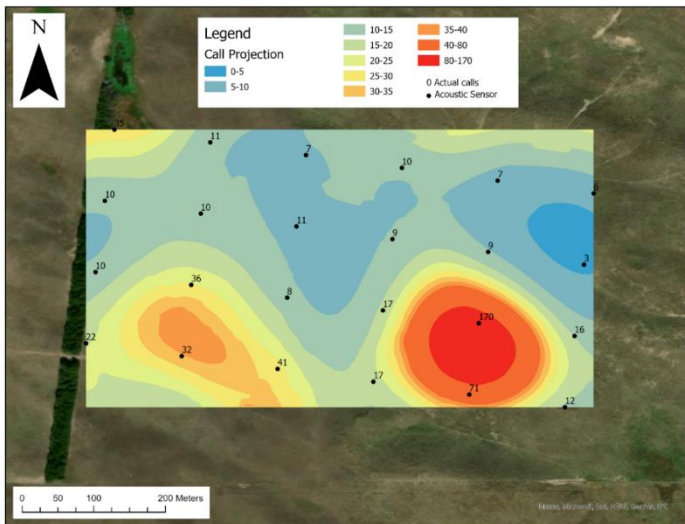
(d) Interpolation map showing the kriging projection for hoary bat (*Lasiurus cinereus*) at location Barta.E on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.



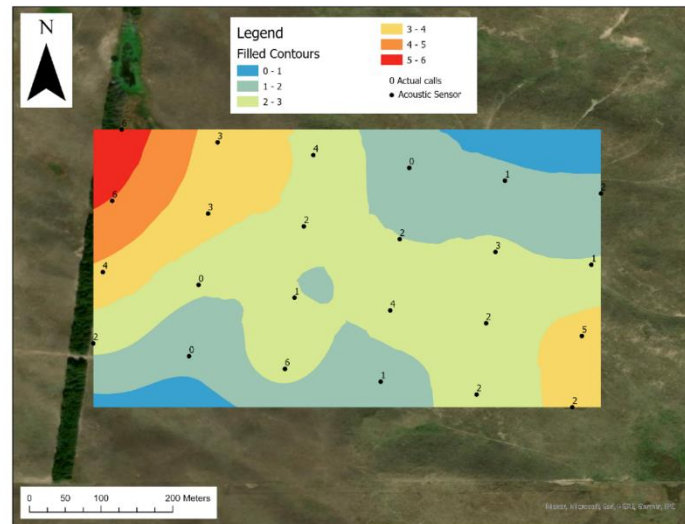
a



b



c



d

Figure 4(a) Interpolation map showing the kriging projection for silver-haired bat (*Lasionycteris noctivagans*) at location Barta.E on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

(b) Interpolation map showing the kriging projection for hoary bat (*Lasiurus cinereus*) at location Barta.F on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid. The kriging layer was made transparent to show the tree stand at the center of the grid.

(c) Interpolation map showing the kriging projection for hoary bat (*Lasiurus cinereus*) at location Barta.G on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

(d) Interpolation map showing the kriging projection for silver-haired bat (*Lasionycteris noctivagans*) at location Barta.G on Barta Brothers Ranch. Markers indicate the location of an acoustic sensor and numbers indicate the actual recorded number of recorded calls at each sensor in the grid.

## CHAPTER 3 – MULTI MODEL INFERENCE OF BAT RICHNESS AND CALL COUNT IN THE NEBRASKA SANDHILLS

### Introduction

Thirteen insectivorous bats occur in Nebraska, including the federally endangered northern long-eared bat (*Myotis septentrionalis*). Bats in Nebraska provide important ecosystem services from their consumption of insects. The US farming industry saves an estimated 3.7 billion to 53 billion US dollars per year from bats consumption of pest insects (Boyles et al. 2011), though the assumptions about bat activity in crop fields made to calculate these numbers have been questioned (Fill et al. 2022). Insect consumption by bats also has indirect effects, such as minimizing disease by vectors such as mosquitos. A single bat has the ability to consume 1000 mosquito-sized insects an hour, though bats prefer to target higher calorie insects such as moths (Ducummon 2000).

Bats inhabiting higher latitudes compensate for cooler winter temperatures and insect numbers by either migrating South to warmer habitats or by spending their winter in hibernacula. Migration is usually undertaken by tree roosting species such as the hoary bat, which is common in Nebraska during the summer months. Big brown bat, another common species, can spend all year in Nebraska, mostly hibernating during the colder months, although there can be some activity in winter (White 2014). Depending on the species and the weather, bats are generally present and active in Nebraska roughly from March through October (Geluso 2004).

North America has seen bat population declines, presumably due to habitat degradation, white-nose syndrome, and mortality from wind turbines. White-nose

syndrome is a fungal infection that disrupts bat hibernation, causing them to burn stores of fat which lowers survival rate (Reynolds et al. 2016). For example, northern-long eared bats are extremely susceptible to White-nose syndrome spreading through wintering colonies and its population has plummeted and, in some cases, led to its extirpation (Frick et al. 2015, Warnecke et al. 2012). Migratory bats also face dangers from wind turbines. Bat mortality around wind turbines is high especially during bat migrations from their warmer winter habitats to the South (Arnett et al. 2008, Cryan et al. 2014). Forest composition has also changed from pre-European settlement (Nemec et al. 2011), causing increased bat habitat strain. Forest change has been in part due to the introduction and spread of species including russian olive (*Elaeagnus angustifolia*), autumn olive (*Elaeagnus umbellate*), and eastern redcedar (*Juniperus virginiana*), which cause mid-story clutter hindering bat movement for forest adapted species.

The Nebraska Sandhills is a semi-arid region in Nebraska comprised of rolling grass-stabilized sand dunes, vegetated with mixed grass prairie. The Sandhills are currently going through a transition from grassland to cedar dominated woodland due to woody encroachment and planting of eastern redcedar (Scholtz & Twidwell 2022, Fogarty et al. 2022a, 2022b). Eastern redcedar is a popular tree used for windbreaks given its thick canopy and ability to survive in a variety of conditions (Van Haverbeke 1976). These same traits have also had negative impacts on grasslands throughout Nebraska and the Great Plains. Eastern redcedar canopy is 99% closed causing other plant species to decline (Briggs et al. 2002). Around 40,000 acres of land in Nebraska is lost to woody encroachment each year, lowering herbaceous cover and species diversity. This makes this cedar dominated woodland a largely undesired alternative stable state for



Sandhills grasslands (Roberts and Allen 2023). Cedar dominated Sandhills is a novel Sandhills ecosystem that is not considered ‘typical’ bat habitat because of its closed and cluttered canopy (Sleep & Brigham 2003); areas of the Sandhills without trees are also not considered typical habitat, because of the lack of deciduous trees, or other structure. The areas above and around cedar features could act as structural bat habitat and an extension of bat habitat into the Sandhills. There are still large areas of core grasslands in the Sandhills that could act as barriers for bat travel, though these areas are shrinking.

In order to determine how bats use the Sandhills ecoregion and forested patches therein, I examined bat species richness and number of calls using acoustic sensors. Initial analysis in chapter 2 indicated that in the interior of the Sandhills bats were taking longer to appear during the summer season in regard to number of species and calls compared to the periphery of the Sandhills (Figure 2, 3), but that bats did use isolated cedar windbreaks and patches. The objective of this study is to determine which features in the Sandhills are driving higher species richness and call numbers. I hypothesize that bats will take longer to start utilizing the middle of the Sandhills than the edge of the Sandhills, and that proximity of suitable habitat will affect that relationship.

## **Methods**

### **Study Area and Site Selection**

I conducted this study at Barta Brothers Ranch in the Nebraska Sandhills (Figure 1a). Barta Brothers Ranch is a University of Nebraska– Lincoln field station in Rock and Brown Counties in the Northeastern Sandhills (42.226381, -99.635297). Barta has an area of approximately 2,000 hectares representative of the Eastern Sandhills with rolling sand dunes covered in cool and warm season grasses, sedges, forbs, planted cedar tree

lines (i.e., afforestation), and patches of invading cedar (i.e., woody plant encroachment) (Figure 1b,1c). The ranch is used by University of Nebraska students, staff, and partners for agricultural, range and wildlife research and demonstrations. I selected study sites using areal imagery in ArcGIS Pro, ground truthed for accuracy. The study sites consist of planted cedar tree lines or stands adjacent to open rangeland. Sites were free of livestock during the data collection period and stock tanks were dry. Supplemental data for the entire Sandhills was also used and provided by the Nebraska North American Bat Monitoring Program from 2016-2019, 2021. (Figure 1a; North American Bat Monitoring Program [NABat] [usgs.gov]). The NABat program was implemented in Nebraska to answer baseline bat habitat questions and ensure a continuation of monitoring in Nebraska in accordance with federal standards (Seguin 2019). Collection was done using Titley Scientific Anabat Express acoustic sensors placed singly, elevated about 2m to clear ground interference. Sensors were left out for 4 nights for data collection. Much of the setting up and taking down of sensors was done using citizen science with volunteers doing much of the work. Species identification was done using Kaleidoscope Pro. Locations were chosen randomly using the generalized random tessellation stratified survey design algorithm across the state (Seguin 2019).

### **Materials and Equipment**

I recorded bat calls using Titley Scientific Anabat Express acoustic sensors with an effective range of 50 meters in a grid of 24 sensors 100 meters apart from one another resulting in a sensor area of 400 meters by 600 meters (Figure 1d). The acoustic sensors were mounted on extendable painter's poles customized to hold the sensors approximately 4 meters off the ground to improve detection. The poles were held up

using metal stakes attached to a piece of PVC pipe to ensure sensors are kept in place during the data collection period. I created the sensor grid in ArcGIS Pro, with 24 fishnet points 100 meters away from each other in a 4 x 6 formation. These points were converted into GPX points using ArcGIS Pro tools and transferred to a Garmin GPSMAP 64, these points were used to place acoustic sensors. The sensor grid was placed adjacent to or around the cedar tree feature with sensors in range of cedar habitat or open grassland habitat (Figure 1d). Acoustic sensors were set to night mode turning on 30 minutes before sunset and turning off 30 minutes after sunrise. The data collection period is 4 nights of non-inclement weather to ensure bat activity. Inclement weather, in this study, included nightly temperatures below freezing, high winds Beaufort 5 or above, and precipitation. If any of these events occurred during data collection the sensors were left up another night to reach the target of four collection nights. Calls from all data collection nights at a site were combined for total counts. Grid Barta.A was done as an initial test grid and sensors were spaced 75 meters from each other.

### **Bat Identification**

Detected bat calls were stored on an SD card inside of the acoustic sensor as raw nightly acoustic files. These raw files were uploaded into AnalookW (version 4.6c) which separates the raw file into individual bat passes or individual sound sequences. These files are output from AnalookW as ZCA files can be bat passes, insect noise, wind, or any other sound within the range of the acoustic sensor. For species identification the ZCA files are uploaded to Wildlife Acoustics Kaleidoscope Pro (Version 5.4.2). Kaleidoscope uses signal parameter settings selected by the user to identify bat calls. These parameters include frequency range, length of detected pulses, maximum inter syllable gap and

minimum number of bat pulses. For this study, frequency range was set to 8-120 kHz, length was set to 2-500 ms, inter-syllable gap was set to 500 ms, and minimum number of pulses was set to 3. Species included for consideration for identification in Kaleidoscope included Species included for consideration for identification in Kaleidoscope included big brown bat, eastern red bat, hoary bat, silver-haired bat, western small-footed myotis, little brown bat, northern long-eared bat, long legged myotis, evening bat, and tri-colored bat (Table 6). Kaleidoscope identifies characteristics of the sound waves and compares them with their database of confirmed calls to give a species identification. Kaleidoscope categories each ZCA file either as noise, a known bat call or an unidentified bat call. Species IDs are given as a percentage of matched pulses. For example, a bat may fly by a sensor and emit 10 pulses inside of the sensor range, if 8 out of 10 of the pulses match a given species Kaleidoscope will display 80% match for that species. Kaleidoscope will, on some occasions, give a second and third alternative identification in decreasing percent match, when characteristics of a pulse match more than one species. The highest percentage match was taken and used for data analysis. Unidentified bat calls were not used in statistical analysis or data interpolation. Kaleidoscope also outputs presence probability (Britzke et al. 2002), which I collected at the site level.

Data censoring occurred when a sensor malfunctioned. The only instance of data censoring occurred at site Barta.D for sensor 8, the sensor had blown over from wind and the microphone was bent. No data was recorded from this sensor throughout the collection period.

## Statistical Analysis

Statistical analysis was completed using R (R Core Team 2021), using the tidyverse, lme4 and MuMIn packages. Grid data from Barta Brother Ranch was combined at the site level resulting in seven points. Data from statewide monitoring were separated to each individual monitoring session between 2016 and 2021 resulting in 54 points. Statewide data and Barta data were organized by richness and bat call counts and combined into one data set of 61 points. A collection order was assigned to each point chronologically, by date, from May 31st – September 11th. Points collected on the same calendar day were assigned the same collection order number. Distances to features were measured in ArcGIS Pro using the ruler tool with satellite imagery to the nearest meter. ArcGIS uses Maxar imagery at 0.3 - 0.5 meter resolution across the United States. Different tree types, mainly deciduous and cedar, were differentiated manually. The feature distances measured to were distance to the edge of the Sandhills, distance to the closest cedar tree, distance to the closest deciduous tree, distance to the closest tree patch (group of trees  $\geq 5$ ), and distance to the closest tree of any species (Table 1). Distance to the edge of the Sandhills was found using the measure tool and measuring the distance to a Sandhills shapefile in ArcGIS, the shapefile of the Sandhills is from the Nebraska Legacy Project (Schneider 2005). 19 plausible linear regression models were developed to explain both species richness and bat call count as the dependent variables.

The nineteen models included a global model (GLOBAL), which included all variables, a null model (null), which includes no variables, a time model (TIME) which predicts bat activity intensity or richness as a function of time from May 31st – September 11th, and eight models run with and without time. These models included

combinations of distance to the edge of the Sandhills, distance to the closest cedar tree, distance to the closest deciduous tree, distance to the closest tree patch with  $\geq 5$  trees of any kind, and distance to the closest tree of any kind as variables.

The model TREES predicts bat activity intensity or richness as a function of the distance to cedar and deciduous trees. If supported, this model suggests that proximity to trees best predicts the expansion of bats into the core of the Sandhills. Richness and bat activity would be best predicted by the presence and proximity of deciduous or cedar trees. The model TREES.TIME incorporates time into the tree model, and if supported would suggest the importance of trees for the movement of bats into the Sandhills, but further suggest that time through the summer also accounts for the patterns observed.

The model EDGE predicts bat activity intensity or richness as a function of the distance to the edge of the Sandhills. If supported, this model suggests that the Sandhills are acting as a barrier for bats, hindering their expansion into the interior. Richness and bat activity would best be predicted by proximity to the periphery of the Sandhills. The model EDGE.TIME incorporates time into the edge model and if supported would suggest that the Sandhills act as a hindrance instead of a barrier suggesting that with time through the summer bats can move into the interior of the Sandhills.

The model HARD predicts bat activity intensity or richness as a function of the distance to deciduous trees (hardwoods). If supported, this model suggests that proximity to deciduous trees best predicts the expansion of bats into the Sandhills. Richness and bat activity would be best predicted by the presence and proximity of deciduous trees. The model HARD.TIME incorporates time into the deciduous model and if supported would

suggest the importance of deciduous trees for the movement of bats into the Sandhills but further suggest that time through the summer also accounts for the patterns observed.

The model PATCH predicts bat activity or richness as a function of the distance to tree patches, in this case homogeneous or mixed species tree patches with >5 trees present. If supported, this model suggests that proximity to tree patches best predicts the expansion of bats into the Sandhills rather than individual trees. The model PATCH.TIME incorporates time into the tree patch model and if supported would suggest the importance of tree patches for the movement of bats into the Sandhills but further suggest that time through the summer also accounts for the patterns observed.

The model CEDAR predicts bat activity intensity or richness as a function of the distance to eastern redcedar trees. If supported, this model suggests that proximity to redcedar best predicts the expansion of bats into the Sandhills. Richness and bat activity would be best predicted by the presence and proximity of redcedar trees. The model CEDAR.TIME incorporates time into the redcedar model and if supported would suggest the importance of redcedar trees for the movement of bats into the Sandhills but further suggest that time through the summer also accounts for the patterns observed.

The model CloseTREE predicts bat activity intensity or richness as a function of the distance to trees of any kind. If supported, this model suggests that proximity to trees, regardless of species, best predicts the expansion of bats into the Sandhills. Richness and bat activity would be best predicted by the presence and proximity to any tree. The model CloseTREE.TIME incorporates time into the closest tree model and if supported would suggest the importance of trees regardless of species for the movement of bats into the

Sandhills but further suggests that time through the summer also accounts for the patterns observed.

The model EDGE.CEDAR predicts bat activity intensity or richness as a function of the distance to eastern redcedar trees and distance to the edge of the Sandhills. If supported, this model suggests that bats are staying near the periphery of the Sandhills and utilizing redcedar trees there. Richness and bat activity would best be predicted by the presence of redcedar trees and proximity to the edge of the Sandhills. The model EDGE.CEDAR.TIME incorporates distance into the edge cedar model and if supported would suggest the importance of redcedar trees and proximity to the edge of the Sandhills but further suggests that time through the summer also accounts for the patterns observed.

The model EDGE.CloseTREE predicts bat activity intensity or richness as a function of the distance to trees of any kind and distance to the edge of the Sandhills. If supported, this model suggests that bats are staying near the periphery of the Sandhills and utilizing all trees there. Richness and bat activity would best be predicted by the presence of trees, regardless of species, and proximity to the edge of the Sandhills. The model EDGE.CloseTREE.TIME incorporates distance into the edge close tree model and if supported would suggest the importance of trees of any kind and proximity to the edge of the Sandhills but further suggests that time through the summer also accounts for the patterns observed.

Multi model inferencing was used to assess the support of each model. For each model the AICc,  $\Delta$ AICc and model weight was calculated and the models with a  $\Delta$ AICc of less than 4 were included in the confidence set. Both richness and call count had more than one model with an  $\Delta$ AICc of less than 4 so model averaging was utilized. Averaging



was done using weights of all accepted models giving higher priority to models with higher weights. A confidence interval was calculated for all variables inside of the averaged models.

## Results

When analyzing which model was best describes richness there were two models that had a  $\Delta\text{AICc}$  less than 4, TREES.TIME and HARD.TIME (Table 2). TREES.TIME was the strongest model with an AICc of 247.81 (K 3,  $\Delta\text{AICc}$  0, w 0.67). HARD.TIME was the second strongest model with an AICc of 251.06 (K 2,  $\Delta\text{AICc}$  3.25, w 0.13). The averaged model for richness showed that as distance from deciduous trees increases that bat richness decreases (Estimate -1.76, SE 0.57, lower CI -2.88, upper CI -0.63), as distance to cedar decreases richness increases (Estimate -0.64, SE 0.27, lower CI -1.19, upper CI -0.09, as time goes on in the summer richness increases (Estimate 0.09, SE 0.03, lower CI 0.03, upper CI 0.15) (Table 3).

When analyzing which model best describes bat call count there were four models that had a  $\Delta\text{AICc}$  less than 4, TREES.TIME, TREES, HARD.TIME, and CloseTREE.TIME (Table 4). TREES.TIME was the strongest model with an AICc of 124.48 (K 3,  $\Delta\text{AICc}$  0, w 0.47). TREES was the second best model with an AICc of 125.87 (K 2,  $\Delta\text{AICc}$  1.39, w 0.23). HARD.TIME was the third strongest with an AICc 127.63 (K 2,  $\Delta\text{AICc}$  3.15, w 0.10). CloseTREE.TIME was the fourth strongest with an AICc of 128.37 (K 2,  $\Delta\text{AICc}$  3.89, w 0.07). The averaged model for bat call counts showed that as distance from deciduous trees increases bat calls decrease (Estimate -0.76, SE 0.20, lower CI -1.16, upper CI -0.35), as distance to cedar decrease calls increase (Estimate -0.24, SE 0.10, lower CI -0.45, upper CI -0.04), earlier the collection time in

the summer calls increase (Estimate -0.02, SE 0.01, lower CI -0.04, upper CI 0.00), as distance from the closest tree decreases calls increase (Estimate -1.10, SE 0.16, lower CI -1.43, upper CI -0.77) (Table 5).

## **Discussion**

When determining richness from the measured variables TREES.TIME and HARD.TIME were the models with the highest weights. Distance to deciduous trees and time were in both of these models meaning that these are the strongest variables when testing for richness, followed by distance to cedar.

When determining bat call counts from the measured variables four models had acceptable  $\Delta AICc$  values, these included TREES, with and without time as a variable, HARD.TIME, and CloseTREE.TIME. Distance to the closest tree was the best indicator of call count. Distance to deciduous, distance to cedar and time were also good indicators. When examining the standard error and confidence intervals for distance to cedar and time in the averaged model, the estimates are close to zero, indicating that in the real world these variables may not affect bat activity.

When comparing the estimates of distance to deciduous or cedar to distance to tree patch for count analysis it appears that individual trees are utilized by bats and that patches of trees are not necessary. When comparing distances to deciduous, cedar and the closest tree for bat call counts it appears that the closest tree is the best indicator followed by deciduous then by cedar. This suggests that tree type does not play a factor in bat use, but a preference for deciduous trees may exist. When comparing estimates for bat richness these results are similar though the closest tree variable is not included. This

suggests that more species may have a preference for the deciduous trees but bat activity is driven by any tree species. These results provide a pattern that as you move from tree structure bat activity and bat richness decrease. These same patterns were observed in chapter one of this thesis. Literature has also supported these trends in other parts of the world showing that bat activity decreasing and species diversity decreasing as you move away from forest structure into grasslands (Treitler et al. 2016). As the Sandhills become more woody dominated available habitat for bats could also increase reducing the stress already applied to bats around North America.

Time was a variable included in both the richness and count analyses. Richness analysis saw an increase in richness as time in the summer progressed. Count analysis saw a slight decrease in counts as time in the summer progressed though it is very near zero and the upper confidence interval is zero. Looking at these analyses together, the inference could be made that species arrival in the state from migration and species emergence from hibernation happens at different times. This could be due to insect availability or temperature requirements for each individual species or species willingness to enter the Sandhills. Comparing these Sandhill analyses with the same test for other areas in the state would help show a wider scope of when bats are active in the state and how and if this is different for unique landscapes in Nebraska. In other parts of the state Big brown bat and eastern red bat have recorded being active during winter months (White 2014). It is possible that hibernating species are spending winters in the Sandhills or near the periphery, in turn entering the Sandhills more quickly than migrating species leading to a richness increase through the summer.

The number of counts seemed to have stayed stable or slightly decreased through the summer, this could be due to a drop off call counts towards the end of the summer as bats move back south for the colder months or reentering hibernacula. There does not seem to be an increase in activity that would logically be seen from the introduction of volant young starting to feed on their own, whether from resource availability or movement bat activity stays about the same through time in Sandhills. Lactating females have been caught from the end of May to the end of July and volant young have been caught from July to September in Nebraska depending on species (Geluso 2004). These dates overlap on a species basis and often stretch multiple months for a single species. This makes the attribution of activity change to new bats being born difficult. Year long sensing and capture would answer many additional questions about when bats arrive and use the Sandhills so further research on this topic is advised.

Distance to the edge of the Sandhills was not in any of the accepted models and EDGE.TIME showed a weight of zero for both richness and count analyses. This disproves my hypothesis and shows that distance to trees is a much stronger indicator of bat activity than how deep in the Sandhills habitat is located. Bats can fly quickly and long distances, this shows that the areas tested are within travel corridors accessible to bats. Core areas of grassland in the Sandhills are shrinking and species composition is changing, reflecting a rapidly changing landscape.

Incentives in Nebraska and the Sandhills still exist to control the spread of woody plants due to their negative side effects to forage quality and plant species diversity. Bats in Nebraska and North America have been in decline and any management of woody structure that bats are shown to have higher activity and richness near would put more

pressure on an already pressured order. Land Managers and government entities will have to make decisions on what species and ecosystem services to support when their presence contradicts the other.

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## Tables

Table (1) Variables for bat sensor locations in the Sandhills. Distances are reported in kilometers.

Location	Richness	Count	Distance to Edge of Sandhills	Distance to Cedar	Distance to Deciduous	Distance to Tree Patch	Distance to Closest Tree	Collection Order (Time)
161821	1	2	28.24	0.74	1.26	0.74	0.74	26
161822	1	1	27.03	0.48	2.16	0.48	0.48	9
161823	1	1	34.13	5	1.42	3.42	1.42	26
161831	2	2	27.93	3.14	0.58	0.58	0.58	26
161891	1	1	26.82	3.28	1.67	1.67	1.67	9
161891	2	3	26.82	3.28	1.67	1.67	1.67	13
161891	3	4	26.82	3.28	1.67	1.67	1.67	26
193841	8	227	0.82	0	0	0	0	18
193841	8	953	0.82	0	0	0	0	15
193841	6	29	0.82	0	0	0	0	16
193841	7	108	0.82	0	0	0	0	19
193841	7	302	0.82	0	0	0	0	27
193881	6	145	4.29	0	0	0	0	18
193881	5	85	4.29	0	0	0	0	15
193881	7	47	4.29	0	0	0	0	16
193881	5	7	4.29	0	0	0	0	27
195421	1	1	54.02	0.74	1.07	0.74	0.74	10
195421	1	1	54.02	0.74	1.07	0.74	0.74	14
195421	6	11	54.02	0.74	1.07	0.74	0.74	21
195431	5	7	55.3	0.19	0.13	0.19	0.13	21
195432	1	1	52.09	2.93	1.39	2.31	1.39	14

195432	4	6	52.09	2.93	1.39	2.31	1.39	21
195491	1	2	51.26	1.5	1.28	1.39	1.28	10
195491	1	1	51.26	1.5	1.28	1.39	1.28	14
195491	4	10	51.26	1.5	1.28	1.39	1.28	21
259411	4	40	7.23	0.62	0.34	0.62	0.34	4
259421	7	227	10.93	0.08	0.05	0.08	0.05	4
259421	5	820	10.93	0.08	0.05	0.08	0.05	3
259421	6	64	10.93	0.08	0.05	0.08	0.05	24
259421	5	212	10.93	0.8	0.05	0.08	0.05	8
259422	6	145	13.88	0	0.17	0	0	6
259431	5	45	7.23	0.62	0.34	0.62	0.34	4
259431	4	147	7.23	0.62	0.34	0.62	0.34	3
259431	3	11	7.23	0.62	0.34	0.62	0.34	24
259432	5	246	7.38	1.33	0.06	0.65	0.06	8
259432	5	42	7.38	1.33	0.06	0.65	0.06	5
264211	6	10	2.91	0.44	0.14	0.14	0.14	20
264211	3	5	2.91	0.44	0.14	0.14	0.14	12
264211	7	38	2.91	0.44	0.14	0.14	0.14	19
264211	4	17	2.91	0.44	0.14	0.14	0.14	23
264221	4	14	1.85	0	0.45	0	0	20
264221	1	3	1.85	0	0.45	0	0	12
264221	7	74	1.85	0	0.45	0	0	19
264221	7	117	1.85	0	0.45	0	0	23
270611	3	13	2.14	0.87	0	0	0	17
270611	3	12	2.14	0.87	0	0	0	11
270611	5	18	2.14	0.87	0	0	0	16
270611	5	56	2.14	0.87	0	0	0	19
270611	6	21	2.14	0.87	0	0	0	28

270641	4	14	1.99	1.17	0	0	0	17
270641	2	14	1.99	1.17	0	0	0	11
270641	6	25	1.99	1.17	0	0	0	16
270641	6	33	1.99	1.17	0	0	0	19
270641	3	11	1.99	1.17	0	0	0	28
BartaB	2	300	29.36	0	0.28	0	0	1
BartaD	3	685	25.66	0	0.35	0	0	4
BartaC	3	417	25.1	0	0.4	0	0	2
BartaE	3	265	25.59	0	0	0	0	7
BartaF	4	173	25.34	0	0.16	0	0	10
BartaA	10	1583	29.36	0	0.28	0	0	22
BartaG	10	1118	29.36	0	0.28	0	0	25

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Table (2) Analysis of models with bat species richness as the dependent variable.

Richness Model	K	AICc	Delta AICc	Weights
TREES.TIME	3	247.81	0.00	0.67
HARD.TIME	2	251.06	3.25	0.13
GLOBAL	6	253.19	5.38	0.05
CloseTREE.TIME	2	253.25	5.44	0.04
EDGE.CEDAR.TIME	3	253.46	5.65	0.04
TREES	2	254.72	6.91	0.02
HARD	1	255.22	7.41	0.02
EDGE.CloseTREE.TIME	3	255.54	7.73	0.01
CEDAR.TIME	2	255.67	7.86	0.01
PATCH.TIME	2	258.12	10.31	0.00
CloseTREE	1	258.63	10.82	0.00
EDGE.CEDAR	2	259.92	12.11	0.00
EDGE.CloseTREE	2	260.50	12.69	0.00
PATCH	1	262.60	14.79	0.00
CEDAR	1	263.06	15.25	0.00
EDGE.TIME	2	266.92	19.11	0.00
EDGE	1	268.37	20.56	0.00
TIME	1	277.14	29.33	0.00
null	0	278.43	30.62	0.00

Table (3) Averaged model of TREES.TIME and HARD.TIME for richness as the dependent variable.

Variable	Parameter Estimate	Standard error	lower CI	upper CI
Intercept	4.17	0.53	3.11	5.24
Distance to Deciduous	-1.76	0.57	-2.88	-0.63
Distance to Cedar	-0.64	0.27	-1.19	-0.09
Time	0.09	0.03	0.03	0.15

Table (4) Analysis of models with number of bat calls as the dependent variable.

Bat Call Count Model	K	AICc	Delta AICc	Weights
TREES.TIME	3	124.48	0.00	0.47
TREES	2	125.87	1.39	0.23
HARD.TIME	2	127.63	3.15	0.10
CloseTREE.TIME	2	128.37	3.89	0.07
CloseTREE	1	129.97	5.49	0.03
GLOBAL.TIME	6	130.54	6.06	0.02
EDGE.CEDAR.TIME	3	130.63	6.15	0.02
EDGE.CloseTREE.TIME	3	130.63	6.15	0.02
HARD	1	130.83	6.35	0.02
EDGE.CEDAR	2	131.92	7.44	0.01
EDGE.CloseTREE	2	131.92	7.44	0.01
CEDAR	1	136.71	12.23	0.00
CEDAR.TIME	2	137.12	12.64	0.00
PATCH.TIME	2	137.38	12.90	0.00
PATCH	1	138.82	14.34	0.00
EDGE.TIME	2	150.66	26.18	0.00
EDGE	1	153.58	29.10	0.00
TIME	1	161.07	36.59	0.00
null	0	162.64	38.16	0.00

Table (5) Averaged model of TREES.TIME, TREES, HARD.TIME, and  
CloseTREE.TIME.

Variable	Parameter Estimate	Standard error	lower CI	upper CI
Intercept	2.17	0.22	1.73	2.61
Distance to Deciduous	-0.76	0.20	-1.16	-0.35
Distance to Cedar	-0.24	0.10	-0.45	-0.04
Time	-0.02	0.01	-0.04	0.00
Distance to Closest Tree	-1.10	0.16	-1.43	-0.77



Table (6) A list of bats that were sampled at Barta Brothers Ranch with corresponding scientific names and species codes.

common name	Scientific name	Species code
big brown bat	<i>Eptesicus fuscus</i>	EPTFUS
eastern red bat	<i>Lasiurus borealis</i>	LASBOR
hoary bat	<i>Lasiurus cinereus</i>	LASCIN
silver-haired bat	<i>Lasionycteris noctivagans</i>	LASNOC
western small footed myotis	<i>Myotis ciliolabrum</i>	MYOCIL
little brown bat	<i>Myotis lucifugus</i>	MYOLUC
northern long-eared bat	<i>Myotis septentrionalis</i>	MYOSEP
long-legged myotis	<i>Myotis volans</i>	MYOVOL
evening bat	<i>Nycticeius humeralis</i>	NYCHUM
tri-colored bat	<i>Perimyotis subflavus</i>	PERSUB

# Figures

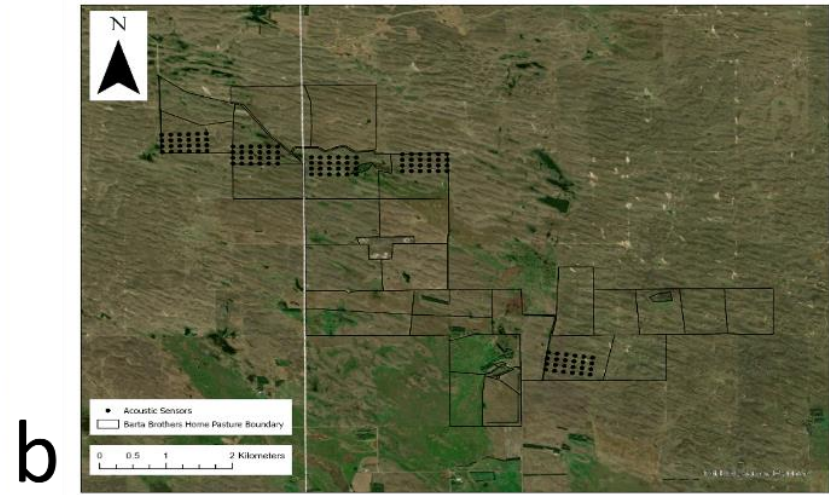
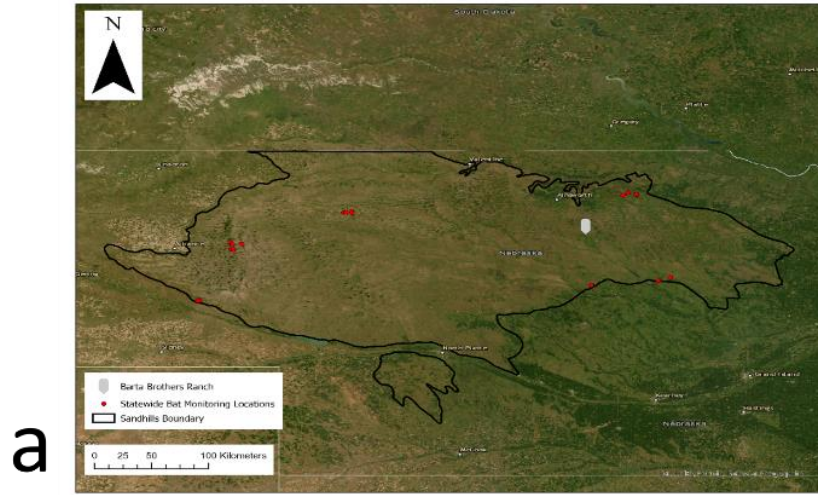


Figure 1(a) Satellite imagery showing the boundary of the Nebraska Sandhills with markers denoting the location of Barta Brothers Ranch and statewide bat monitoring locations.

(b) Satellite imagery showing the boundary of Barta Brothers Ranch home pasture with markers showing the locations of acoustic sensor grids.

(c) Satellite imagery showing an example of planted eastern redcedar windbreak complexes at Barta Brothers Ranch in the Nebraska Sandhills.

(d) Satellite imagery showing an example of how acoustic sensors were set up in relation to planted eastern redcedar windbreaks. This example is the location for Barta.B and Barta.G at Barta Brothers Ranch.

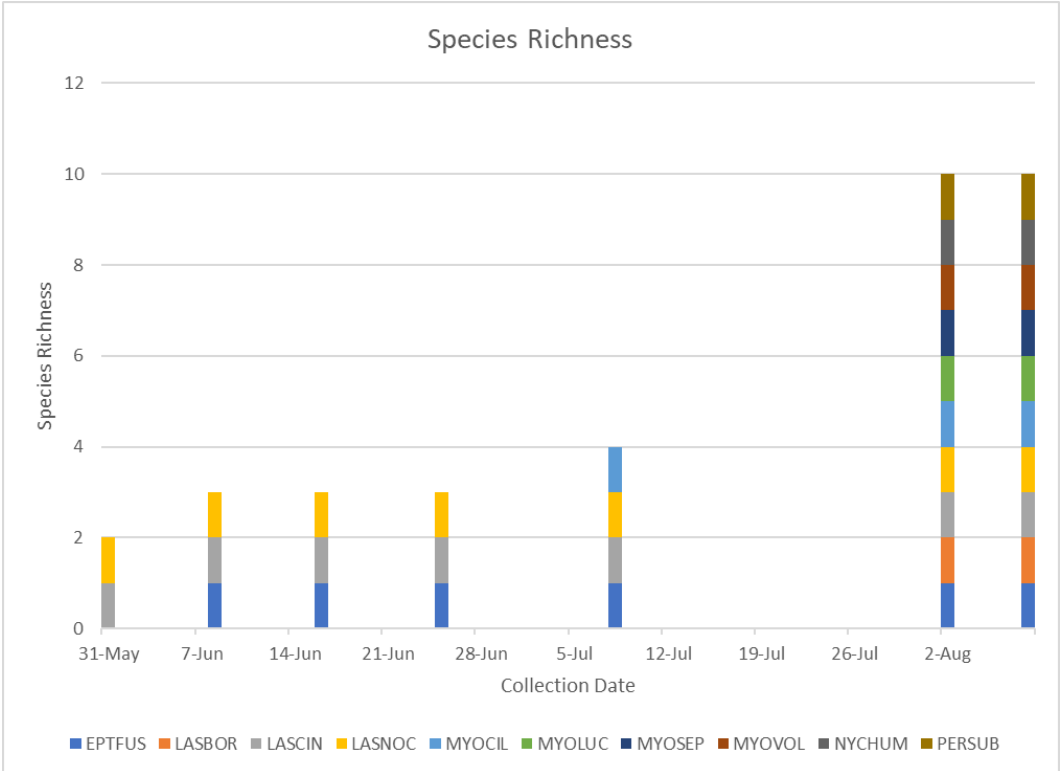


Figure (2) Species richness through time at Barta Brothers Ranch in the Nebraska Sandhills.

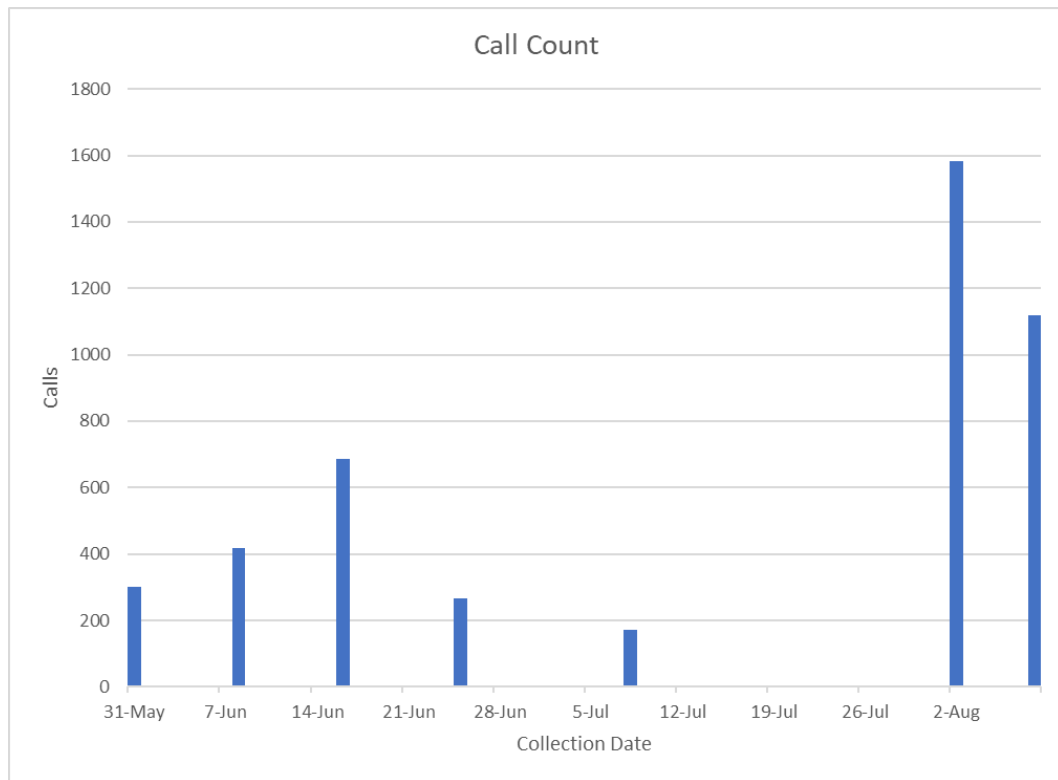


Figure (3) Number of bat calls recorded through time at Barta Brothers Ranch in the Nebraska Sandhills.

## CHAPTER 4 – USING FIRST INSTANCE TO UNDERSTAND THE UTILIZATION OF WINDBREAKS BY BATS IN THE NEBRASKA SANDHILLS

### Introduction

There are thirteen insectivorous bats in Nebraska including the federally endangered northern long-eared bat (*Myotis septentrionalis*). Bats provide important ecosystem services in Nebraska in the form of pest control. United States farmers are estimated to save anywhere from 3.7 billion to 53 billion US dollars per year by bats consumption of pest species (Boyles et al. 2011), although some assumptions of that study have been questioned (Fill et al. 2022). A single bat has the ability to consume ~1000 mosquito sized insects per hour, indirectly minimizing populations of disease vectors, though bats do prefer and target higher calorie insects such as moths (Ducummon 2000). Bats are found throughout the state but are generally found along waterways and wooded areas in Nebraska.

Nebraska bats have been found roosting in a wide array of roosts depending on location in Nebraska and species of bat. Recorded roost locations include living and dead pine and hardwood trees, rock crevices, and anthropologic structures such as houses and fences (Anderson and Geluso 2022, Fill et al. 2021). There is also evidence of eastern red bat (*Lasiurus borealis*), which is found in Nebraska, roosting in eastern redcedar trees, though moving to more insulated roosts during colder conditions (Mormann and Robbins 2007). Bats do not always hunt directly next to their roost; bats in Nebraska may travel multiple kilometers from roost to feeding grounds (Fill et al. 2021).

Bat population throughout North America have been in decline presumably due to White-nose syndrome, habitat degradation and anthropologic inhibitions such as wind turbines. Habitat change has occurred heavily since European expansion into the Americas. Forest structure has changed though time (Nemec et al. 2011), the introduction of species such as Russian olive (*Elaeagnus angustifolia*), autumn olive (*Elaeagnus umbellate*) and eastern redcedar (*Juniperus virginiana*) changes the midstory of forests causing obstructions for flying bats. White nose syndrome is a fungal infection found in bats that is especially detrimental to northern long-eared bats. White nose syndrome infects winter colonies, spreading throughout, causing populations to rapidly decline and in some cases leading to local extirpation (Frick et al. 2015, Warnecke et al. 2012). White nose syndrome interrupts hibernation, causing increased activity in hibernacula and lowering survival rate (Reynolds et al. 2016). Migratory bat species also face obstacles as they are at a higher risk of mortality around wind turbines as they seasonally move (Arnett et al. 2008, Cryan et al. 2014).

The Nebraska Sandhills is a semi-arid grassland in central Nebraska consisting of rolling grass-stabilized sand dunes. The Sandhills have undergone immense pressure from planted and encroached woody cover causing the region to transition to a cedar dominated woodland, with approximately 40,000 acres of land is lost each year to woody encroachment (Scholtz & Twidwell 2022, Fogarty et al. 2022a, 2022b). Eastern redcedar trees are popularly used as windbreaks as their thick canopies effectively stop wind and their ability to survive in many habitats (Van Haverbeke 1976). Cedar tree branches can be wide and their twisting branches form thick umbrellas at the base causing herbaceous cover to decline which lowers diversity and forage quality for grazing animals (Briggs et

al. 2002), causing redcedar dominated areas to be undesirable to most Sandhills residents because they may decrease forage production (Roberts and Allen 2023).

Eastern redcedar windbreaks have thick canopies and cluttered structure but the outside and top of windbreaks may provide usable edge habitat, stretching in straight lines throughout the Sandhills. The purpose of this study was to determine if bat species in Nebraska are using these cedar features as roost habitat or localized travel corridors from their roosts by analyzing first instance calls inside within acoustic sensor grids. I hypothesize that bats are utilizing these features as roosts or traveling from roost to feeding ground using the windbreaks, showing an increased number of first instance calls along the cedar windbreak.

## **Methods**

### **Study Area and Site Selection**

I conducted this study at Barta Brothers Ranch in the Nebraska Sandhills (Figure 1a). Barta Brothers Ranch is a University of Nebraska–Lincoln field station in Rock and Brown Counties in the Northeastern Sandhills (42.226381, -99.635297). Barta has an area of approximately 2,000 hectares representative of the Eastern Sandhills with rolling sand dunes covered in cool and warm season grasses, sedges, forbs, planted cedar tree lines, and patches of invading cedar (Figure 1b,1c). The ranch is used by University of Nebraska students, staff, and partners for agricultural, range and wildlife research and demonstrations. I selected study sites using areal imagery in ArcGIS Pro, ground truthed for accuracy. The study sites consist of planted cedar tree lines or stands adjacent to open rangeland. Sites were free of livestock during the data collection period and stock tanks were dry.



## **Materials and Equipment**

I recorded bat calls using Titley Scientific Anabat Express acoustic sensors with an effective range of 50 meters in a grid of 24 sensors 100 meters apart from one another resulting in a sensor area of 400 meters by 600 meters (Figure 1d). The acoustic sensors were mounted on extendable painter's poles customized to hold the sensors approximately 4 meters off the ground to improve detection. The poles were held up using metal stakes attached to a piece of PVC pipe to ensure sensors are kept in place during the data collection period. I created the sensor grid in ArcGIS Pro, with 24 fishnet points 100 meters away from each other in a 4 x 6 formation (Figure 1d). These points were converted into GPX points using ArcGIS Pro tools, transferred to a Garmin GPSMAP 64, and acoustic sensors were placed at each point. The sensor grid was adjacent to or around the cedar tree feature with sensors in range of cedar habitat or open grassland habitat (Figure 1d). Acoustic sensors were set to night mode turning on 30 minutes before sunset and turning off 30 minutes after sunrise. The data collection period is 4 nights of non-inclement weather to ensure bat activity. Inclement weather, in this study, included nightly temperatures below freezing, high winds Beaufort 5 or above, and precipitation. If any of these events occurred during data collection the sensors were left up another night to reach the target of 4 collection nights. Calls from all data collection nights at a site were combined for total counts.

## **Bat Identification**

Detected bat calls were stored on an SD card inside of the acoustic sensor as raw nightly acoustic files. These raw files were uploaded into Analoow (version 4.6c) which separates the raw file into individual bat passes or individual sound sequences. These

files are output from AnalookW as ZCA files and can be bat passes, insect noise, wind, or any other sound within the frequency and spatial range of the acoustic sensor. For species identification the ZCA files are uploaded to Wildlife Acoustics Kaleidoscope Pro (Version 5.4.2). Kaleidoscope uses signal parameter settings selected by the user to identify bat calls. These parameters include frequency range, length of detected pulses, maximum inter syllable gap and minimum number of bat pulses. For this study, frequency range was set to 8-120 kHz, length was set to 2-500 ms, inter-syllable gap was set to 500 ms, and minimum number of pulses was set to 3. Species included for consideration for identification in Kaleidoscope included big brown bat, eastern red bat, hoary bat, silver-haired bat, western small-footed myotis, little brown bat, northern long-eared bat, long legged myotis, evening bat, and tri-colored bat (Table 3). Kaleidoscope identifies characteristics of the sound waves and compares them with their database of confirmed calls to give a species identification. Kaleidoscope categorizes each ZCA file either as noise, a known bat call or an unidentified bat call. Species IDs are given as a percentage of matched pulses. For example, a bat may fly by a sensor and emit 10 pulses inside of the sensor range, if 8 out of 10 of the pulses match a given species Kaleidoscope will display 80% match for that species. Kaleidoscope will, on some occasions, give a second and third alternative identification in decreasing percent match, when characteristics of a pulse match more than one species. The highest percentage match was taken and used for data analysis. Unidentified bat calls were not used in statistical analysis or data interpolation. Kaleidoscope also outputs presence probability (Britzke et al. 2002), which I utilized at the site level.

Data censoring happened when a sensor malfunctioned. The only instance of data censoring occurred at site Barta.D for sensor 8, the sensor had blown over from wind and the microphone was bent. No data was recorded from this sensor throughout the collection period.

### **Statistical Analysis**

Statistical analysis was completed using R (R Core Team 2021). Collected calls were viewed at the site level and the first call each night after sunset for each available species was recorded as one first instance – the first location of a given species on a given night. Distance measurements to the nearest windbreak were taken in ArcGIS Pro using the measure tool to each sensor that a first instance was recorded. First instances were binned based on their distance from the windbreaks. Distances from 0-100 meters were placed into the 0.1km bin, distances from 101-200 meters were placed into the 0.2km bin, distances from 201-300 meters were placed into the 0.3km bin, distances from 301-400 meters were placed into the 0.4km bin, and distances from 401-500 meters were placed into the 0.5km bin. Binned first instances from all sites at Barta Ranch were organized by species, by all species combined, and by all *Myotis* bat species combined (table 1). Statistical analysis was done on species or groups of species with  $\geq 10$  first instances, this included *Eptesicus fuscus*, *Lasiurus cinereus*, *Lasionycteris noctivagans*, all species combined, and all *Myotis* species combined. Statistical analysis was linear regression that predicts number of first instances as a function of distance to the planted windbreak. Graphs were also constructed using the plotted first instances vs distance from the windbreak, with the fitted regression line and error.

## Results

There were 10 species with a recorded first instance equaling a total of 89 first instance calls. Species groups and species that met  $\geq 10$  first instance calls needed for analysis included *Eptesicus fuscus*, *Lasiurus cinereus*, *Lasionycteris noctivagans*, all species combined, and all *Myotis* species combined. All species combined had 89 first instance calls, all *Myotis* species combined had 10 first instance calls, *Eptesicus fuscus* had 13 first instance calls, *Lasiurus cinereus* had 26 first instance calls, the maximum number a species could have over the 26 nights of data collection, and *Lasionycteris noctivagans* had 19 first instance calls.

The linear regression for all combined species at Barta Ranch had a negative distance estimate of -63.00, a p-value of 0.01 and an adjusted  $R^2$  of 0.89 (intercept estimate 36.70, intercept SE 3.63, intercept p-value  $< 0.00$ , distance SE 10.94) (Table 2) (Figure 2). The linear regression for *Eptesicus fuscus* had a negative distance estimate of -10.00, a p-value of 0.10 and an adjusted  $R^2$  of 0.54 (intercept estimate 5.60, intercept SE 1.38, intercept p-value 0.03, distance SE 4.16) (Table 2) (Figure 3a). The linear regression for *Lasionycteris noctivagans* had a negative distance estimate of -12.00, a p-value of 0.08 and an adjusted  $R^2$  of 0.59 (intercept estimate 7.40, intercept SE 1.53, intercept p-value 0.02 distance SE 4.62) (Table 2) (Figure 3b). The linear regression for *Lasiurus cinereus* had a negative distance estimate of -7.00, a p-value of 0.51, and an adjusted  $R^2$  of -0.12 (intercept estimate 7.30, intercept SE 3.08, intercept p-value 0.09, distance SE 9.29) (Table 2) (Figure 3c). The linear regression for all combined *Myotis* had a negative distance estimate of -13.00, a p-value of 0.16 and an adjusted  $R^2$  of 0.37

(intercept estimate 5.90, intercept SE of 2.35, intercept p-value 0.09, distance SE 7.10) (Table 2) (Figure 3d).

## Discussion

For all bat species combined, there is a significantly higher number of first instances close to windbreaks and a steady decrease with distance from tree line (Figure 2). The adjusted  $R^2$  for the combined species regression was 0.89; 89% of variance found in first instances was accounted for with distance from windbreaks. First calls from bats inside of the sensor grids happen near to the windbreak, documenting that bat nightly activity starts near these planted redcedar features. *Eptesicus fuscus* and *Lasionycteris noctivagans* had similar results with negative distance estimates showing higher numbers of first calls near to the planted windbreaks (Figure 3a, 3b), with adjusted  $R^2$  of 0.54 and 0.59 respectively, explaining over half of the variance in first instance. *Eptesicus fuscus* and *Lasionycteris noctivagans* fits were less pronounced (p-values of 0.10 and 0.08, respectively). Combined *Myotis* species showed a negative estimate but with a p-value of 0.16 (Figure 3c). *Myotis* species only had 10 first instances spread between 4 species over 26 nights, giving this regression low statistical power. The four *Myotis* species could also exhibit different behavior for different species, given a larger sample size, causing the data to show a nonsignificant trend. Given these reasons *Myotis* species results cannot be supported with the available data. *Lasiurus cinereus* had a unique result compared to the other regressions, reporting a negative distance estimate of -7.00 but error of 9.29 and a p-value of 0.51 (Figure 3d). These results show no negative or positive change in the number of first instances as you move from the windbreak, with an estimate including zero when error is accounted for. This species showed similar results

in chapter 2 where *Lasiurus cinereus* used the whole sensor grid evenly throughout nightly activity.

Bats, in general, are using Planted redcedar windbreaks for roosting or traveling to their feeding ground to roost; this behavior, in the case of *Lasiurus cinereus*, can differ from species to species. Most species saw their first instance of the night close to windbreaks leading to the conclusion that they are using cedar trees for roosting and if not for roosting to travel from their roost to feeding grounds, which can be multiple kilometers (Fill et al. 2021). Another possibility with windbreak travel is bats are using these structures to enter and fill in the Sandhills not as just roost – feeding ground travel paths but long distance ‘highways’ connecting the entirety of the Sandhills.

Other explanations for the trend observed are possible. One such possibility is that bats are flying higher than 50 meters, higher than the acoustic sensor range, when coming from their roost locations and they make their descent around usable structure for feeding. Another possibility is that since nightly usage is already clustered around the windbreaks as shown in chapter 2 then the first call would be close to the windbreaks as well. This theory falls short as you would see more calls towards the end of the grid if bats were crossing the grasslands from their roosts to reach feeding grounds. One way to have concrete evidence of roosting would be to physically capture bats with mist nets and attach radio transmitters, then during the day time, follow the signals and try to find where bats are roosting.

*Lasiurus cinereus* behaved differently from other bat species present with no significant spatial relationship of where first instances are occurring, showing this species may be flying across large sections of open grassland when traveling from roost to

feeding ground (Figure 4), and/or that their roosts are spatially dispersed and not, or less closely, associated with cedar windbreaks. *Lasiurus cinereus* tends to favor open habitats and higher altitudes (Norberg & Raynor 1987; Jones et al., 2016), Sandhills with strips of vertical structure throughout could offer these open habitats that it prefers.

Roost locations are important for the survival of bat species as they play a large part in the bat's lives. Bats use their roosts for a place to sleep during the day and to digest their food, for copulation and raising their young, and for protection from weather conditions and predators (Kunz 1982). In Nebraska Bats have found roosting in human built structures such as houses and fences, living and dead deciduous and pine trees, and rock crevasses (Anderson and Geluso 2022, Fill et al. 2021). There is even an instance that an eastern red bat, which is common in Nebraska, was found using an Eastern redcedar for its daytime roost (Mormann and Robbins 2007). Further study of this topic is warranted in the Sandhills. Physical capture and attachment of radio antennae could be done to track bats back to their roost and find their true roost locations.

As the Sandhills become more redcedar dominated bats will likely start using the area more as core grasslands continue to shrink. If bats are roosting in cedar windbreaks, especially the endangered northern-long eared, dilemmas of what threatened species or landscape to focus conservation efforts on could arise. Land managers will have to make decisions on differing ecosystem services provided by bats and those provided by diverse productive grasslands.

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## Tables

Table (1) Number of first instances in each binned distance for each species at Barta Brothers Ranch.

Species	0.1km	0.2km	0.3km	0.4km	0.5km
EPTFUS	6	2	2	2	1
LASBOR	2	2	0	0	0
LASCIN	4	10	4	5	3
LASNOC	6	4	6	2	1
NYCHUM	1	1	1	1	0
MYOCIL	2	1	0	0	1
MYOLUC	2	0	1	0	0
MYOSEP	1	0	0	0	0
MYOVOL	2	0	0	0	0
PERSUB	1	0	1	0	1
Combined Myotis	7	1	1	0	1
Combined Species	34	21	16	10	8

Table (2) Outputs from linear regression analysis for first instance analysis at Barta Brothers Ranch.

Species	Intercept Estimate	Intercept SE	Intercept p-value	Distance Estimate	Distance SE	Distance p-value	Adjusted R <sup>2</sup>
Combined Species	36.70	3.63	0.00	-63.00	10.94	0.01	0.89
EPTFUS	5.60	1.38	0.03	-10.00	4.16	0.10	0.54
LASNOC	7.40	1.53	0.02	-12.00	4.62	0.08	0.59
LASCIN	7.30	3.08	0.10	-7.00	9.29	0.51	-0.12
Combined Myotis	5.90	2.35	0.09	-13.00	7.10	0.16	0.37

Table (3) A list of bats that were sampled at Barta Brothers Ranch with corresponding scientific names and species codes.

common name	Scientific name	Species code
big brown bat	<i>Eptesicus fuscus</i>	EPTFUS
eastern red bat	<i>Lasiurus borealis</i>	LASBOR
hoary bat	<i>Lasiurus cinereus</i>	LASCIN
silver-haired bat	<i>Lasionycteris noctivagans</i>	LASNOC
western small footed myotis	<i>Myotis ciliolabrum</i>	MYOCIL
little brown bat	<i>Myotis lucifugus</i>	MYOLUC
northern long-eared bat	<i>Myotis septentrionalis</i>	MYOSEP
long-legged myotis	<i>Myotis volans</i>	MYOVOL
evening bat	<i>Nycticeius humeralis</i>	NYCHUM
tri-colored bat	<i>Perimyotis subflavus</i>	PERSUB

# Figures

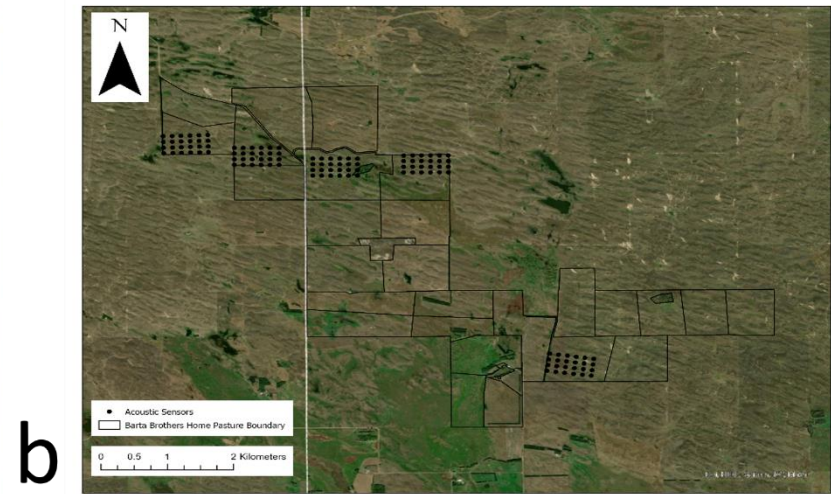
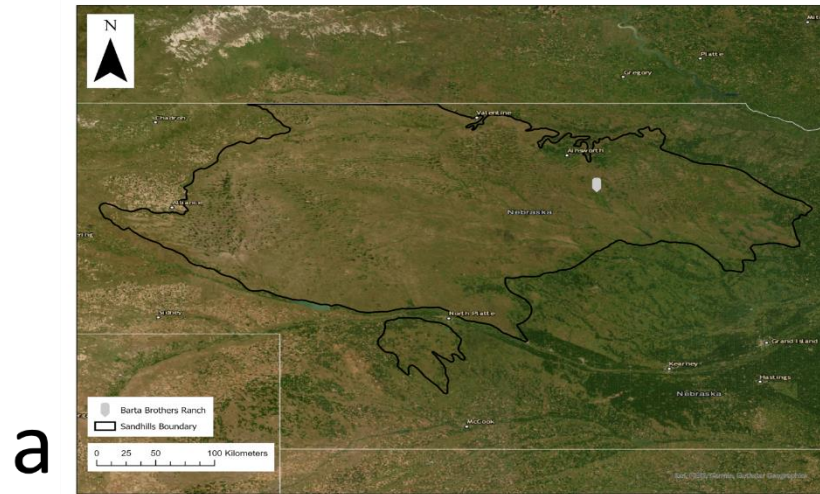


Figure 1(a) Satellite imagery showing the boundary of the Nebraska Sandhills with a marker denoting the location of Barta Brothers Ranch.

(b) Satellite imagery showing the boundary of Barta Brothers Ranch home pasture with markers showing the locations of acoustic sensor grids.

(c) Satellite imagery showing an example of planted eastern redcedar windbreak complexes at Barta Brothers Ranch in the Nebraska Sandhills.

(d) Satellite imagery showing an example of how acoustic sensors were set up in relation to planted eastern redcedar windbreaks. This example is the location for Barta.B and Barta.G at Barta Brothers Ranch.



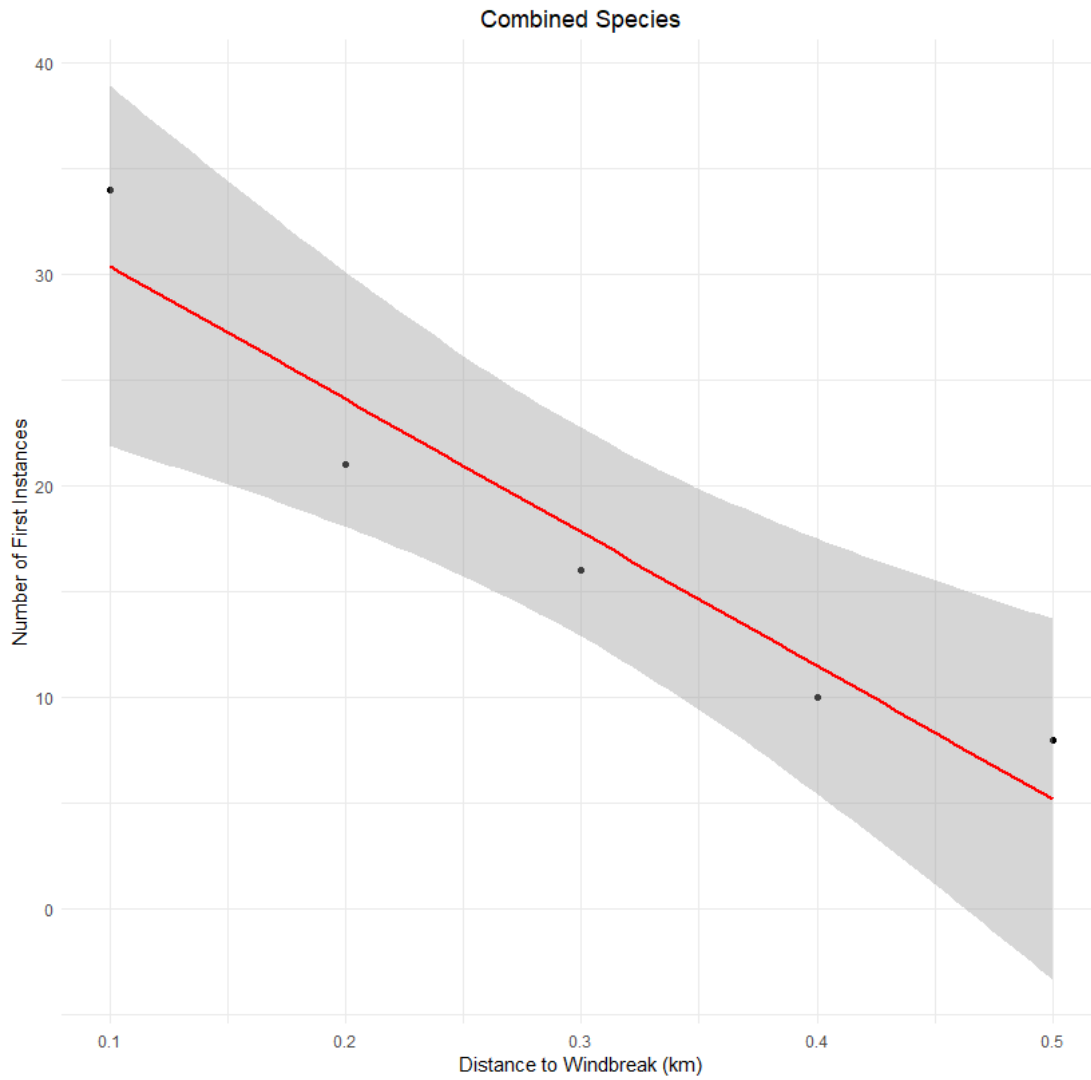


Figure (2) Graph showing plotted points for the number of first instances for all species in each binned distance value and a fitted regression line with error at each point in the line.

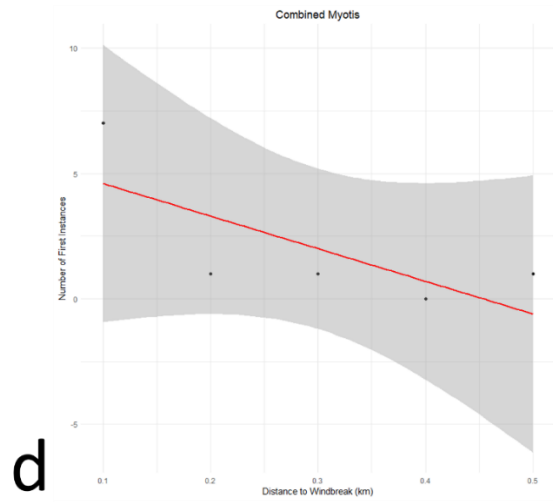
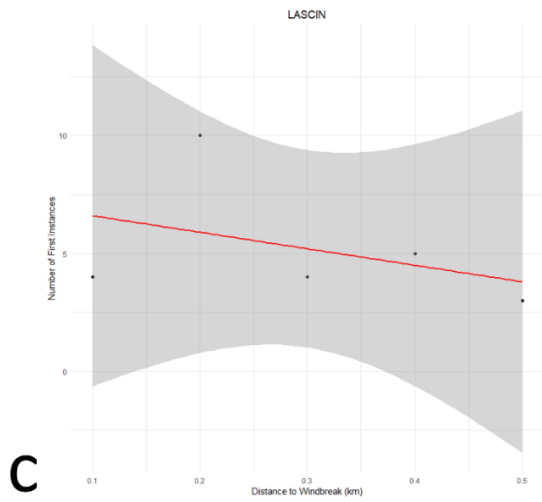
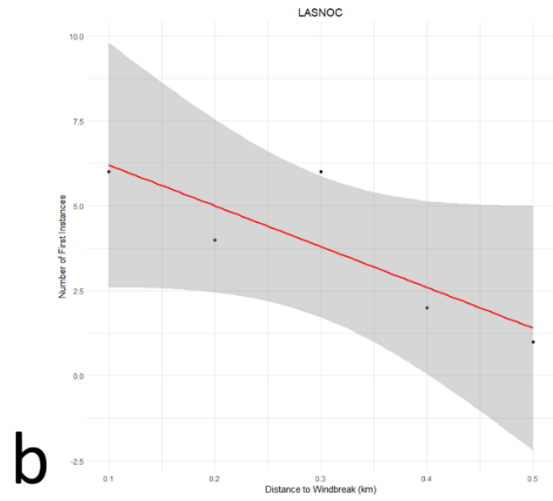
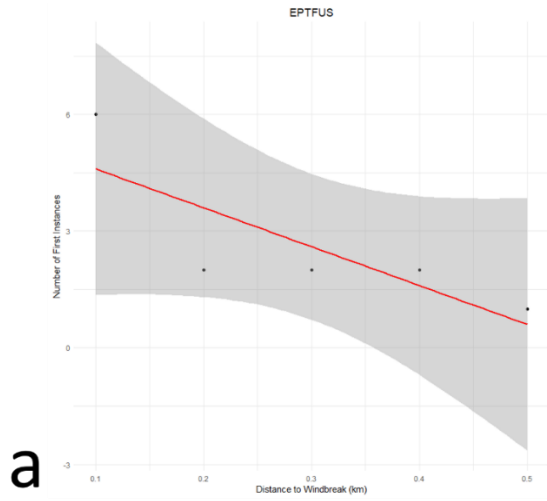


Figure 3 (a) Graph showing plotted points for the number of first instances for big brown bat (*Eptesicus fuscus*) in each binned distance value and a fitted regression line with error at each point in the line.

(b) Graph showing plotted points for the number of first instances for silver-haired bat (*Lasionycteris noctivagans*) in each binned distance value and a fitted regression line with error at each point in the line.

(c) Graph showing plotted points for the number of first instances for hoary bat (*Lasiurus cinereus*) in each binned distance value and a fitted regression line with error at each point in the line.

(d) Graph showing plotted points for the number of first instances for combined Myotis species including: northern long-eared bat (*Myotis septentrionalis*), little brown myotis (*Myotis lucifugus*), western small-footed myotis (*Myotis ciliolabrum*), and long-legged myotis (*Myotis volans*) in each binned distance value and a fitted regression line with error at each point in the line.

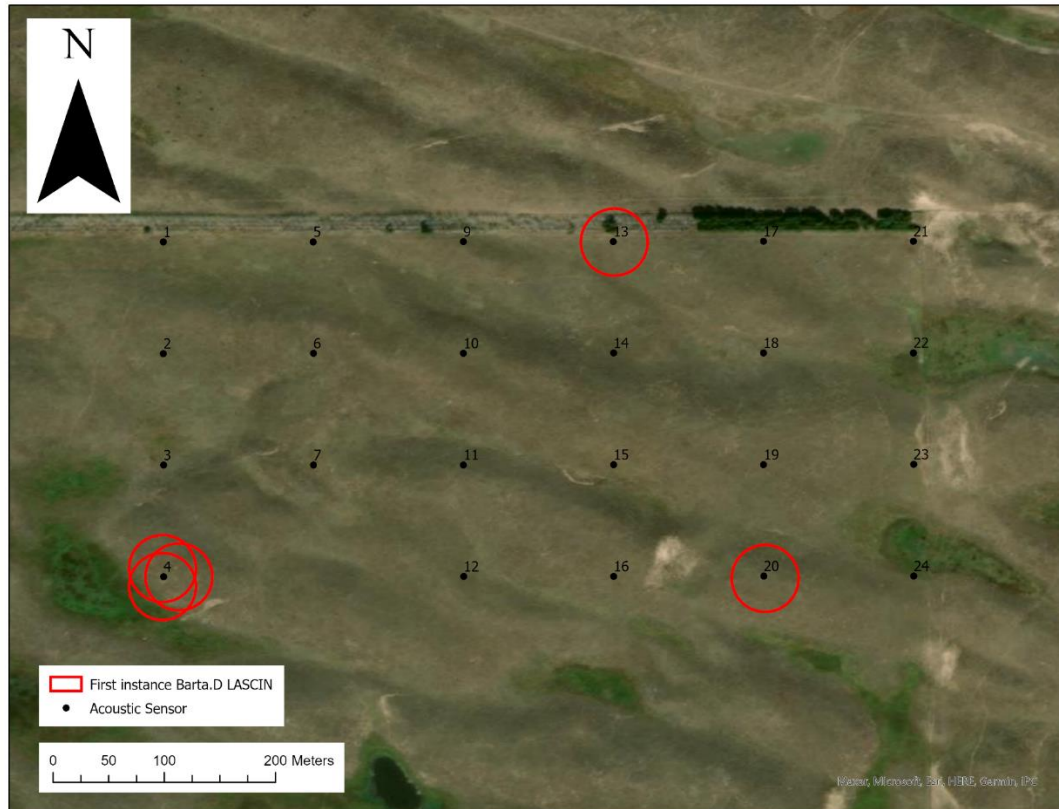


Figure (4) Map showing the sensors where first instances occurred for *Lasiurus cinereus* over five nights at site Barta.D.

## CHAPTER 5 - CONCLUSION

Bats in the Nebraska Sandhills are utilizing planted eastern redcedar windbreaks during nightly activity. Activity and how closely bats stay to the windbreaks is species dependent. Big brown bat (*Eptesicus fuscus*), eastern red bat (*Lasiurus borealis*), and evening bat (*Nycticeius humeralis*) stayed close to windbreaks and the majority of their nightly activity occurred near these features, linear regressions showed significant relationships between distance from windbreaks and bat activity intensity. These results were also clear in kriging interpolation maps which showed much higher activity near the windbreak and limited activity as you move into the grassland. Hoary bat (*Lasiurus cinereus*) and silver-haired bat (*Lasionycteris noctivagans*) used the limited sensor range of the grid evenly as no relationship was found. Both of these species did show a significant relationship to trees on a larger scale when examining NABat data, showing that hoary bat and silver-haired bat likely also exhibit patterns of usage near the windbreak and less activity the further into the grassland you go, though their usage area is likely further than the 600m range the Barta study sites allowed.

Bat richness and call count can be predicted with distance to trees and time through the summer when examining data from the Sandhills. When running multi model analysis for richness the results showed that distance to deciduous trees, redcedar trees, and tree patches and time were the best predictors. Distance to deciduous trees was the highest weighted variable, showing that bats likely show preference to hardwoods but also will use redcedar trees. Time, when accounting for richness showed that as the summer progressed more species show up in the Sandhills, this is likely do to bats awaking from hibernacula or arriving from migration at different times. This could also

be due to bats taking more time to filter into the Sandhills, possibly due to the difficulties that open grasslands present. When running multi model analysis for bat call count or bat activity the results showed that distance to deciduous trees, redcedar trees, closest tree and tree patches and time were the best predictors. Distance to deciduous trees was the highest weighted variable, showing that bats likely show preference to hardwoods but also will use redcedar trees. Time, when accounting for bat call count or activity showed that as summer progressed call count stayed pretty flat showing a very small decrease in call count.

Bats use planted eastern redcedar windbreaks either for roosting habitat or to travel from roost to feeding grounds. When examining the first call each night by all species combined the data showed a very strong favor to having the first call of the night near the windbreak. This tells us that bats are either emerging directly from the windbreak to feed or using these features as a 'highway' of sorts to travel from where they prefer to roost to their feeding grounds, this conceivably could be multiple kilometers away, around the redcedar windbreaks. When breaking this analysis down to the species level, though having much lower data points and power, showed that this pattern is indeed species dependent. Hoary bat showed no favoritism for their first call being close to the windbreak. Combining this knowledge with results from my chapter on nightly activity this lines up. The usage area for hoary bat appears to be larger than the grid of 400 by 600 meters. The assumption I can make from this is hoary bat likely traverses larger areas of open grassland compared to other species and may even travel from roost to feeding grounds over large areas of open grassland. Future roost studies in

the Sandhills would benefit from using typical mist net capturing and radio transmitters to find roosting locations and compare them with the findings here.

This thesis utilized multiple analysis techniques to answer the question of whether bats are utilizing the Nebraska Sandhills and redcedar windbreaks. I believe the conclusion is that they are indeed utilizing the Sandhills and redcedar windbreaks but show a preference for deciduous trees. This use likely due to the afforestation and encroachment of woody plants into the Sandhills. Bats are likely using long planted windbreaks as highways of travel through the Sandhills, avoiding open areas of grassland.

There were in total ten species of bat recorded at Barta Brothers Ranch, these include: *Eptesicus fuscus*, *Lasiurus borealis*, *Lasiurus cinereus*, *Lasionycteris noctivagans*, *Myotis ciliolabrum*, *Myotis lucifugus*, *Myotis septentrionalis*, *Myotis volans*, *Nycticeius humeralis*, and *Perimyotis subflavus*. When accounting for presence probability only six reported a significant likelihood of presence, this could be attributed to low numbers of calls recorded for some species. The six species at Barta Ranch with a significant presence probability likelihood include: *Eptesicus fuscus*, *Lasiurus borealis*, *Lasiurus cinereus*, *Lasionycteris noctivagans*, *Myotis volans*, *Nycticeius humeralis*. NABat data recorded 9 bat species throughout the Sandhills, the discrepancy being *Myotis Volans* was not recorded during their data collection.

The northern long-eared bat is of special concern as it is a listed endangered species and highly susceptible to white nose syndrome. This thesis found that if present at all in the Sandhills is only present in very small numbers. Collection at Barta Ranch showed only one instance of a northern long-eared presence represented by 3 calls, and

the presence probability likelihood for these calls was non-significant. NABat data collected 4 northern long-eared calls spread between 3 sites over a 5-year period. These numbers are very low compared to other species with hundreds of recorded calls at multiple sites. Northern long-eared bats are adapted for hunting and flying inside of congested forests. The open and extended edge habitats of the Sandhills may not be suitable for their needs or preferences, it is possible the species is excluding itself from this area.

Eastern redcedar removal and management on invading redcedar is high though the removal of the source of invasion, human planted windbreaks, is low. Windbreaks are still valued for their protection from wind and inclement weather, this is doubly important in largely open landscapes such as the Sandhills. Management of any woody species will put pressure on bats which are already pressured by habitat degradation and population decline. Our planet is becoming more crowded and less stationary as climate changes and human population increases. Humanity will need to grapple with an uptick in dueling ecosystem services and will have to make hard choices on which services, habitats, or species take preference on the landscape.