M.L. 2015 Project Abstract

For the Period Ending June 30, 2018

PROJECT TITLE: Genetic and Camera Techniques to Estimate Carnivore Populations
PROJECT MANAGER: Ron Moen
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FUNDING SOURCE: Environment and Natural Resources Trust Fund
LEGAL CITATION: M.L. 2015, Chp. 76, Sec. 2, Subd. 03a as extended
M.L. 2017, Chapter 96, Section 2, Subdivision 18

APPROPRIATION AMOUNT: \$200,000 AMOUNT SPENT: \$159,506 AMOUNT REMAINING: \$40,494

Overall Project Outcome and Results

Minnesota has 20 carnivore species, 3 of which are very rare. Current monitoring methods of summer scent station surveys, winter track surveys, and population modelling could be complemented by camera traps and genetic DNA analysis. We used camera traps to obtain 3,400 images of carnivores over 12,000 camera-nights. American marten, fisher, short-tailed weasel, wolf, red fox, and gray fox were most frequently photographed. Occupancy analysis showed habitats used by each of these species. Mark-recapture estimation of population size was not possible. Camera traps could include significant public involvement, as is being done by the Wisconsin DNR. A second outcome of camera trap data is testing a Random Encounter Model to determine if population densities can be estimated without identifying individuals.

We implemented sampling protocols to obtain hairs non-invasively from weasels and larger carnivores. Hair collection was less efficient than camera traps. Wolf scat collection in snow was unpredictable. However, DNA analysis identified individuals in the collected samples. Hair and scat collection is technically feasible but logistically difficult to implement.

A consistent conclusion from genetic sampling protocols is that the cost to obtain and analyze genetic samples, at present, would make it difficult to implement a mark/recapture population estimate for management on a large spatial scale. We did not fully expend the ENRTF funding because genetics collaborators were fully occupied with their own research. One tangible outcome of this project is that a genetic collaborator with time to do the analysis is critical.

The Minnesota Carnivore website has descriptions, pictures from the camera trap project, and historical harvest data in Minnesota and adjoining jurisdictions. The website will be updated periodically to provide new information—it is the only Minnesota-specific Carnivore website available. In addition, we will finish 4 Technical Reports and a peer-reviewed paper on occupancy modelling in Fall 2018.

Project Results Use and Dissemination

- 1. Technical reports summarizing the entire project.
 - a. **Camera Trapping**: Moen, R. and B. Houck. 2018. Monitoring Carnivore Populations in Northeast Minnesota with Camera Traps. NRRI Technical Report No. NRRI/TR-2018/44. University of Minnesota Duluth.
 - b. Weasel tube hair snares: Houck, B. and R. Moen. 2018. Use of Tube Hair Snares to Detect Weasels in Minnesota. NRRI Technical Report No. NRRI/TR-2018/45. University of Minnesota Duluth.
 - c. **Cable-Restraint hair snares**: Houck, B. and R. Moen. 2018. Use of Single-Capture Hair Snares to Detect Carnivores in Minnesota. NRRI Technical Report No. NRRI/TR-2018/43. University of Minnesota Duluth.
 - d. Wolf Scat Collection and Genetic Analysis. Moen, R. 2018. Genetic Analysis of Wolf Scats Collected from Snow. NRRI Technical Report No. NRRI/TR-2018/51. University of Minnesota Duluth.
- 2. Occupancy modelling manuscript. Houck, B. and R. Moen. 2018. Occupancy modelling of carnivores in northeastern Minnesota from camera trap data. Manuscript to be submitted for peer review.
- 3. The Minnesota Carnivore website is currently located at https://champ.d.umn.edu/mc. It is being relocated to https://www.nrri.umn.edu/mc.



Environment and Natural Resources Trust Fund (ENRTF) M.L. 2015 Work Plan Final Report

Date of Report: November 12, 2018			
Date of Next Status Update Report:	Final Report		
Date of Work Plan Approval:	January 14, 2015		
Project Completion Date:	June 30, 2018		

PROJECT TITLE: Genetic and Camera Techniques to Estimate Carnivore Populations

Project Manager: Ron Moen

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Location: Aitkin, Beltrami, Carlton, Cook, Itasca, Koochiching, Lake, St. Louis, statewide

Total ENRTF Project Budget:	ENRTF Appropriation:	\$200,000
	Amount Spent:	<u>\$159,506</u>
	Balance:	<u>\$40,494</u>

Legal Citation: M.L. 2015, Chp. 76, Sec. 2, Subd. 03a M.L. 2017, Chapter 96, Section 2, Subdivision 18

Appropriation Language:

\$200,000 the first year is from the trust fund to the Board of Regents of the University of Minnesota – Duluth for the Natural Resources Research Institute to use genetic sampling and remote cameras to improve monitoring of distributions and estimate population sizes of carnivore species.

Carryforward (a) The availability of the appropriations for the following projects are extended to June 30, 2018: (6) Laws 2015, chapter 76, section 2, subdivision 3, paragraph (I), Genetic and Camera Techniques to Estimate Carnivore Populations.

I. PROJECT TITLE: Genetic and Camera Techniques to Estimate Carnivore Populations

II. PROJECT STATEMENT:

Minnesota has 17 carnivore species which are more common, and 3 other species which are very rare or only occasional visitors. The carnivore species in Minnesota range in size from the tiny least weasel to the black bear. Species with high harvest levels such as bobcat, fisher, marten, and coyote are tracked well, while there is much less known about species with lower harvest levels such as badgers. Some carnivore species such as the coyote, mink, black bear, and raccoon are common throughout much or all of Minnesota. Other species are either at the extreme edge of their distribution or are only known from historical accounts or recent occasional reports. Relatively little is known about these rarer species.

Current methods for monitoring carnivores are the summer scent station survey, a winter track survey, and population modelling from harvest data. While these methods are useful, other techniques have been developed for monitoring carnivores over the past 20 years that could be tested in Minnesota. Namely, the development of genetic DNA analysis and increasing use of trail cameras make it possible to improve existing methods of monitoring distribution and numbers of carnivores. Trail cameras would also provide a mechanism for significant public involvement with future development of a citizen science component.

There are many reasons why it would be beneficial to increase monitoring of carnivores and improve understanding of current distributions. One benefit of increased carnivore monitoring would be an independent estimate of wolf populations in Minnesota. Wolves were removed from U.S. Endangered Species Act protection in January 2012, and species management was transferred to the DNR. Regulated hunting and trapping seasons for wolves were held in 2012 and 2013, and continued harvest management will require accurate population estimates. An accurate estimate of wolf numbers will also be useful for considering effects wolves might be having on other species (e.g., moose and deer). Independent estimates using either camera trap or genetic mark-recapture testing could help wolf management.

Additionally, wildlife diseases are also of growing concern, especially in urban areas where there is an increased potential for transmission of diseases to humans and domestic animals. Some of the diseases that can be transmitted from wildlife to humans or companion animals include canine distemper, canine heartworm, sarcoptic mange, leptospirosis, erlichiosis, and canine parvovirus. Other diseases also affect humans and domestic animals. These diseases could be of increasing concern in the future, in part because generalist carnivore species such as coyotes, raccoons, and skunks appear to thrive in urban areas.

The specific goals of this proposal for carnivore populations in Minnesota are to:

- 1. Develop a remote camera based protocol for occupancy modelling and for independent estimates of census population size if applicable to a species.
- 2. Develop a genetic sample collection and analysis protocol to estimate genetic effective population size for carnivore species. We will also make genetic mark-recapture estimates of census population size when applicable to a species.
- 3. Review historical trends in distribution and abundance of Minnesota carnivores from museum records and DNR data.

III. OVERALL PROJECT STATUS UPDATES:

Project Status as of December 31, 2015:

We have developed a field protocol for remote camera based occupancy modeling. Initial deployments of cameras have resulted in detection of 10 of 16 common carnivore species with over 500 visits to cameras. Early indications are that we can estimate occupancy for at least 6 carnivore species using remote cameras. We have also identified candidate species for genetic work, and have developed protocols for genetic sample collection, processing, and storage. We have begun testing techniques to collect genetic samples from small and large carnivores. We will increase the number of samples in winter with snow cover.

Project Status as of June 30, 2016:

Remote cameras have been deployed at 97 sites with 13 of 16 common carnivore species in Minnesota identified. We have obtained about 1 picture event every 3 camera-days. The protocol for collection of genetic samples has been developed. Genetic samples are collected using tubes with wire brushes that smaller carnivore species walk through, or with cable wire strands set up for larger carnivore species. The small tube collectors have been more efficient than cable wire strands, with 1 sample collected per 20 snare-days. The cable wire strands for obtaining large carnivore samples have been less efficient, with about 1 sample collected per 75 snare-days. However, larger carnivores have lower population densities, so this loss in efficiency would be expected. We will continue deployment of trail cameras and genetic sampling devices this summer.

Project Status as of September 30, 2016:

Remote cameras have been deployed at 97 sites with 13 of 16 common carnivore species in Minnesota identified. We have obtained about 1 picture event every 2.25 camera-days. Species not detected to date include the four uncommon species that we would not expect to be present (mountain lion, wolverine, spotted skunk, and least weasel). The other carnivore species not detected are the river otter (not seen because deployments have been terrestrial), badger (present in areas sampled, but lower densities), and long-tailed weasel (not expected or only at low densities in the regions we have deployed cameras).

The protocol for collection of genetic samples was revised slightly. The general technique remains the same, with tubes with wire brushes for smaller carnivore species to walk through, and cable wire strands set up for larger carnivore species. The small tube collectors have become slightly less efficient, with 1 sample collected per 26 snare-days. The cable wire strands for obtaining large carnivore samples were also less efficient over the summer, with about 1 sample collected per 100 snare-days. The difference between larger and smaller carnivores is expected, because larger carnivores have lower population densities. A possible explanation for the lower efficiency in summer is that there is more prey available than in the winter months, but this can only be confirmed with longer term sampling. We will continue deployment of trail cameras and genetic sampling devices in Fall 2016.

Project Status as of December 31, 2016:

We have continued with the developed field protocol for detection of carnivore species using remote camera surveys with almost 10,000 camera-nights through 12/31/2016. There have been almost 3,700 animals captured, of which about 1,200 are carnivores. We remain at 13 of 17 carnivore species detected. We have also continued with collecting genetic material. Hair snare tube traps, cable restraint hair snares, and wolf scat have been collected.

The project has also encountered some obstacles that have delayed completion and will result in a request for an extension. The biggest loss was the genetics co-investigator is no longer working on the project. In addition,

the technician who had worked on camera deployment moved on to graduate school at the University of Wyoming, and the graduate student on this project chose to leave graduate school. Throughout these changes we have kept the field sampling going with relatively few breaks. However, this has resulted in delays in some of the data processing, genetics analysis, and website development.

Completion dates will need to be revised. I am working on a revised schedule that would allow completion of project goals with a 6-month extension and would like to submit an amendment request for this project. Specifically, I have obtained a new genetics collaborator here at NRRI, and I have a new technician to oversee camera deployment and picture processing. I have also hired 3 new UMD undergraduates to do the picture processing. The one personnel gap that I still have is website development, and I will fill that gap in February. These issues have also resulted in less money being spent on the project, 56% of funds are left although the original 2 year period is 75% completed.

Amendment Request February 14, 2017:

I would like to request to change the completion date for this project from 6/30/2017 to 6/30/2018. The reason for this request is primarily several personnel changes on the project that will make it very difficult to finish project activities by the original end date. No changes in the budget are required.

Amendment Approved: May 30, 2017.

Project Status as of June 30, 2017:

Progress has been made on each aspect of the project: analysis of photographs, techniques reports, genetics, and the project website.

Photographs: We have analyzed pictures from 124 of 127 camera deployments, with almost 13,000 camera nights represented. During those deployments we have almost 9,000 animal events, and 2,600 events with carnivores.

Techniques reports. We have written 3 Technical Reports describing trail camera methods, hair snare tube trap methods, and cable restraint hair snare methods. These reports describe methods used for each activity, and initial results in terms of efficiency of sample collection.

Genetics. We have successfully extracted DNA from wolf scats collected this winter. The DNA is being extracted in the laboratory of Dr. Chanlan Chun at the NRRI. The remainder of the wolf scats are being analyzed in summer 2017.

Project website. We have created all of the content for the initial website release. This includes 54 text files and about 140 pictures and figures that provide background information on carnivores in Minnesota. We will have this website available to the public by July 15, 2017. Files are currently being converted from MS-Word format to HTML. The development website URL is https://champ.d.umn.edu/mc. We will be adding pages and pictures and checking links with a revised goal of public release no later than August 1, 2017.

Project Status as of December 31, 2017:

Progress has been made on each aspect of the project: analysis of photographs, techniques reports, genetics, and the project website.

Photographs: We recorded about 800,000 pictures with 3,143 camera-events of 14 carnivore species from 111 camera deployments. Pictures of wolves were taken at almost 2/3 of camera sites, more than any other carnivore species.

Techniques reports. The 3 Technical Reports (trail camera methods, hair snare tube trap methods, and cable restraint hair snare methods) are complete except for the discussion. These reports describe methods used for each activity, and initial results in terms of efficiency of sample collection.

Genetics. We have successfully extracted DNA from wolf scats collected this winter. The DNA was extracted in the laboratory of Dr. Chanlan Chun at the NRRI.

Project website. We have created all of the content for the initial website release. The first website is at this URL: https://www.nrri.umn.edu/mc. We will be editing this content and releasing the final version on the UMD website: https://champ.d.umn.edu/mc with a redirection from the first website hosted at NRRI.

Occupancy modelling. With the completion of the picture analysis, we created the data set that is to be used for occupancy modelling. The goal of Occupancy modelling is to estimate a probability of presence for each species that we obtained enough pictures of, based on habitat variables in the surrounding habitat.

Project Status as of April 11, 2018:

Progress has been made on each aspect of the project: analysis of photographs, techniques reports, genetics, and the project website.

Photographs: We worked through about 500,000 trail camera pictures that provide a historical record of carnivore presence and distribution. We will use these results to test occupancy model. We also deployed additional trail cameras and are processing those pictures.

Techniques reports. The 3 Technical Reports (trail camera methods, hair snare tube trap methods, and cable restraint hair snare methods) are complete except for the discussion section. These reports describe methods used for each activity, and initial results in terms of efficiency of sample collection.

Genetics. We have finished a draft Technical Report of the genetic analysis at NRRI.

Project website. We have continued editing content for the website at this URL: https://www.nrri.umn.edu/mc. The original site is still online while we edit the UMD website version: https://champ.d.umn.edu/mc.

Occupancy modelling. The focus over the past 3 months has been occupancy modelling. A paper for peer review is written through the Results section.

Overall Project Outcomes and Results:

Minnesota has 20 carnivore species, 3 of which are very rare. Current monitoring methods of summer scent station surveys, winter track surveys, and population modelling could be complemented by camera traps and genetic DNA analysis. We used camera traps to obtain 3,400 images of carnivores over 12,000 camera-nights. American marten, fisher, short-tailed weasel, wolf, red fox, and gray fox were most frequently photographed. Occupancy analysis showed habitats used by each of these species. Mark-recapture estimation of population size was not possible. Camera traps could include significant public involvement, as is being done by the Wisconsin DNR. A second outcome of camera trap data is testing a Random Encounter Model to determine if population densities can be estimated without identifying individuals. We implemented sampling protocols to obtain hairs non-invasively from weasels and larger carnivores. Hair collection was less efficient than camera traps. Wolf scat collection in snow was unpredictable. However, DNA analysis identified individuals in the collected samples. Hair and scat collection is technically feasible but logistically difficult to implement.

A consistent conclusion from genetic sampling protocols is that the cost to obtain and analyze genetic samples, at present, would make it difficult to implement a mark/recapture population estimate for management on a large spatial scale. We did not fully expend the ENRTF funding because genetics collaborators were fully occupied with their own research. One tangible outcome of this project is that a genetic collaborator with time to do the analysis is critical.

The Minnesota Carnivore website has descriptions, pictures from the camera trap project, and historical harvest data in Minnesota and adjoining jurisdictions. The website will be updated periodically to provide new information—it is the only Minnesota-specific Carnivore website available. In addition, we will finish 4 Technical Reports and a peer-reviewed paper on occupancy modelling in Fall 2018.

IV. PROJECT ACTIVITIES AND OUTCOMES:

ACTIVITY 1: Determine Population Sizes of Carnivores

Description: Activity 1 will be divided into three steps.

First, we will develop a protocol for field collection of samples for DNA extraction and analysis from several carnivore species, in addition to using samples already available for some species. The protocol will identify three different groups of carnivores which break down somewhat into small, medium, and large body sizes. We will vary detection methods for each group to account for differences in average home-range size.

The second step in determining the population sizes of carnivores will be to conduct genetic surveys in a noninvasive manner. This method is especially effective at monitoring species that are difficult or expensive to monitor using other methods such as radio-telemetry. Studies involving non-invasive genetic sampling usually acquire DNA from hair or scat (feces) that are collected systematically or opportunistically throughout a study area. This genetic material can be used to determine the species, gender, and individual identity of its source. We will collect both scat and hair samples throughout the study area.

Samples collected will be analyzed genetically and then statistical analyses will enable us to estimate genetic effective and census population sizes. The genetic effective population size is an important measure of the amount of genetic variation in a species, as species with large census population sizes may still have low genetic variation. In addition, genetic data sampled throughout species' ranges in the state will allow us to infer patterns of migration and dispersal by tracking the spread of genetic variants.

Genetic and camera-based estimates of population size will provide complementary information about the status of each species in Minnesota. Pictures of carnivores will enable us to estimate population size and perform occupancy modeling. Museum records, harvest records, DNA data, and other reports will be used to estimate species distributions and abundances across time, providing a comprehensive picture of MN carnivore species population sizes and trends from a genetic and organismal perspective.

ENRTF Budget: \$ 200,000

Amount Spent: \$ 159,506

Balance: \$ 40,494

Outcome	Completion Date
1. Genetic estimate of wolf population size from mark-recapture in Year 1	June 30, 2018
2. Population size estimate from mark-recapture completed for selected carnivores	June 30, 2018
3. Genetic effective population size and dispersal events identified for selected carnivores	June 30, 2018
4. Report summarizing census and genetic effective population size estimates and trends	June 30, 2018

Activity Status as of December 31, 2015:

We have developed a field protocol for remote camera based occupancy modeling. We have deployed 46 cameras in St. Louis and Lake counties for 2,428 camera-days through 12/30/2015. Twenty-four of these deployments are now complete with 89,768 photographs. There are 10 carnivore species in these photographs. We have developed a data management protocol for remote camera photographs, and have processed 11 of the 24 completed remote camera deployments, documenting 527 camera visits by animals in 260 camera-days. Early indications are that we can estimate occupancy for at least 6 carnivore species using remote cameras.

We have also identified candidate species for genetic work, and have developed protocols for genetic sample collection, processing, and storage. We have developed field protocols for non-invasive DNA collection for eight species (least weasel, short-tailed weasel, long-tailed weasel, mink, otter, bobcat, lynx, and wolf). We have begun collecting weasel and wolf genetic samples. We will increase the number of samples as we fine-tune the weasel hair snag design, deploy hair snares, and collect scat in snow. We expect to develop techniques to estimate occupancy for 5 additional species using genetic sampling techniques.

Activity Status as of June 30, 2016:

We have continued with the developed field protocol for carnivore species using remote cameras. We have deployed 97 cameras in St. Louis and Lake counties for 4,721 camera-days. Sixty-four of these deployments are now complete. There still have been 13 carnivore species observed on these cameras. We have processed 23 of the 64 completed remote camera deployments, with 1,598 camera visits by animals. Camera deployments will continue through the summer.

We have also developed non-invasive hair snare traps to target least, short-tailed, and long-tailed weasels. Traps are constructed using polyvinyl chloride (PVC) tubing 5.1 cm and 7.6 cm in diameter. Each trap is cut to a length of 30.5 cm. One wire brush for collecting hairs is mounted on each end of the small diameter trap. We are using two wire brushes on each end of the large diameter trap. Each trap is baited using commercial trapping lures and a piece of hunter donated white-tail deer liver. Traps are attached to down trees or logs using a 2.5 cm screw on each end of the PVC tube.

Field methods using non-invasive genetic sampling have been successful in collecting genetic material that will be used to estimate carnivore species. We have developed hair snares from cable restraint snares to target larger carnivores (red fox, gray fox, bobcat, wolf, and black bear) using 7 x 7 stranded cable. Hair samples are collected on loose wires that are created by cutting individual cable strands approximately 1 cm in length along the last 60 cm of the snare loop. As the animal walks through the snare wire, hairs making contact with the wire strands are shed from the animal and are retained on the hair snare trap. The locking mechanism of the snare

has been replaced with a small paper clip so the animal just walks through and brushes against the wire. Commercial trapping lures are placed at the trap location to attract carnivores.

We have used the hair snares to collect genetic material in St. Louis and Lake counties. The hair snares target 6 species (least weasel, short-tailed weasel, long-tailed weasel, red fox, bobcat, lynx, and wolf). The weasel hair snare traps have captured 95 hair samples during 1,769 snare days in St. Louis county, about 1 sample per 20 snare-days. Cable restraint hair snare traps designed for larger carnivores have captured 9 hair samples in 740 snare-days, about 1 sample per 75 snare-days. We will continue to survey new locations using the developed non-invasive genetic sampling techniques for weasels and large carnivores in summer 2016.

Activity Status as of September 30, 2016:

Camera trap surveys for carnivores using remote trail cameras have continued in St. Louis and Lake County. We have deployed 118 cameras for 6,672 camera-days. Ninety-three of these deployments are now complete. We have processed 39 of the 93 completed remote camera deployments, with 2,962 camera visits by animals. As of October, 2016 there still have been 13 carnivore species documented (Table 1). We will continue deploying cameras through 2016.

Common name	Latin name	Events
Coyote	Canis latrans	17
Gray wolf	Canis lupus	93
Canada lynx	Lynx canadensis	13
Bobcat	Lynx rufus	7
American marten	Martes americana	375
Fisher	Martes pennanti	95
Striped skunk	Mephitis mephitis	29
Ermine	Mustela erminea	176
Mink	Neovison vison	2
Raccoon	Procyon lotor	7
Black bear	Ursus americanus	7
Gray fox	Urocyon cinereoargenteus	76
Red fox	Vulpes vulpes	79

Table 1. Number of carnivores captured on remote trail cameras in St. Louis, Lake, and Cook County as of October, 2016.

Species not detected to date include the four uncommon species that we would not expect to be present (mountain lion, wolverine, spotted skunk, and least weasel). The other carnivore species not detected are the river otter (not seen because deployments have been terrestrial), badger (present in areas sampled, but lower densities), and long-tailed weasel (not expected or only at low densities in the regions we have deployed cameras).

The weasel hair snare traps have captured 102 hair samples during 2,609 snare-days, a slightly lower success ratio of about 1 sample per 26 snare-days than we obtained in winter and spring 2016. The cable snares for larger carnivores have captures 24 hare samples in 2,410 snare-days, a slightly lower success ratio of about 1 sample per 100 snare-days than we obtained in winter and spring 2016. We have continued deploying hair snares of both types in Fall 2016.

Activity Status as of December 31, 2016:

We have continued with the developed field protocol for detection of carnivore species using remote camera surveys. We have deployed 127 cameras in St. Louis and Lake Counties for 9,573 camera-nights through 12/31/2016. One hundred and nine of these camera deployments have been completed. We have processed 60 of the completed remote camera surveys, with 3,697 camera visits by animals and 1,210 camera visits by carnivores. There have been 13 carnivore species observed on these cameras. We will continue to increase the number of locations surveyed using the developed field protocol for remote camera sampling of carnivore species in 2017.

We have also continued with collecting genetic material to estimate occupancy of carnivore species in St. Louis and Lake Counties. We have utilized non-invasive genetic sampling methods in effort to capture seven species (least weasel, short-tailed weasel, long-tailed weasel, gray fox, red fox, bobcat, lynx, wolf, and black bear). The weasel hair snare traps have captured 109 hair samples during 2,324 trap nights in St. Louis County. Cable restraint hair snare traps designed for larger carnivores have captured 25 hair samples in 2,800 trap nights. We will continue to increase the number of locations surveyed using the developed non-invasive genetic sampling techniques for weasels and larger carnivores in 2017. Scat collection surveys have resumed with snow present.

Activity Status as of June 30, 2017:

We have not deployed additional cameras, ending up with 127 cameras in St. Louis and Lake Counties for 12,989 camera-nights through 3/31/2017. We have processed 124 of the completed remote camera surveys, with 8,951 camera visits by animals and 2,616 camera visits by carnivores, with 3 trail camera surveys left to process. There have been 13 carnivore species observed on these cameras. After initial analysis of pictures we will redeploy cameras in Fall 2017.

We have successfully extracted DNA from wolf scats collected in winter on snow. This work is being done in the genetics laboratory of Dr. Chanlan Chun at the Natural Resources Research Institute. We are using standard methods with a Quiagen DNA extraction kit. The remaining wolf scats that have been collected are being extracted and analyzed in summer 2017.

Hairs collected using the hair snare collection technique with cable restraint and tube snares will be analyzed next, if we find that the efficiency of collection would justify using this technique to estimate population sizes of carnivore species in NE MN. Because of the collection efficiency (< 1 hair / 100 nights for cable restraint, 5 hairs / 100 nights for tube hair snares), it may be more economically feasible to obtain hair samples from harvested animals or other sources.

The content for the release 1 of the website has been fully developed. We include species descriptions for every Minnesota carnivore species, harvest trends for Minnesota and surrounding states and provinces, methods sections for trail camera, cable restraint hair snare, and tube hair snare techniques, a unique method of contrasting different species that are easily confused from pictures, species ranges, and other information. We have a URL set up and UMD students are assisting in converting the MS-Word format mockup of the website to HTML code in the Drupal content management system. The website will be released to the public with a revised goal of August 1, 2017.

Activity Status as of December 31, 2017:

Progress has been made on each aspect of the project: analysis of photographs, techniques reports, genetics, and the project website.

Photographs and cameras: In addition to finishing up the analysis of the original pictures, we deployed cameras in the field again. Some of these cameras were placed in the same locations that were previously used to test repeatability of detection. Others were placed within 1 mile of the previous deployments to test adjacency.

Techniques reports. We continued work on the Discussion section of each of the Technical reports.

Genetics. In addition to finishing DNA extraction, we have begun work on a written summary of the protocol and results.

Project website. We continued work on the initial website content, and began creating images that will make it possible for people to differentiate between different carnivore species based on a comparison of side-by-side pictures.

Occupancy modelling. We created the picture data set (animal presence) and we also created the data set on spatial characteristics (e.g., cover type, road density, distance to water, edge length) for each camera location. We then began doing the occupancy modelling on the American marten data set.

Activity Status as of April 11, 2018:

Progress has been made on each aspect of the project: analysis of photographs, techniques reports, genetics, and the project website.

Photographs: We worked through about 500,000 trail camera pictures that provide a historical record of carnivore presence and distribution. These camera deployments were mostly within the current study area. We also deployed additional trail cameras and are processing those pictures. Some of these deployments are in the same location that we used 10 to 12 years ago, and will provide data on changes in species presence.

Techniques reports. The 3 Technical Reports (trail camera methods, hair snare tube trap methods, and cable restraint hair snare methods) are complete except for the discussion section. These reports describe methods used for each activity, and initial results in terms of efficiency of sample collection and ability to use the method for analysis of different carnivore species.

Genetics. We have nearly finished a draft Technical Report of the genetic analysis done at NRRI.

Project website. We have continued editing content for the website at this URL: https://www.nrri.umn.edu/mc. The original site is still online while we edit the UMD website version: https://champ.d.umn.edu/mc.

Occupancy modelling. The focus over the past 3 months has been occupancy modelling. A paper for peer review is written through the Results section. This paper covers all carnivore species for which we obtained enough pictures to use occupancy modelling.

Final Report Summary:

This activity can be broken down into 3 main tasks: Camera traps, Genetic Sampling, and Website Development.

1. Camera Trap Analysis. Camera traps were successful in obtaining pictures of many carnivore species. In Phase 1 we deployed 127 cameras in St. Louis and Lake Counties for 12,213 usable camera-nights, 7,200 images of mammals, and 3,400 images of carnivores. Other camera deployments resulted in a total data set of 17,000 images of mammals, and 7,200 images of carnivores. Species most frequently photographed were American marten, fisher, short-tailed weasel, wolf, red fox, and gray fox. American marten were most common with about 30% of events, other frequently photographed species ranged from 5 to 15% of events. Occupancy analysis could be done on 6 species: marten and fisher were associated with conifer forests and increased canopy cover, wolves were more common at lower human impact and lower road densities, coyote and gray fox tended to be present in landscapes with more agriculture and human presence, and red fox occupancy was increased in mixed forest and wooded wetland cover types.

It was not possible to individually identify any carnivore species with enough certainty to calculate a markrecapture estimate of population size. Some individual wolves could be identified by coloration, and some bobcats by spotting pattern. However, we could not identify all individuals, and not all images of a species were of high enough quality to view spots. Camera traps can be used for an index of relative density, but camera traps could not be used to estimate population sizes on a large spatial scale. If animals were radio-collared or had some other identifying marks, camera traps could be used to estimate population sizes on a small spatial scale. One outcome of the analysis of camera trap pictures is that we are testing a Random Encounter Model (REM) to determine if deer population densities can be estimated without being able to identify individual deer. We are also evaluating the REM using radiotelemetry locations of deer.

2. Genetic sampling protocol. We developed and implemented sampling protocols using tube hair snares for weasels, cable restraint hair snares for larger carnivores, and scat collection for wolves. A consistent conclusion across the sampling protocols is that while we could obtain genetic samples, the cost to obtain and analyze those samples, at least at the current time, would make it difficult to implement a mark/recapture population estimate for management on a large spatial scale. A second complication, elaborated on below, is having a collaborator who can do genetic analyses, or developing the skills needed to do genetic analyses in house. The most appropriate use of hair sampling or scat sampling protocols at this time would be for localized analysis of population size.

A weasel tube hair snare is a PVC pipe with wire brushes that are used to snag hair. We collected 109 hair samples from tube hair snares over 2,128 trap nights, or 5 hairs collected per 100 trap nights. Weasels were also visible on the camera traps about 5 events / 100 trap nights. The second protocol developed used a cable restraint hair snare, which is a snare cable that has the locking device replaced with a paper clip. We collected 25 hair samples from the cable restraint hair snares over 3,020 trap nights. Cable restraints were much less successful than camera traps, with only 0.9 hairs collected per 100 trap nights, compared to 2.4 picture events per 100 trap nights. If efficiency of sample collection could not be increased, it would be necessary to deploy the cable restraint hair snares for 300,000 trap nights in order to obtain even 100 hairs for analysis.

Wolf scat collection. Wolf scat collection would be most applicable during the winter when snow is on the ground to prevent DNA degredation. We used snowmobiles to search for wolf scat on state trails in winter. We found that obtaining wolf scat was inconsistent. On some days we would obtain 20 wolf scats in 40 miles of snowmobiling, on other days we would find 0 wolf scats. Wolf scats were analyzed successfully to individual based on DNA identifications. To be efficient the scat collection protocol would likely require collecting wolf scats across a broad area, and because of the unpredicatable nature of finding wolf scats, it would likely involve either collection of scats in conjunction with other work, or developing a method for collection by citizen scientists.

 <u>3. Website and associated data. We developed a website with descriptions of each carnivore species in</u> <u>Minnesota, pictures from the camera trap project, historical harvest data in Minnesota and adjoining</u> <u>states and provinces, and also some comparative pictures for identification purposes. The website will</u> <u>be updated periodically to provide new information—it is the only Minnesota-specific Carnivore website</u> <u>available. Much information has been added since the Mammals of Minnesota book, and in addition</u> <u>there are new sources for carnivore data—for example the ENRTF funded project to digitize the Bell</u> <u>Museum of Natural History collections.</u> The Minnesota Carnivore website is currently located at <u>https://champ.d.umn.edu/mc. It is being relocated to https://www.nrri.umn.edu/mc.</u> We did not fully expend the ENRTF funding. The field data collection aspect of the project was successful, but a genetic collaborator is needed. A major difficulty that would be encountered if genetic work were to be contemplated in the future is identifying a genetic collaborator. We had one genetics faculty who had to drop off of the project, and while we identified a second collaborator, that collaborator was busy with the own work and not able to analyze all of the genetic samples that we collected.

V. DISSEMINATION:

Description: We will create a website to distribute information to the public, but this will be done after the project starts. The website will be modelled after other websites we maintain (e.g., www.nrri.umn.edu/moose).

In addition, we will also prepare and submit papers for publication in peer-reviewed journals, and present our results at regional and national scientific meetings (using other funds for travel outside of Minnesota). All genetic data will be submitted to the Dryad Digital Repository (http://datadryad.org/), a freely accessible non-profit database, upon publication.

We will also probably have periodic contact with print and broadcast media, given the nature of the project.

Status as of December 31, 2015:

We began work on website design in the first 6 months of the project. This included text and selecting pictures for use on the website. The website will become public later in 2016 with implementation of the Drupal website management system at UMD.

Status as of June 30, 2016:

No changes other than picture selection as the trail camera pictures were processed.

Status as of Sepember 30, 2016:

We continued developing page content for the website. Trail cameras were deployed at hair snare sites to document animals moving through the hair-snaring equipment. These pages will be part of the website, in addition to highlights showing different species. Website will become public in December 2016.

Status as of February 2, 2017:

Because of the personnel issues summarized in the Project Status Update above, I failed to accomplish the goal of making the website available to the public. I have a revised goal of posting the initial website by March 1, 2017. This is only 2 weeks away, but I believe with the current status of the project that it is essential that we have an initial website online by then.

Status as of June 30, 2017:

We have prepared 4 products for release as information dissemination. These products are currently undergoing final review:

- 1. NRRI Technical Report on cable restraint hair snares
- 2. NRRI Technical Report on tube hair snares
- 3. NRRI Technical Report on camera trap protocol

4. Minnesota Carnivore website

Status as of December 31, 2017:

A short sidebar about the project appeared in the Minnesota Conservation Volunteer in the January-February 2018 issue. The URL is https://www.dnr.state.mn.us/mcvmagazine/issues/2018/jan-feb/game-cameras.html. Although I asked, the editors did not keep in the credit to the ENRTF.

Status as of April 11, 2018:

There were two presentations that incorporated carnivore data collected on this project:

Moen, R. A. and Windels, S. K. *Climate-driven changes in future Minnesota mammal species*. Given at the 2018 Forestry and Wildlife Research Review, Cloquet, Minnesota, United States on January 11, 2018. This meeting was organized by the UM/Sustainable Forests Education Cooperative. Attendance was about 100 individuals, primarily resource managers from NE MN.

Moen, R. A. and Windels, S. K. *Climate-driven changes in future Minnesota mammal species*. Given at the 2018 Annual Meeting of the Minnesota Chapter of of The Wildlife Society, St. Cloud, Minnesota, United States on February 14, 2018. This was an improved version of the presentation in Cloquet, about 50 Wildlife Biologists were present from throughout the state.

Final Report Summary:

We are currently completing Technical Reports and a peer-reviewed paper. We expect these papers to be finished in Fall 2018 and will forward to LCCMR as the reports are completed. The occupancy analysis paper will be forwarded when it is published.

- 1. <u>Technical reports summarizing the entire project.</u>
 - a. <u>Camera Trapping</u>: Moen, R. and B. Houck. 2018. Monitoring Carnivore Populations in Northeast Minnesota with Camera Traps. NRRI Technical Report No. NRRI/TR-2018/44. University of Minnesota Duluth.
 - b. Weasel tube hair snares: Houck, B. and R. Moen. 2018. Use of Tube Hair Snares to Detect Weasels in Minnesota. NRRI Technical Report No. NRRI/TR-2018/45. University of Minnesota Duluth.
 - c. <u>Cable-Restraint hair snares</u>: Houck, B. and R. Moen. 2018. Use of Single-Capture Hair Snares to Detect Carnivores in Minnesota. NRRI Technical Report No. NRRI/TR-2018/43. University of Minnesota Duluth.
 - d. Wolf Scat Collection and Genetic Analysis. Moen, R. 2018. Genetic Analysis of Wolf Scats Collected from Snow. NRRI Technical Report No. NRRI/TR-2018/51. University of Minnesota Duluth.
- 2. <u>Occupancy modelling manuscript. Houck, B. and R. Moen. 2018. Occupancy modelling of carnivores in</u> northeastern Minnesota from camera trap data. Manuscript to be submitted for peer review.
- 3. <u>The Minnesota Carnivore website is currently located at https://champ.d.umn.edu/mc. It is being</u> relocated to https://www.nrri.umn.edu/mc.

VI. PROJECT BUDGET SUMMARY:

A. ENRTF Budget Overview:

Budget Category	\$ Amount	Overview Explanation
Personnel:	\$ 149,225	1 project manager (66.4% salary, 33.6%
		benefits) at 6% FTE each year for 2 years; 1 field
		and lab technician (63.2% salary, 36.8%
		benefits)at 25% FTE each year for 2 years; 1
		graduate research assistant (AY-84.3% salary,
		15.7% benefits, and \$17.32/hr tuition
		reimbursement cost; SUM-76.9% salary, 23.1%
		benefits, no tuition costs) at 50% FTE (academic
		year)and 50% FTE (summer) each year for 2
		years; and 1 undergraduate research assistant
		(100% salary, 0% benefits)at 50% FTE each year
		for 2 years. Allocation of effort among
		personnel categories are estimates that may be
		adjusted to best meet project objectives.
Professional/Technical/Service Contracts:	\$0	n/a
Equipment/Tools/Supplies:	\$ 33 <i>,</i> 850	Trail cameras, genetic analysis supplies, and
		field supplies such as batteries, mosquito
		repellent, and flagging
Capital Expenditures over \$5,000:	\$ O	n/a
Fee Title Acquisition:	\$ O	n/a
Easement Acquisition:	\$0	n/a
Professional Services for Acquisition:	\$0	n/a
Printing:	\$0	n/a
Travel Expenses in MN:	\$ 16,925	In-state travel to/from field sites to deploy and
		maintain cameras and collect material for
		genetic analysis
Other:	\$0	n/a
TOTAL ENRTF BUDGET:	\$ 200,000	

Explanation of Use of Classified Staff: N/A

Explanation of Capital Expenditures Greater Than \$5,000: N/A

Number of Full-time Equivalents (FTE) Directly Funded with this ENRTF Appropriation: 4.6 FTEs

Number of Full-time Equivalents (FTE) Estimated to Be Funded through Contracts with this ENRTF Appropriation: N/A

B. Other Funds:

	\$ Amount	\$ Amount	
Source of Funds	Proposed	Spent	Use of Other Funds
Non-state			
Ron Moen salary/fringe (cost-share)	\$ 12,900	\$ <u>7,249</u>	
State			
Foregone by UMN ICR funding	\$ 88,600	\$ <u>88,600</u>	52% indirect costs (excluding graduate
			fringe)
TOTAL OTHER FUNDS:	\$ 101,500	\$ <u>95,849</u>	

VII. PROJECT STRATEGY:

A. Project Partners:

Other individuals will assist or provide advice on parts of this project.

- **Dr. John Erb (**MN DNR) will provide input and some samples, with an intent to make long-term use of techniques developed in this proposal for DNR management purposes.
- **Dr. Steve Windels** (Voyageurs National Park) will cooperate on the trail camera project and genetic sampling. He already has cameras deployed to test a mark-recapture method of estimating presence and abundance of carnivores in Voyageurs.

B. Project Impact and Long-term Strategy:

Our long term strategy is to develop the ability to do genetic analysis of wildlife populations in Minnesota with in-house expertise. Specific applications will vary from an independent estimate of wolf population size to proactive preparation for future wildlife issues. For example, wildlife diseases and parasites may become important if they are transmitted to humans or companion animals like dogs and cats.

We envision this as a project to develop techniques that we will initially focus on carnivore species in northeastern Minnesota. As techniques are successfully developed, we will expand to other parts of the state. Over the long-term, a genetic approach should decrease uncertainty in population estimates and even enable analysis of trends over time.

C. Funding History:

Funding Source and Use of Funds	Funding Timeframe	\$ Amount
We have been developing the genetics and the field sampling	2011 to Present	\$10,000
components of this project over the past 2 years. Wolf scat and		estimated.
tissue samples from DNR wolf collar program are being		
analyzed genetically by an M.S. student, and we have hundreds		
of thousands of trail camera images that can be used as a		
resource from past research.		
ENRTF – M.L. 2013, Ch. 52, Sec. 2, Subd. 04g. This project on	July 1, 2013 – June 30, 2016	\$ 200,000
moose browsing habitat is totally unrelated to the Carnivore		
project in this work plan.		
		\$ 200,000

VIII. FEE TITLE ACQUISITION/CONSERVATION EASEMENT/RESTORATION REQUIREMENTS: A. Parcel List: N/A

N/A

B. Acquisition/Restoration Information:

N/A

IX. VISUAL COMPONENT or MAP(S):

X. RESEARCH ADDENDUM:

See attached Research addendum.

XI. REPORTING REQUIREMENTS:

Periodic work plan status update reports will be submitted no later than December 31, 2015, June 30 2016, December 31, 2016, June 30, 2017, and December 31, 2017. A final report and associated products will be submitted between by August 15, 2018.

Environment and Natural Resources Trust Fund M.L. 2015 Project Budget Final Report

Project Title: Genetic and Camera Techniques to Estimate Carnivore Populations

Legal Citation: M.L. 2015, Chp. 76, Sec. 2, Subd. 03a

Project Manager: Ron Moen

Organization: Natural Resources Research Institute, University of Minnesota Duluth

M.L. 2015 ENRTF Appropriation: \$ 200,000

Project Length and Completion Date: 3 years, June 30, 2018

Date of Report: November 13, 2018



	Activity 1		Activity 1	TOTAL	TOTAL
ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Budget	Amount Spent	Balance	BUDGET	BALANCE
BUDGET ITEM	Determine Population Sizes of Carnivores.				
Personnel (Wages and Benefits) Overall	\$149,225	\$134,529	\$14,696	\$149,225	\$14,696
Project Manager: (66.4% salary, 33.6% benefits); 6% FTE each year for 2 years. Est. total \$10,945					
Co-Investigator: (66.4% salary, 33.6% benefits); 8% FTE each year for 2 years. Est. total \$10,985					
Field & Lab Technician: \$32,000 (63.2% salary, 36.8% benefits); 25% FTE each year for 2 years. Est. total \$19,945					
Graduate Research Assistant: \$46,000 (84.3% salary, 15.7% benefits, and \$17.32/hr tuition reimbursement cost; summer-76.9% salary, 23.1% benefits, no tuition costs); Academic Year-50% FTE, Summer-50% FTE each year for 2 years. Est. total \$83,880					
Undergraduate Research Assistant: \$20,800 (100% salary, 0% benefits); Academic Year-50% FTE, and Summer-50% FTE each year for 2 years. Est. total \$23,470					
Equipment/Tools/Supplies					
Trail cameras (20 estimated @ \$339 each), purchased in year 1	\$6,770	\$6,754	\$17	\$6,770	\$17
Genetic analysis supplies (\$12,694 per year)	\$25,388	\$4,052	\$21,336	\$25,388	\$21,336
Field supplies (\$846 per year) (Batteries, mosquito repellent, flagging, etc.)	\$1,692	\$1,687	\$5	\$1,692	\$5
Travel expenses in Minnesota					
In-state travel to/from field sites (\$10,000 per year). Cameras will need to be deployed and maintained at sites in NE Minnesota and material will be collected for genetic analysis, requiring the use of University vehicles. Some trips will involve longer- distance travel and require overnight expenses (camping or motel) and food expenses.	\$16,925	\$12,484	\$4,441	\$16,925	\$4,441
COLUMN TOTAL	\$200,000	\$159,506	\$40,494	\$200,000	\$40,494

Historical northern long-eared bat occurrence in Minnesota based on acoustic surveys



Ron Moen, Ph.D. Morgan Swingen, M.S.

Report Number: NRRI/TR-2018/40 Release 1.0

Natural Resources Research Institute

UNIVERSITY OF MINNESOTA DULUTH

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Summary

Although long thought to exist throughout the forested region of Minnesota, occurrence records for northern long-eared bats (Myotis septentrionalis) were historically based on winter hibernacula records and sporadic summer observations. The ability to record and identify bats by their echolocation calls allowed scientists to more systematically survey for bats in Minnesota beginning in the 2000s; however, these data were not compiled in a central database. With the arrival of whitenose syndrome in Minnesota and the federal listing of the northern long-eared bat in 2015 as threatened under the Endangered Species Act, the need for a more detailed and current distribution map for this species was evident. In this report, we summarize the occurrence records for northern long-eared bats based on specimens collected, existing acoustic survey data from various sources collected prior to 2015, and acoustic survey data collected from 2015 to 2017. Northern long-eared bats do appear to be distributed throughout the forested region of Minnesota. Presence has been documented in the northern half of the state, surrounding the Twin Cities metropolitan area, and in the southeast corner of the state. Detection of the northern long-eared bat in almost every attempt suggests that the species is also present in unsurveyed regions of the forested regions of the state, although it is less common than the little brown bat (M. lucifugus), especially after white-nose syndrome has led to mortalities in Minnesota.

Cover photograph:	The cover shows an Anabat detector enclosed in a protective box with a
	reflector plate that would reflect bat calls into the microphone.

Author Affiliations: RM and MS – UMD Natural Resources Research Institute

Funder: Environment and Natural Resources Trust Fund

Report Number: NRRI/TR-2018/40 Release 1.0

Please contact authors before citing, as manuscripts are in review and in preparation.

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Introduction

Northern long-eared bats (*Myotis septentrionalis*) are small bats that hibernate in caves and mines in the winter. Other names for the northern long-eared bat in the historical literature include *Vespertilio gryphus* var. *septentrionalis* and *Myotis keeni septentrionalis* (Caceres and Barclay 2000). In some older literature, the northern long-eared bat is referred to as *Myotis subulatus* (Jackson 1961). The northern long-eared bat ranges throughout much of eastern North America based on the species distribution map, although specimen records in Minnesota are relatively scarce (Fig. 1).

Figure 1. Distribution of northern long-eared bat in North America (Caceres and Barclay 2000) and in Minnesota (Hazard 1982). The North American distribution map was originally published in a Mammalian Species account, and the Minnesota map was created by the Bemidji State University Biology Department and originally published as Map 13 in *The Mammals of Minnesota* (Hazard 1982).



Older references often indicate that the northern long-eared bat is relatively common in Minnesota forests in the summer, although the little brown bat (*Myotis lucifugus*) appears to be the most common *Myotis* species in Minnesota. Occurrence records of *Myotis* species come from winter hibernacula records and from occasional specimens captured in the months when bats are not in hibernacula. The little brown bat has more historical records because it often roosts in buildings during the summer, while the northern long-eared bat usually roosts in trees and therefore is less likely to be encountered by humans.

The first publications on mammals in Minnesota did not list the northern long-eared bat as present. For example, Herrick (1892) listed only the little brown bat in the *Myotis* genus. The northern long-eared bat was likely present; this is probably another instance of the little brown bat being easier to find because it roosts in buildings in the summer.

The first published indication of the northern long-eared bat in Minnesota was in a list of Minnesota mammals compiled by University of Minnesota professor C.E. Johnson in 1916, in which the range of the northern long-eared bat was described as including the "entire state" (Johnson 1916). The northern long-eared bat was described as very common throughout Itasca County in north central Minnesota in 1919 (Cahn 1921), although Cahn called it *M. subulatus* in that publication. Cahn also listed the little brown bat as present but did not say that it was either common or very common.

In a second compilation of the Mammals of Minnesota, the little brown bat was considered the most common *Myotis* bat in the summer, although hibernacula locations appeared to be unknown because winter locations were not discussed (Surber 1932). The northern long-eared bat was described as having only a local distribution in Minnesota, although it could be found throughout the state. One known specimen from Elk River in Sherburne county was referenced.

There are few published records of the northern long-eared bat from the mid 1900s. A few northern long-eared bats were found hibernating in caves during winter surveys in 1940–1941 in Nicollet, Goodhue, Fillmore, and Wabasha counties (Rysgaard 1942). Rysgaard indicates the northern long-eared bat is thought to be relatively common throughout Minnesota, although he also says that it is rarely found in hibernacula compared to other bat species. Other counties with documented presence of the northern long-eared bat included Clearwater, Itasca, St. Louis, Lake, Pine, Sherburne, and Stearns counties. Northern long-eared bats were found hibernating in sewers in St. Cloud, MN in 1952 (Goehring 1954).

In a third compilation of the mammals of Minnesota by Gunderson and Beer in 1953, the theme of the little brown bat being more common than the northern long-eared bat continues. The little brown bat was documented present in 19 counties in Minnesota, while the northern long-eared bat was listed as present in 7 counties in Gunderson and Beer's Mammals of Minnesota. The authors again indicate that the northern long-eared bat is more common than specimen records indicate, although also saying that in hibernacula the little brown bat is much more common.

In 1982, Bemidji State University biology professor Evan B. Hazard published another compilation of the mammals of Minnesota, with maps based on specimen records at the township level (Fig. 1). Counties with northern long-eared bat presence in Fig. 1 include Cass, Cook, Sherburne, and Ramsey. Several of the counties listed in earlier references (e.g., Rysgaard 1942, Goehring 1954) are not included because specimens were not associated with those observations. As in Gunderson and Beer (1953), the little brown bat was listed as present in many more counties than the northern long-eared bat in Hazard's book (30 counties for the little brown bat compared to 4 counties for the northern long-eared bat).

A literature review and additional surveys for Minnesota bat species were conducted by Gerda Nordquist and Elmer Birney in the early 1980s, leading to updated distribution maps which included known museum specimens, literature records, and observations from summer and winter field surveys (Fig. 2). In their literature review the little brown bat was documented in 55 counties, compared to 15 counties for the northern long-eared bat. Several hibernacula were identified, including the largest known hibernating populations of the little brown bat and the northern long-eared bat in Minnesota, at the Soudan Underground Mine in St. Louis County (Nordquist and Birney 1985).

Figure 2. Maps of known northern long-eared bat winter (left) and summer (right) distribution records in Minnesota as of 1985. Circles appear in counties for which records of northern long-eared bats occur, and do not indicate exact locations. Maps in Figure 2 were originally published as Figure 3 in the report "Distribution and Status of Bats in Minnesota" (Nordquist and Birney 1985).



Most of the earlier records of bat presence in Minnesota are from hibernacula, captures of bats in summer roosts, or specimens obtained incidentally. It became easier to document presence of bat species during the summer when the technology to record and identify the ultrasonic calls of bats became available to field biologists in the early 1980s (Fenton and Bell 1981). Acoustic surveys for bats were first conducted by the Minnesota Department of Natural Resources (MN DNR) in Minnesota in the early 2000s. Additional survey work has been conducted by state and federal agencies, universities, and private consulting companies. Some of these projects were published in peer-reviewed literature or as theses; others are only present in gray literature or are unavailable.

Ultrasonic recording technology has advanced greatly in the last decade, but even now not every call that is recorded can be identified to species. Early acoustic detectors recorded data in zero-crossing (ZC) format, a format which stored a limited amount of acoustic information compactly due to data storage limitations. More recently, acoustic detectors that record data in full-spectrum (FS) formats were developed. The FS format stores a greater amount of information about each call, which can make it easier to identify bat species.

The echolocation calls of bat species vary depending on their body size and foraging strategy. Minnesota's seven bat species make either low-frequency calls (hoary bat (*Lasiurus cinereus*), big brown bat (*Eptesicus fuscus*), silver-haired bat (*Lasionycteris noctivagans*)) or high-frequency calls (northern long-eared bat, little brown bat, tricolored bat (*Pipistrellus subflavus*), eastern red bat (*Lasiurus borealis*)). Low-frequency calls and high-frequency calls are easily differentiated, but it can be difficult to assign calls to a species within either the low-frequency or the high-frequency group. As might be expected, the calls of some bats within the same genus, such as the northern long-eared bat and the little brown bat, can be difficult to assign to a species. Northern long-eared bats and little brown bats are particularly difficult to distinguish due to the overlapping range of many call characteristics. In addition, for high frequency bats, some little brown bat and eastern red bat calls have similar characteristics. For low-frequency bats, big brown bat and silver-haired bat calls are very similar. Examples of the calls made by different species and additional discussion of identification of bat calls to species are in Swingen et al. (2018a).

Since 2006, cave-hibernating bat populations in the United States have declined sharply from whitenose syndrome (WNS). First documented in New York state, WNS has spread westward to 32 U.S. states and 7 Canadian provinces, killing millions of bats (U.S. Fish and Wildlife Service 2012). The northern long-eared bat is particularly susceptible to WNS, with declines of 90–100% in many eastern U.S. hibernacula (Turner et al. 2011). WNS was first confirmed in Minnesota in 2016, and subsequent winter surveys have confirmed decreasing numbers of bats (MN DNR 2016, 2017). When the northern long-eared bat was listed as Threatened under the Endangered Species Act in 2015, it became necessary to increase understanding of the distribution of this species in Minnesota. We used historic locations reviewed above, records downloaded from the Global Biodiversity Information Facility, the Minnesota Biodiversity Atlas, and available acoustic data to create an updated map of northern long-eared bat detections.

Methods

We compiled bat acoustic data collected in Minnesota by various entities prior to 2015. We attempted to identify all potential sources of bat acoustic data, including state agencies, federal agencies, universities, private consulting firms, and industry partners. If the original data was available, it was obtained in addition to a summary of the dataset and/or file identifications.

If the files were identified to species by the original source or author, we used the results of the original analysis. If the files were not identified by the original source or author, and the original recording data was available to us, we analyzed the files using the software program Kaleidoscope Pro (version 4.0.4). Data were processed in Kaleidoscope using the "Moderate" setting, with the "Minnesota" set of candidate species:

Big brown bat	Eptesicus fuscus (EPFU)
Eastern red bat	Lasiurus borealis (LABO)
Hoary bat	Lasiurus cinereus (LACI)
Silver-haired bat	Lasionycteris noctivagans (LANO)
Little brown bat	Myotis lucifugus (MYLU)
Northern long-eared bat	Myotis septentrionalis (MYSE)
Tricolored bat	Perimyotis subflavus (PESU)

We did not analyze any of the acoustic data using the Sonobat software program, because most of the existing data was from zero-crossing detectors, which cannot be analyzed by Sonobat.

There are some legal filings for wind turbine projects that can be found with an internet search. The acoustic data has been collected for these projects, but the data presentation in earlier projects is limited to high-frequency and low-frequency bat groups. Because the calls are not differentiated to species, the high-frequency bat calls could be from the northern long-eared bat, the little brown bat, or the eastern red bat. An example of this type of analysis is Derby and Dahl (2008).

Other legal filings that were done after the northern long-eared bat was listed as threatened under the ESA could be used. One example of this type of project is the Palmers Creek project in Yellow Medicine County (MDOC 2018), in which northern long-eared bats were not found.

Results

We compiled acoustic data and results from 2003–2014 from 10 sources (Table 1). Data were from 208 passive surveys, 47 active surveys, and 13 driving transects located in 21 Minnesota counties. Sources included the Minnesota Biological Survey, U.S. Forest Service, University of Minnesota, and WEST Inc. environmental consulting company. Acoustic records that are not publicly available, such as an acoustic study for the proposed new route for the Line 3 pipeline project by Enbridge, Inc. (https://www.enbridge.com/Line3ReplacementProgram.aspx), are not included in Table 1.

Table 1. Sources of bat acoustic data collected in Minnesota compiled for this analysis. For data type, ZC = zero-crossing and FS = full-spectrum.

Source	Years Data Collected	# Locations Data Collected	# Files Recorded	Type of Data	Files Identified by Source?
UMD – NRRI	2009-2014	106	52,790	ZC	Yes/No
Superior National Forest	2009–2014	7^1	4,554	ZC	Yes (WEST)
Chippewa National Forest	2011–2014	5^1	3,283	FS	Yes (WEST)
MN DNR – Biological Survey	2003–2014	Unknown ²	Unknown ²	ZC	Yes
MN DNR/ MN DOT	2014	16	25,547	ZC	Yes
National Park Service	2003	3	1,488	ZC	Yes
UPM Blandin	2014	3	790	ZC	Yes
Camp Ripley Training Center	2006–2014	11 ³	4,834	ZC	No
Dixon (2012)		47		ZC	Yes ⁴
Carlton County ⁵	2016	2	1,450	FS	Yes
Total		259			

¹ These locations are all driving transects.

² MBS data include confirmed MYSE calls. Call file data is not available.

³ One of these 11 locations is a driving transect.

⁴ Did not differentiate between MYSE and MYLU.

⁵ Sichmeller and Hammond 2017.

There were 16 records for *Myotis septentrionalis* and *Myotis keenii* in the Global Biodiversity Information Facility (GBIF) and the Minnesota Biodiversity Atlas databases (GBIF.org 2018a, b). The GBIF database search for *Myotis septentrionalis* and *Myotis keenii* returned one record from Elk River and one record from St. Cloud (GBIF_1, GBIF_2). The Minnesota Biodiversity Atlas (MBA) of the Bell Museum of Natural History has 14 specimens from Minnesota that were collected from 1934 to 1983 (MBA 2018). Hazard, Gunderson and Beer, and Nordquist probably looked at some of these specimens from the Bell Museum to make their maps! Because the northern long-eared bat is listed as a threatened species, the location is only reported at the county level in the MBA. Counties included Cook, Goodhue, Hennepin, Nicollet, Ramsey, St. Louis, and Stearns, all of which had been reported in the earlier literature. As a result of the northern long-eared bat being listed under the ESA, the MN DNR maintains records of locations of known roost trees. These records are currently located in 28 counties spread throughout the forested area of Minnesota. For all of these analyses of northern long-eared bat presence, an important consideration of this map is that absence of a record does not mean that northern long-eared bats are not present.

Figure 3. Records of northern long-eared bat roost trees by township stored in the database maintained by the MN DNR. Records are current as of April 2018.



Our review of the publications, museum records, and recent acoustic datasets resulted in documentation of northern long-eared bat presence in 38 of the 88 Minnesota Counties (Table 2). The main outcome of the synthesis of current knowledge of northern long-eared bat presence was to fill in some of the vacant spots present in earlier reviews.

Table 2. Counties in Minnesota with documented presence of northern long-eared bat. Column labels refer to publication date for Cahn (1921), Surber (1932), Rysgaard (1942), Goehring (1954), Gunderson and Beer (1953), Hazard (1982), Nordquist and Birney (1985). The column labelled "MNDNR" refers to known locations of northern long-eared bat roosts (Fig. 3), and the column labelled "T-1" refers to the sources compiled in Table 1. The column labelled "All" includes all counties in this table with northern long-eared bat presence documented.

County	1921	1932	1942	1954	1953	1982	1985	MNDNR	T-1	All
Aitkin								1		1
Anoka								1		1
Becker								1	1	1
Beltrami									1	1
Benton								1		1
Big Stone										
Blue Earth										
Brown										
Carlton								1	1	1
Carver								1		1
Cass						1	1	1		1
Chippewa										
Chisago										
Clay										
Clearwater				1				1	1	1
Cook						1	1	1	1	1
Cottonwood										
Crow Wing								1		1
Dakota								1		1
Dodge										
Douglas									1	1
Faribault										
Fillmore			1				1	1		1
Freeborn										
Goodhue			1		1		1	1		1
Grant										
Hennepin							1			1
Houston								1		1
Hubbard								1	1	1
Isanti								1		1
Itasca	1			1			1	1	1	1
Jackson										
Kanabec										
Kandiyohi										
Kittson										
Koochiching									1	1
Lac qui Parle										
Lake				1				1	1	1
Lake of the								1	1	1
Woods										

Table 2, Continued.

County	1921	1932	1942	1954	1953	1982	1985	MNDNR	Fig. 3	All
Le Sueur								1		1
Lincoln										
Lyon										
Mahnomen										
Marshall										
Martin										
McLeod										
Meeker										
Mille Lacs										
Morrison								1		1
Mower										
Murray										
Nicollet			1		1		1			1
Nobles										
Norman										
Olmsted										
Otter Tail										
Pennington										
Pine				1	1		1	1		1
Pipestone										
Polk										
Pope										
Ramsev					1	1	1	1		1
Red Lake				-						
Redwood										
Renville				-	-		-			
Rice										
Rock										
Roseau										
Saint Louis				1	1		1	1	1	1
Scott				1	1		-	1	1	1
Sherburne		1		1	1	1		1	1	1
Sibley		-		1	1	-	1			1
Stearns				1	1		1	1		1
Steele				1	1		1	1		1
Stevens							1			1
Swift										
Todd										
Traverse										
Wabasha			1	-	1		1			1
Wadena			1		1		1		1	1
Wasaca									1	1
Washington							1	1	1	1
Watonwon							1	1	1	1
Wilkin										
Winona					1			1		1
Wright					1			1		1
Vallow										
1 enow										
wiedicine										

Combining pre-2015 acoustic survey data with other known locations shows that northern long-eared bats are distributed throughout the forested region of Minnesota (Fig. 4). Acoustic surveys and occurrence records have been focused on the northern half of Minnesota, around the Twin Cities metropolitan area, and in the southeastern corner of the state. However, based on the documented distribution of northern long-eared bats from recent research projects, it is likely that the species is present throughout the forested portion of the state, consistent with Johnson (1918), Surber (1932), Gunderson and Beer (1953), Hazard (1982), and Nordquist and Birney (1985).

Figure 4. Map of pre-2015 bat acoustic survey locations in Minnesota showing detection of northern long-eared bat (MYSE) calls. Sites marked with yellow are from past records, and sites marked with blue are from the current project (Swingen et al. 2018a). Counties with records are shaded green.



Discussion

Historically, northern long-eared bats were thought to be distributed across Minnesota, although this was originally based on relatively few documented specimen locations. Hibernacula sites were unknown in the early 1900s, and northern long-eared bats are less likely to be found because of the tendency of northern long-eared bats to roost in trees instead of buildings. Most references to abundance after the 1940s indicate that the little brown bat is the most common bat in Minnesota and that the northern long-eared bat is less common. Many of the references also indicate that the northern long-eared bat is more common than occurrence records indicate.

The several books that have been published on the Mammals of Minnesota generally indicate a summer range throughout the state, although there were few documented locations to support this (e.g., 8 specimens in Hazard (1982), 9 locations in Gunderson and Beer (1953)). Even as late as 1985, there were only 8 counties in Minnesota with documented locations of the northern long-eared bat in summer (Fig. 2, from Nordquist and Birney 1985). Thus, although specimens had been identified in counties distributed from north to south in Minnesota, the validity of the extent of summer range could have been challenged.

Acoustic detectors made it possible to more efficiently find northern long-eared bats in the summer, and the analysis of acoustic data has provided strong support for northern long-eared bats being distributed throughout at least the forested part of Minnesota. There were few deployments of acoustic detectors in the southern half of Minnesota prior to 2015 (Fig. 4), but deployments and mistnet captures from 2015 to 2018 provided additional support for presence of northern long-eared bats in the southern half of Minnesota (Swingen et al. 2018a, b).

One important aspect of acoustic data is that it provides evidence of distribution, but it is still not possible to use acoustic data to determine abundance of different species. As discussed in the Introduction (p. 6), while it is easy to differentiate high-frequency and low-frequency species from the bat calls, it can be difficult to differentiate species within each frequency group. Because of similarities of calls among species, relative abundance calculations must also be qualified with the identification criteria used. Human interpreters and software programs do not always agree when assigning species identifications to a call file (Lemen et al. 2015).

Although recording equipment and analysis software have made bat surveys more practical, acoustic file identifications are still less reliable than confirming species presence through mist-netting. The northern long-eared bat in particular is difficult to confirm positively from acoustic records because its calls are so similar to calls made by the closely related little brown bat. The automated programs appear to be more likely to identify a call to be from a little brown bat, because not every call made by a northern long-eared bat has the distinctive high-frequency part of the call present.

Another important aspect of both acoustic surveys and mist-netting is that it is difficult to impossible to prove absence. The only area of the state where northern long-eared bats have not been found during any survey is in the southwestern counties, where forested areas cover a small portion of the landscape. In all other areas that have been surveyed, at least some surveys have indicated presence of northern long-eared bats. It is probably a safe assumption that in the forested portion of Minnesota, even if one acoustic survey fails to detect northern long-eared bats, another acoustic survey in the area would detect their presence.

Overall, based on documented locations and acoustic surveys, the northern long-eared bat is present throughout the forested region of Minnesota. The mist-netting and acoustic detection parts of this project, conducted from 2015 to 2017, provided additional data on the distribution of the northern long-eared bat in Minnesota (Swingen et al. 2018a, b).

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