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SOLUTIONS FOR IMPERILED BAT CONSERVATION: INTEGRATING ECOLOGY, TECHNOLOGY, AND THE PUBLIC

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A Thesis Submitted to the Graduate School in Partial Fulfillment of the Requirements for the Degree of Master of Science

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SOLUTIONS FOR IMPERILED BAT CONSERVATION: INTEGRATING ECOLOGY, TECHNOLOGY, AND THE PUBLIC

An Abstract of the Thesis by
Amy Marie Hammesfahr

Bat populations have plummeted in Missouri since the introduction of white-nose syndrome (WNS) in 2012, presenting challenges in researching understudied species' habitat ecology. Frequently incorporated survey techniques, such as mist netting and radio-telemetry, have become unreliable post-WNS. In response to address the challenge of studying rare species, we explored the alternative strategies of acoustic monitoring, acoustic lures, and human dimension surveys that may enhance surveys. Our goals from these objectives included comparing the methods to recommend better management decisions for imperiled bat species post-WNS. For Chapter I, we surveyed three imperiled bat species in southeastern Missouri, including the northern long-eared bat *Myotis septentrionalis*, little brown bat *Myotis lucifugus*, and tricolored bat *Perimyotis subflavus* using mist-netting and acoustic monitoring. We assessed the efficacy of modern acoustic monitoring activities to more traditional approaches of mist-netting and radio-telemetry. We never captured northern long-eared bats or little brown bats during our mist net surveys, but we did detect them acoustically. Chapter II evaluated the acoustic lures' success in increasing detection success of mist net and acoustic detector surveys. We captured two tricolored bats when we used an acoustic lure and detected them acoustically during the two years of the study. Our capture success allowed us to identify the first tricolored bat maternity roost within a Missouri Department of

Conservation (MDC) area in Carter County through radio-telemetry. We found our acoustic lure positively affected the acoustic activity of the endangered Indiana bat *Myotis sodalis* and big brown bat *Eptesicus fuscus*. We also assessed a human dimensions analysis to assess the level of public familiarity of bat species in Missouri, WNS awareness, perceived attitudes, and trust in the MDC. We found that respondents were less knowledgeable about WNS and bat natural history, despite their overall positive or neutral perception of bats. The public in our study trusted the MDC as a natural resource management agency. Both public trust and accurate knowledge of bat natural history and threats must be accounted for when suggesting forest management modifications to benefit our three imperiled target species.

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CHAPTER I

HABITAT ECOLOGY AND ACOUSTIC ACTIVITY OF THREE IMPERILED BAT SPECIES IN SOUTHEASTERN MISSOURI

ABSTRACT

White-nose syndrome (WNS) has caused severe declines in northern long-eared bat *Myotis septentrionalis*, little brown bat *Myotis lucifugus*, and tricolored bat *Perimyotis subflavus* populations in southeastern Missouri. Information about the species' population health, spatial distribution, and habitat ecology are all understudied in the area. Traditionally used mist net techniques may be unreliable survey methods post-WNS since individuals are rare on the landscape. In this study, we compared the efficacy of mist netting and acoustic monitoring as post-WNS survey tools. We assessed species distributions across Shannon, Carter, and Reynolds counties in southeastern Missouri. Captured female and juvenile target species were radio-tagged and tracked to maternity roosts. We described the first maternity roost habitat for tricolored bats within Carter County's Missouri Department of Conservation (MDC) areas. Although we detected them acoustically across this study's three years, we did not capture little brown bats or northern long-eared bats. We did not find any relationship between the three species'

habitat use and acoustic activity. The rare occurrence of the three species in the area limited our results; however, our findings contribute to habitat research for tricolored bats. Before timber sales or management activity occur, biologists should monitor species presence by deploying acoustic detectors and mist nets. Both techniques convey essential details about species population health, although we argue that acoustic monitoring is more efficient for documenting imperiled species post-WNS.

INTRODUCTION

Bats in North America face many threats, including habitat loss from timber harvesting and agricultural conversion, disease, and wind energy development (Frick et al. 2019). One of these threats, white-nose syndrome (WNS), has proven to be challenging to control since the disease is complex and highly detrimental to many species (Pettit and O'Keefe 2017). Hibernacula studies since the introduction of WNS in Missouri in 2012 suggest three species have significantly declined: Northern long-eared bat *Myotis septentrionalis* (99.9% decline), little brown bat *Myotis lucifugus* (86.7% decline), and the tricolored bat *Perimyotis subflavus* (53.8% decline) (Colatskie 2017). Range-wide declines observed throughout North America led scientists to petition tricolored bats and little brown bats for inclusion as a federally endangered species through a five-year endangered species listing working plan for the U.S. Fish and Wildlife Service (USFWS) (USFWS 2017, 2019a). White-nose syndrome associated declines for the northern long-eared bat population declines resulted in the species listing as federally threatened in 2015 (USFWS 2018).

Roosting habitat availability is influenced by forest management activities, emphasizing the need for more ecological roost data (Silvis et al. 2016). Each of our three focal species shares similar preferences for maternity roosts, although there are some key differences among species. Northern long-eared bats often inhabit cluttered interior forests with dense canopy cover and large diameter trees (Broders et al. 2004; Starbuck et al. 2015). Substrates used as roosts include tree cavities and crevices under sloughing bark (Barclay and Kurta 2007). Little brown bats use similar roost substrates as northern long-eared bats, although they frequently use anthropogenic roosts (Crampton and Barclay, Robert M. 1998; Olson and Barclay 2013; Schwartz et al. 2016; Thomas and Jung 2019). The maternity roosts used by tricolored bats include solitarily or small colonies of bats roosting in leaf clusters in live and dead oak species *Quercus spp.*, and occasionally tree cavities (Veilleux et al. 2003; Perry and Thill 2007). Vegetation characteristics identified in areas occupied with tricolored bats include those with taller and larger trees, high basal areas, and within riparian areas, although habitat studies for this species are limited (Ford et al. 2005; O'Keefe et al. 2009).

This study evaluated post-WNS maternity ecology and spatial distribution through acoustic activity and capture of northern long-eared bats, little brown bats, and tricolored bats in southeastern Missouri. Populations of the three species are rare, and therefore is essential to compare the efficacy of mist net and acoustic surveys to determine which method is more reliable for documenting species presence post-WNS. We collected vegetation data at acoustic and roost sites to identify habitat associations with the species and to compare our results with other studies. We predict that the three species will be rare on the landscape based on recent hibernacula data; thus, acoustic surveys will offer better insight into species distribution within the three counties than mist netting.

METHODS

Study Areas

We included ten survey areas within MDC areas in the counties of Shannon, Carter, and Reynolds in southeastern Missouri. Missouri Department of Conservation areas we surveyed included Angeline, Peck Ranch, Current River, Riverside, Birch Creek, Rocky Creek, Clearwater, Sunklands, Powder Mill Cave, and Logan Creek (Fig. 1.1). Dense mixed forests, extensive river systems, and hilly terrain characterize the study area's natural landscape (Steyermark and Yatskievych 2006). Summers in Missouri are warm, with an average monthly maximum temperature of 29.3°C between May and August (SD ± 3.14) (NOAA 2019). Most of the rainfall in the area falls during May and June, with an average of 10.3 cm (SD ± 1.19) between the two months (NOAA 2019).

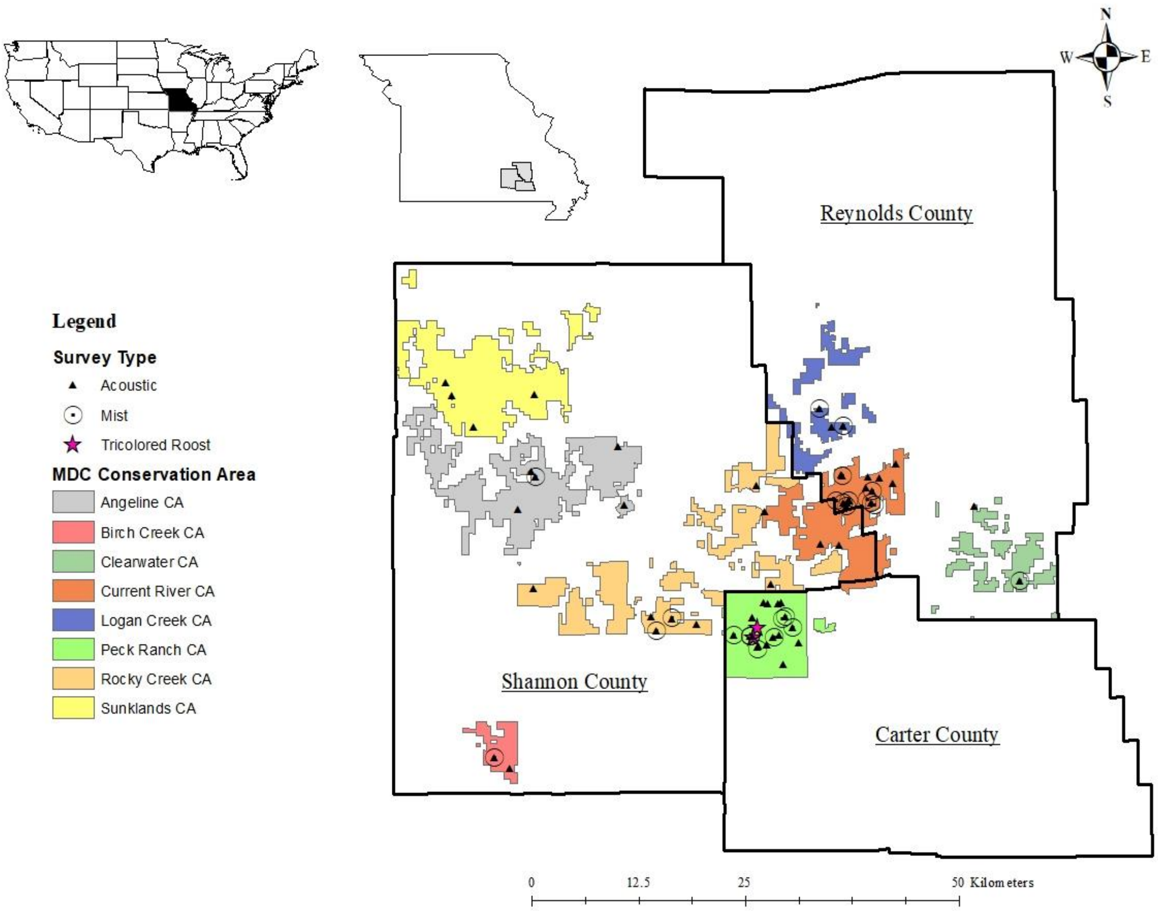


Figure 1.1 Mist net and acoustic survey sites sampled between 2018 and 2020. We did not include sites that did not record or capture bats

Mist Net Surveys

We mist netted over water (e.g., small wildlife ponds and creeks) up to three nights at eight MDC areas between May and August 2018 – 2019 (Fig. 1.1). We included 23 mist net nights in 2018 and 33 mist net nights in 2019. We separated each survey at a site by at least a week to increase capture success (Kunz and Parsons 2009). We suspended our capture efforts in 2020 due to insufficient personal protective equipment that would be necessary to mitigate SARS-CoV-2 reverse zoonosis to bats (MDC 2020b). We followed procedures and recommendations for wildlife handling under the most current version of the *USFWS Range-wide Indiana Bat Survey Protocol* and through the U.S. Department of Agriculture and MDC's Institutional Animal Care and Use Committee. Additionally, we adhered to the most recent WNS decontamination protocol procedures to prevent the spread of WNS to other bats (WNS Decontamination Team 2018). We used Kunz and Parsons (2009) methods for net placement at each site to achieve the greatest coverage during surveys. Other compliance to complete research on state-owned lands was covered under an approved MDC grant that served as the graduate research assistant's primary research funding. Mist netting was permitted under Amy Hammesfahr's Federal Permit TE61451C-1 and Missouri Wildlife Collector Permits (17893, 18119, and 18685 for 2018 – 2020, respectively).

Acoustic Surveys

We passively sampled for bat activity at each site with a full spectrum SM2BAT+ bat detector (Wildlife Acoustics, Maynard, MA) for two to four nights at each site over a three-year (2018 – 2020) period between May and August. We sampled for 53 nights in

2018, 52 nights in 2019, and 60 nights in 2020. We upgraded our microphones in 2019 to address recording quality issues we experienced during 2018. We verified the microphones' sensitivity and functionality with the manufacture's ultrasonic calibrator before deploying the detectors.

Extendable painter poles secured the detector's ultrasonic microphones three meters above the ground. We used recording settings that were sensitive enough to detect quietly echolocating bats and minimized the effects of noise from insects and vegetative clutter. We programmed detectors to begin recording at sunset and end at sunrise. Detectors were placed at least three meters from the water's edge and vegetation; however, some sites were limited by the availability of natural openings, which are ideal for recording bat calls (Reichert et al. 2018). Data collected at dry streambed sites yielded no acoustic bat activity in 2018, and we did not reattempt sampling such habitat in future surveys. We resampled three sites at Peck Ranch in 2020 that were sampled in 2018 to maximize the number of streams surveyed. This resampling allowed us to record better quality data that improved species identification compared to the data collected during 2018.

Acoustic Analysis

We used an automatic classifier, Kaleidoscope Pro v.5.1.9 (Wildlife Acoustics, Maynard, MA), for full-spectrum acoustic data analysis in addition to manually vetting each bat pass. We used *Sonobat Echolocation Call Characteristics of Eastern U.S. Bats* v.4x key and a reference library to verify each auto-classified bat pass (Szewczak et al. 2017). We described a 'bat pass' as three or more echolocation calls (called a pulse) that originated from a single species; one recorded bat pass per species equaled one recorded file (Britzke et al. 2013; Reichert et al. 2018). Bat passes included in our assessment were within the search phase and included more than three echolocation pulses per file to maintain consistency during analysis (Loeb et al. 2015; Reichert et al. 2018). Each bat pass was viewed alternatively in the 'real-time' and 'compressed' modes to verify that they were within the search phase and originated from bats (Reichert et al. 2018).

We followed published acoustic bat species vetting protocols to complete our analysis, focusing on each bat passes' maximum and minimum frequencies, characteristic frequency, duration, pulses per second, and bandwidth (Szewczak et al. 2017; Reichert et al. 2018). We documented occurrences where multiple species were present within a bat pass file, but not those that originated from multiple individuals of the same species to avoid bias and oversampling. Bat passes that were visually derived from a bat but contained less than three pulses in the bat pass and those that were social vocalizations were labeled 'noise' and omitted from the analysis. Only one of the authors manually vetted each collected file to avoid bias. For bat pass sequences unidentifiable at the species level, we grouped similar echolocation calls (Reichert et al. 2018).

Radio-telemetry, Tracking, and Roost Characteristics

We radio-tagged captured female or juvenile target species by gluing a 0.27 g transmitter on the bat's back to track it to its maternity roost (Holohil Systems Ltd., Ontario, Canada). We ensured that the transmitter and glue's weight was not more than five percent of the bat's weight. We listened to the bat's frequency during the day with a three-element Yagi antenna to locate their roost. Once we located the maternity roost, we collected vegetation data (Table 1.1) around an 11.3 m radius around the maternity tree (James and Shugart 1970).

Table 1.1 Forest variables measured at maternity roosts and random non-roost trees. Roost tree DBH refers to the diameter breast height of a selected roost.

Forest Variables for Maternity Roosts	
Roost Tree DBH	Roost Tree Species
Basal Area	Avg. Canopy Density
Roost Tree Height	Roost Tree Decay Score
Roost Substrate	Avg. Canopy Height
Tree Type	Avg. Mid-story Height

We measured canopy cover with a spherical convex densiometer (Forestry Suppliers, Inc, Jackson, MS), the basal area with a 10-factor prism, tree class size with a Biltmore stick and diameter height breast (DBH) tape, and tree height with a digital rangefinder (Forestry Pro Laser Rangefinder, Nikon, Melville, NY). ‘Tree type’ was described as deciduous or coniferous. We collected additional forest characteristics at the maternity roost, including decay score (USFWS 2019b) and roost substrate type (cavity, crevice, exfoliating bark, coniferous leaf cluster, squirrel nest, deciduous leaf cluster, or roost suspended in hanging branch). We grouped each tree’s measured values into size classes based on their DBH (Table 1.2).

Table 1.2 Description and conditions between different size classes of trees.

Term	**Conditions/DBH**
Saplings/small trees	DBH 5-14.5 cm
Poles	DBH 14.6-27 cm
Sawtimber	DBH $\geq$27.1 cm

For each identified roost tree, we paired the roost tree with a random non-roost tree to compare forest characteristics. To select our random non-roost tree, we traveled 500 –1000 m from the occupied roost tree at a randomized azimuth number between 0 – 360 degrees. Once we traveled 500 –1000 m from the roost tree, we selected the nearest tree with a DBH ≥7 cm, the smallest diameter tree used for roosting by a northern long-eared bat (USFWS 2019b). At our random non-roost tree sites, we collected vegetation measurements identical to those collected for our occupied roost tree forest measurements, except for roost height. We recorded any presence of a potential roost substrate for our random non-roost tree vegetation surveys.

We counted the number of bats that emerged from a maternity roost from twenty minutes before sunset until ten minutes passed from the last bat emerged or when the contrast of the sky and forest dissipated (USFWS 2019b). For each maternity tree used by the female, we monitored emergence twice.

Acoustic Site Vegetation Sampling

We collected vegetation characteristics at acoustic survey sites during all survey years. Our methods varied between 2018 and 2019 – 2020. For this study's analysis, we only include the vegetation methods we used during 2019 and 2020. We collected similar vegetation data for 2019 and 2020 as we collected for the maternity roosts, except we had three plots per site. The first plot included the acoustic detector area, with the detector established as the plot center in place of the roost's center. The second and third plots were 30 m away from the detector in two standardized directions, southeast and southwest. We averaged each plot's data then averaged the three plots to obtain representative characteristics for a site. We did not include roost substrate type as a measured forest characteristic in our acoustic site vegetation surveys.

Data Analysis

Our maternity roost sample size and species capture results limited statistical analysis. We averaged the number of bat passes collected per site by the number of recorded nights to account for this difference in sampling between years. We used linear models coupled with Akaike's information criteria (AIC_c) adjusted for small sample sizes to investigate relationships between bat activity and forest characteristics. We developed four to five a priori hypotheses for each species to assess which forest characteristics were most important for our species. Sample sizes for little brown bat detections were too small for analysis, so we performed statistical analyses for northern long-eared bats and tricolored bats.

Our models for northern long-eared bats included characteristics associated with densely cluttered environments, such as canopy coverage, the number of saplings per plot, and average sapling height (Broders and Forbes 2004; Starbuck et al. 2015). We compared habitats between ponds and streams to evaluate if the species were more likely to be recorded in one of the habitats. We explored if northern long-eared bats used areas with a higher number of sawtimber trees per plot since previous studies found support for that hypothesis (Badin 2014). We included forest characteristics associated with mature forests for our tricolored bat models, such as the number of sawtimber per plot and sawtimber height (Perry and Thill 2007; Carpenter 2017). Additional variables we assessed were canopy coverage and riparian/pond environments (Perry and Thill 2007; O'Keefe et al. 2009).

Our small sample sizes for each species permitted only one habitat predictor variable to be evaluated per linear model, but bat passes were always the response variable. Null models (e.g., intercept only) were included in our assessment, but we did not include a global model since we were limited by sample size. We assessed each model output by evaluating models with the lowest AIC_c and model weights ω_i to compare the strength of each model (Burnham and Anderson 2002). Models with $\Delta AIC_i < 2$ were considered to have equivalent support (Burnham and Anderson 2002). We used the coefficient of determination (r^2) to assess the explanatory power of best models. We used R Studio version 1.1.463 for all statistical analyses (R Core Team 2020).

RESULTS

Capture Results

We captured twice as many bats during 2019 compared to 2018, with 56.3% of the total captures representing red bats *Lasiurus borealis* (Table 1.3). We did not capture northern long-eared bats or little brown bats. We captured two tricolored bats, one male at Birch Creek Conservation Area and one female at Peck Ranch Conservation Area.

Table 1.3 Capture results for mist net surveys completed during 2018 and 2019. We order our adult and juvenile captures by most to least frequently captured. We captured one target species (e.g., tricolored bat, bolded). We did not capture northern long-eared or little brown bats.

		2018 Totals			**2019 Totals**			
Age	**Species Captured**	**Male**	**Female**	**Unknown**	**Male**	**Female**	**Unknown**	**Total Captures**
Adult	Red bat	26	23	0	69	33	0	151
	Red bats (Escaped)	13	0	0	2	0	32	47
	Evening bat	13	0	0	21	0	1	35
	Silver-haired bat	0	0	0	11	1	0	12
	Gray bat	5	1	3	7	0	1	17
	Seminole bat	1	0	0	1	0	0	2
	Hoary bat	7	0	0	6	0	0	13
	Big brown bat	0	0	0	1	4	0	5
	Tricolored bat	0	0	0	1	1	0	2
	Indiana bat	1	0	0	1	0	0	2
Juvenile	Red bat	15	8	0	29	12	0	64
	Evening bat	0	0	0	1	0	0	1
	Big brown bat	1	0	0	0	0	0	1
	Totals	**82**	**32**	**3**	**150**	**51**	**34**	**352**

Acoustic Analysis

The most frequently recorded species across the three years was red bats, followed by gray bats and evening bats (Table 1.4). During 2018, we recorded tricolored bats at one site at Current River Conservation Area (Table 1.5). We recorded tricolored bat passes during each of the three years, northern long-eared bats during 2019 and 2020, and little brown bats in 2020. Northern long-eared bats were only recorded in Shannon and Reynolds Counties, and the other species were recorded in all three counties. None of our habitat models for tricolored or northern long-eared bat acoustic activity were an improvement over the null hypothesis.

Table 1.4 The number of raw count bat passes recorded by species for each year 2018 – 2020.

Species	**2018 Bat Passes**	**2019 Bat Passes**	**2020 Bat Passes**	**Total Bat Passes**
Big brown bat	35	36	166	237
Big brown/silver-haired bats	274	203	875	1352
Red bat	372	1026	3873	5271
Hoary bat	73	55	172	300
Silver-haired bat	8	79	338	425
Low frequency bats	43	28	61	132
Gray bat	13	45	1952	2010
Evening bat	3	341	739	1083
Evening bat/red bat	0	359	1309	1668
Tricolored bat	44	122	96	262
Little brown bat	0	0	26	26
Indiana/little brown bat	0	0	44	44
Northern long-eared bat	0	3	41	44
Indiana bat	0	6	35	41
40 kHz Myotis	78	265	2027	2370
40 kHz Bat	1187	487	809	2483
Totals	**2130**	**3055**	**12563**	**17748**

Table 1.5 Number of bat passes/number of nights surveyed for the three imperiled species. Values within the parentheses represent the percentage of target species bat passes compared to the sum of all bat passes at the site.

Year	Site	Conservation Area	County	Little brown bat	Northern long-eared bat	Tricolored bat
2018	CRCA18A11	Current River	Reynolds			13/3 (8.9)
2019	CRCA19A06	Current River	Reynolds		2/4 (0.19)	234/4 (43.98)
	CRCA19A15	Current River	Reynolds			2/4 (0.92)
	LCCA19A02	Logan Creek	Reynolds	1/4 (0.26)		30/4 (7.81)
	PRCA19A01	Peck Ranch	Carter		3/4 (1.07)	
	PRCA19A09	Peck Ranch	Carter			2/4 (16.67)
	RCCA19A04	Rocky Creek	Shannon			14/4 (1.85)
2020	BCCA20A02	Birch Creek	Shannon	11/2 (0.86)	1/2 (0.08)	11/2 (0.86)
	CRCA20A18	Current River	Reynolds	6/2 (1.04)	3/2 (0.52)	2/2 (0.35)
	CRCA20A19	Current River	Reynolds		4/2 (1.18)	
	LCCA20A03	Logan Creek	Reynolds		5/2 (0.55)	72/2 (7.94)
	POCA20A01	Powder Mill Cave	Shannon		2/2 (0.38)	
	PRCA20A12	Peck Ranch	Carter		11/2 (3.22)	
	PRCA20A16	Peck Ranch	Carter		1/2 (33.33)	
	PRCA20A17	Peck Ranch	Carter			2/2 (5.26)
	PRCA20A18	Peck Ranch	Carter			8/2 (1.81)
	PRCA20A20	Peck Ranch	Carter		2/2 (0.96)	
	PRCA20A03	Peck Ranch	Carter			1/2 (0.09)
	RCCA20A01	Rocky Creek	Shannon		1/2 (0.36)	
	RCCA20A07	Rocky Creek	Shannon		2/2 (0.26)	
	SUCA20A01	Sunklands	Shannon		9/2 (2.05)	
	SUCA20A05	Sunklands	Shannon	9/2 (0.95)		

Roost Ecology

We failed to capture the northern long-eared bat or little brown bat during our mist net surveys. We captured one male tricolored bat at Birch Creek Conservation Area in Shannon County on July 23, 2019. One non-reproductive female tricolored bat was captured and radio-tagged at Peck Ranch Conservation Area in Carter County on May 17, 2019. The female used two roosts; the first roost, Roost One, was located in a dead white oak *Quercus alba* tree with a decay score of three (Fig. 1.3). We observed at least four individuals emerge with the tagged female from the same dead leaf cluster near the top of the tree; average emergence counts were six (SD±1.41) individuals observed between two observation periods. Some individuals emerged from leaf clusters near our tagged female.

Figure 1.2 Peck Ranch Roost One was located in a leaf cluster in a white oak snag. The roost location is in red.

The second roost, Roost Two, used by the female was an oak species *Quercus spp*. snag on May 22, 2019 (Fig. 1.4). We were unable to perform an emergence count at this location due to other research priorities. Roost Two had more decay than Roost One with suitable roosting substrates present, such as cavities and exfoliating bark that made identifying the exact roost location challenging. We tracked the individual to Roost Two after a heavy rain event (7.6 cm) that occurred over one evening. The day after the rain event, the female left Roost Two and returned to Roost One until we could not pick up the battery's transmission, which occurred on June 1, 2019. The female flew 2.4 km from the capture location to Roost One, and then 1.5 km to Roost Two. Roost Two was near the capture location.

Figure 1.3 Peck Ranch Roost Two used by the same individual that used Roost One. The roost location is outlined in red.

Both roosts that the female used were sawtimber trees with a DBH >30 cm (Table 1.6). Canopy cover at both roost sites was high, >95% (Table 1.6). The roost substrate differed between the two roosts; one substrate included a leaf cluster in a snag, and the other was located in exfoliating bark or a cavity. Both sites included deciduous trees with several sapling trees and no observed conifers. The associated heights of both roost types fell within the average pole timber height for each plot, and the average height of the trees around the roost tree was taller than the roost's height. Roost location resided within the mid-canopy of the forest. The basal area between the two sites was similar, 11-13 m^2/ha.

Table 1.6 Roost structure characteristics and vegetation characteristics observed in roost trees. Standard deviations follow averaged values.

Forest Characteristic	Roost 1	Roost 2
Tree species	*Quercus alba*	*Quercus spp.*
DBH (cm)	47.5	30.2
Roost tree decay score	3	6
Roost tree height (m)	32.7	10.6
Roost height (m)	21.9	6.6
Roost substrate	Deciduous leaf cluster	Exfoliating bark
Canopy coverage at roost	95.83% ± 3.70	97.65% ± 0.99
Canopy coverage 5 m from roost	95.57% ± 3.84	98.44% ± 0.60
Average deciduous saplings	23	9
Average deciduous poles	6	8
Average deciduous sawtimber	5	3
Average conifer saplings	0	0
Average conifer poles	0	0
Average conifer sawtimber	0	0
Average snag saplings	2	2
Average snag poles	1	0
Average snag sawtimber	0	0
Average sapling height (m)	13.53 ± 11.27	4.21 ± 2.97
Average pole height (m)	26.88 ± 11.47	14.68 ± 6.79
Average sawtimber height (m)	41.1 ± 15.92	22.2 ± 4.33
Snag basal area m^2/ha	0	0
Live Tree basal area m^2/ha	11.47	13.76

DISCUSSION

Capture Success and Roost Ecology

Assessing the population health of the species: the northern long-eared bat, the tricolored bat, and the little brown bat through mist net surveys has become a laborious task post-WNS due to their infrequent occurrence (Frick et al. 2010). Compared to our two targeted *Myotis* species, we had greater success capturing the tricolored bat. We successfully captured two tricolored bats out of the 352 bats captured across two years in southeastern Missouri. This result reflects diminished winter population status observed during hibernacula surveys (Ingersoll et al. 2016; Colatskie 2017; Pettit and O'Keefe 2017). We described the first documented maternity roosts of a tricolored bat in an MDC area within the research counties. This discovery provides insight into potential roost habitat use in southeastern Missouri and warrants additional research efforts.

The reduced capture success hinders the study of these rare species and stymies the development of supportive, science-based management decisions that could help protect impacted populations. This study discovered that mist netting and radio-tagging females and juveniles post-WNS was a slow and unreliable approach in southeastern Missouri. Similarly, a study in Tennessee successfully captured male tricolored bats, but very few females (n=23 males, n=3 females) (Carpenter 2017). While useful to understand male habitat use, the need for more ecological information on female habitat selection is critical for their survival.

Roost One's substrate material was similar to other studies (e.g., an oak leaf cluster; Veilleux et al. 2003; Perry and Thill 2007), although our Roost Two roost substrate did not fit the published descriptions because it was a snag without leaves. Only one study described the use of a cavity as a possible maternity roost (Menzel 1996). In the study, the authors assumed that one tricolored bat adult female and one juvenile fell from a cavity above a pitfall trap where they were captured (Menzel 1996). We hypothesized that heavy precipitation caused our radio-tagged female to search for a less exposed roost. What remains unclear is why the female used Roost Two when it was so far from Roost One. Available literature does not include any roost observations used during extreme rain events, but we argue that such events are uncommonly reported or observed in other studies. The roosting ecology for our three target species still lacks information within southeastern Missouri. The unfortunate reality is that individuals are so rare that gathering any statistical insights from collected data is challenging. The lack of recent, current roost ecology information on these species post-WNS provokes inquires whether the studies were not published due to small sample sizes or if researchers failed to capture these species or study their roosting ecology.

We recorded tricolored bat echolocation bat passes more frequently compared to the other two imperiled species, a result that supported our predictions. We did not anticipate recording as many northern long-eared bat echolocation calls as we collected. Bats likely use habitat based on several forest characteristics available (Jung et al. 2012). The small sample sizes we collected limited our habitat comparison analyses between the species.

Despite the challenges we encountered, we still captured bats; however, the effort required to capture rare species was intensive. Comprehending the full impacts of WNS on our target species requires yearly surveys of affected populations. Successful captures that are radio-tracked to roosts provide essential information needed to update forest management strategies to increase species survival. We do not believe that it is practical for wildlife biologists who are confined to the availability of human labor and budgets to realistically spend more than a few nights in an area searching for rare bat species. What might help maximize survey efforts include collaborating with other agencies, educational institutions, and non-profit partners, and incorporating alternative research methods when mist netting, such as acoustic analysis (Francl et al. 2011; Slough et al. 2014).

A significant contribution from acoustic analysis over mist net surveys was that we documented all three species through acoustic analysis. Our review of the time of night we recorded tricolored bat passes suggests that most of the species' activity occurred before sunrise. This observance suggests that we may have missed opportunities to capture the species by surveying around sunset instead of sunrise (Appendix I). Acoustic monitoring presents challenges in data collection, such as realizing that the number of bat passes recorded at a site does not equal individuals' abundance. Acoustic surveys suggest results as an index of the activity or the relative frequency of use at a site and estimate species richness (Hayes 2000; Britzke et al. 2013). Unlike mist net surveys, acoustic surveys do not provide information about the sex of recorded species or provide roosting ecology information. SARS-CoV-2 prevented us from mist netting in 2020, and agencies such as the MDC and USFWS suggested that acoustic monitoring occurs in place of mist net activity for all bat handling permittees (MDC 2020b). Acoustic monitoring will likely be an essential component in future bat mist net efforts, and our results reaffirm that the method is more useful in documenting species presence.

Management Recommendations

White-nose syndrome has caused significant population declines in our three species, and timber harvesting contributes additional stress on affected populations. Recommending forest management strategies for the three species is difficult, especially since the species required different habitat features and are rare. Our results suggest that little brown bats are rarer than northern long-eared bats. We recommend that the MDC survey areas with mist nets and acoustic detectors before any timber sales or habitat management. Northern long-eared bats are more sensitive to timber harvest than the other species we studied (Pauli et al. 2015). Forest management that strategizes optimizing clutter habitat for the northern long-eared bat and possibly little brown bats is recommended, and we suggest single tree selection occur as a harvesting technique (Guldin et al. 2007). Snags located within conservation areas should be left on site unless the snag threatens human life (Hayes and Loeb 2007). Given the diversity of forest habitat needs required by each species, a matrix of different forested and unforested habitats would optimize the habitat available in the conservation areas for each of the species (Jung et al. 1999; Yates and Muzika 2006).

We did not include landscape-level management activities such as logging intervals, prescribed fire activity, or management objectives (e.g., natural preserve, experimental forest) in our roost ecology or acoustic analysis. Area managers at both tricolored bat capture locations made comments about recent management activity. For example, at Peck Ranch Conservation Area, we were informed that Roost One was in a recently burned area. Based on our observations, we believed the roost tree recently died. The area manager at Birch Tree Conservation Area mentioned that the site was partially harvested within the last year. Tricolored bats may be more flexible and adaptive in their habitat selections, and more research should occur to observe different management objectives and their occupancy.

Our results reflect the challenges and research needs of southeastern bat populations post-WNS. WNS has significantly reduced populations, which restricts statistical analyses that determine habitat needs and trends. Forest management activities can negatively or positively affect already stressed populations, and data that contributes to more knowledge about rare species population status and habitat use is valuable locally and range wide. Biologists and researchers in the area should collaborate with state, federal, and non-profit partners to efficiently maximize effort. Local and regional bat conservation efforts can use this data to provide insight and recommendations for management objectives needed to improve bat habitat.

CHAPTER II

TUNING INTO BAT FREQUENCIES: THE POTENTIAL USE OF ACOUSTIC LURES FOR THE TRICOLORED BAT *PERIMYOTIS SUBFLAVUS*

ABSTRACT

The associated bat population declines from white-nose syndrome (WNS) in southeastern Missouri presents challenges in studying them due to their rarity on the landscape. Post white-nose syndrome surveys should explore additional tools that increase the chances of capturing WNS impacted species, such as acoustic lures. We tested the efficacy of an acoustic lure on one WNS susceptible species, the tricolored bat *Perimyotis subflavus*. We compared the capture success and acoustic activity of tricolored bats and other species within the bat community by randomizing the lure's presence while mist netting and monitoring acoustic activity. While we did not have a large enough sample size to assess tricolored bat acoustic activity, we captured two tricolored bats with the lure and none without the lure. Only big brown bats *Eptesicus fuscus* and Indiana bats *Myotis sodalis* increased their acoustic activity during the presence of the lure, while the lure did not increase or decrease other species detections. Future research efforts on the efficacy

of acoustic lures on tricolored bat behavior should expand into areas where the species is abundant to assess better whether acoustic lures are useful in studying the species.

INTRODUCTION

Acoustic lures are a new method used in bat studies that function similarly to broadcasted calls used to attract birds (Conway and Gibbs 2005), except the speakers play ultrasonic bat calls. Acoustic lures have benefitted population assessments by increasing capture rates of imperiled species such as the bonneted bat *Eumops floridanus*, northern long-eared bat *Myotis septentrionalis*, and Indiana bat *Myotis sodalis* (Quackenbush et al. 2016; Braun De Torrez et al. 2017; Samoray et al. 2019). Most of the research on acoustic lures focused on whether the acoustic lure increased captures, while fewer studies evaluated how the lure influenced the number of bat passes recorded. For instance, in North America, only one study incorporated acoustic detectors during the deployment of an acoustic lure and found no difference in the targeted species' level of bat activity between treatments (Loeb and Britzke 2010). Contrasting the North American study, Panamanian sac-winged bats *Saccopteryx bilineata* significantly increased acoustic activity near roosts when distress calls were played through an acoustic lure (Eckenweber and Knörnschild 2016). Acoustic activity at acoustic lure deployment sites in Scotland experienced an increase in acoustic detections from all species included in the study with some species responding strongly to calls from a particular genus (Lintott et al. 2013). Some research suggests that some vocalizations used on acoustic lures could repel some species (Russ et al. 2005b). As such, the comparison of acoustic activity with and without the presence of an acoustic lure may help identify behavioral changes and species attracted to the lure but are not captured (Flaquer et al. 2007).

Tricolored bats *Perimyotis subflavus* are rare in Missouri, and our understanding of their conservation status and ecology may benefit from the use of acoustic lures. For instance, capturing these bats using traditional methods has proven difficult, given their scarcity across their range (Francl et al. 2012). Tricolored bats served as our primary test subject for our study as their populations are decreasing at a slower rate compared to other WNS susceptible species in the area (Colatskie 2017). We expected the species to be less social than species of *Myotis*, and we hypothesized that the use of a distress call might attract the species better than the use of a social call. A review of a study on Pipistrelles *Pipistrellus* species demonstrated that species within this genera responded strongly to distress calls (Russ et al. 2005b). This success on similar relatives was our deciding factor of why we selected to test this echolocation.

Distress calls are often audible because they are vocalizations bats make when handled. This type of call's lower frequency presented questions in how the vocalization would transmit through the environment since lower frequency calls may not transmit effectively in dense forests compared to more open forests (Penna and Solís 1998; Patriquin et al. 2003). Other acoustic lure studies have not evaluated if differences in forest habitats affect the efficacy of the lure. These assessments help identify optimal locations of which to deplore acoustic lures if an effect is observed.

We designed an acoustic lure study to determine if we could increase the capture rates and acoustic activity of tricolored bats in Missouri, to determine if acoustic lures could improve imperiled bat surveys post-WNS. Results from our research update the U.S. Fish and Wildlife Service (USFWS) and the state of Missouri on post-WNS population status and habitat use. Additional objectives included ensuring that the acoustic lure did not repel any Missouri or federally listed species from the study area (Missouri Heritage Program 2019). We did not want to negatively impact other species' capture success, so we assessed the efficacy of the lure on other species found within the bat community. Finally, we evaluated whether vegetation composition influenced lure effectiveness since broadcasted echolocation calls may be constrained in densely forested habitats (Brigham et al. 1997; Patriquin et al. 2003).

METHODS

Study Areas

We included nine survey areas within the Missouri Department of Conservation (MDC) areas in Shannon, Carter, and Reynolds counties in southeastern Missouri (Fig. 2.1). Conservation Areas surveyed included Angeline, Peck Ranch, Current River, Birch Creek, Rocky Creek, Clearwater, Sunklands, Powder Mill Cave, and Logan Creek (Fig. 2.1). Dense mixed forests, large river systems, and hilly terrain characterized the study area's natural landscape (Steyermark and Yatskievych 2006). Summers in Missouri are warm, with an average monthly maximum temperature of 29.3°C (SD ± 3.14) between May and August (NOAA 2019). Most of the area's rainfall occurs during May and June, with an average of 10.3 cm (SD ± 1.19) of precipitation between the two months (NOAA 2019).

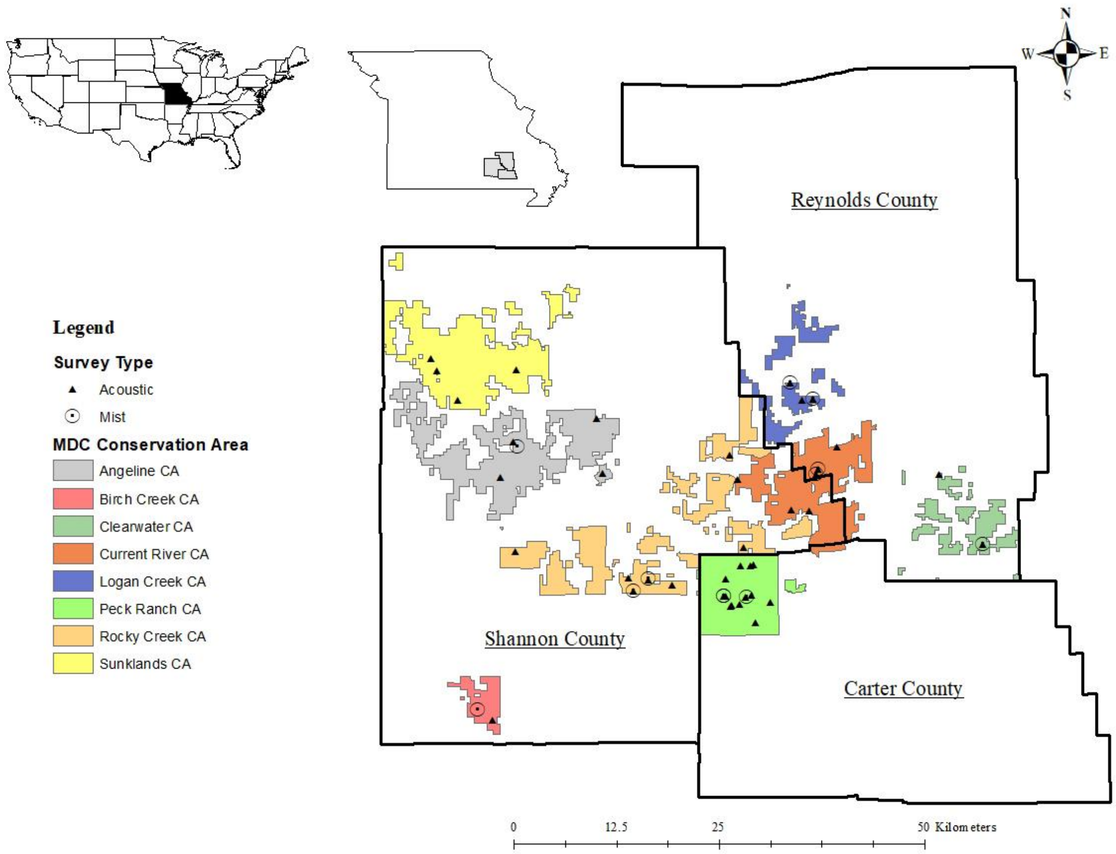


Figure 2.1 Mist net, acoustic survey, and roost locations used in our study.

Acoustic Lure

We broadcasted tricolored bat distress calls with our lure, since studies on similar Pipistrelles *Pipistrellus* species responded positively to this type of echolocation call (Russ et al. 2005a). We recorded distress calls made by handled tricolored bats (n=11 males, 5 females) with a SM4BAT detector and SMM_U2 microphone (Wildlife Acoustics, Maynard, MA) during fall and spring capture efforts during 2018 –2019 in Oklahoma, Missouri, and Kansas. We compiled a SD card with five unique ten-minute tracks made from our 44 randomized distress calls. We only included calls with low noise to sound ratios. We used *Audacity* (v. 2.3.2, 2018) software to create a fluid, continuous ten-minute track. We are unaware of any North American studies that have confirmed if bats recognize individuals based on their echolocation. Based on this lack of research, we assumed echolocation calls were not unique among individuals and therefore included multiple calls collected from the same individual. Other lure studies included fewer echolocation files than we included (Braun De Torrez et al. 2017; Samoray et al. 2019). We used the manufacture's accessory timer kit to control the lure's start and end times and programmed the lure to run ten minutes on and ten minutes off (Eckenweber and Knörnschild 2016; Quackenbush et al. 2016; Braun De Torrez et al. 2017). The lure's volume and other audio settings were set according to the manufacturer's recommendations to avoid clipping.

Mist Net Surveys

We mist netted over water (e.g., small wildlife ponds and creeks) two nights each at eight MDC areas between May and August 2019. We suspended our capture efforts in 2020 due to insufficient personal protective equipment that would be necessary to mitigate SARS-CoV-2 reverse zoonosis to bats (MDC 2020b). We followed procedures and recommendations for wildlife handling under the most current version of the *USFWS Range-wide Indiana Bat Survey Protocol* and through the U.S. Department of Agriculture and MDC's Institutional Animal Care and Use Committee. We adhered to the most recent WNS decontamination protocol procedures to prevent the spread of WNS to other bats (WNS Decontamination Team 2018). Mist net surveys began at sunset and lasted for five hours. We used Kunz and Parsons (2009) methods for net placement at each site to achieve the greatest coverage during surveys. We separated each survey at a site by at least a week to increase capture success (Kunz and Parsons 2009). Other compliance to complete research on state-owned lands was covered under an approved MDC grant that served as the graduate research assistant's primary research. Mist netting was permitted under Amy Hammesfahr's Federal Permit TE61451C-1 and Missouri Wildlife Collector Permits (17893, 18119, and 18685 for 2018-2020, respectively).

Each site was mist net twice, and we randomly flipped a coin to determine which night received the lure treatment. During mist net surveys, the lure faced the center of a triple high mist net with <30.5 cm of space in-between the lure and the mist net (Russ et al. 2005a; Loeb and Britzke 2010; Braun De Torrez et al. 2017). Our replicates included only ponds and streams; therefore, the lure floated on the water's surface on a modified boogie board (Fig. 2.2). The boogie board's frame secured the lure onto it with a camera mount, making the lure 127 cm from the water's surface. We angled the lure 120 degrees towards the net with the camera mount and balanced the boogie board with two grounded 4.5 – 5.5 kg weights to prevent it from spinning. At shallow creeks, we used a fence t-post to elevate the lure (see stake deployment methods in the proceeding section for a description) to maintain consistent deployment conditions.

Figure 2.2 Acoustic lure deployed on a modified boogie board.

Acoustic Activity Surveys

We passively sampled for bat activity at each mist net site with a full spectrum SM2BAT+ bat detector (Wildlife Acoustics, Maynard, MA) for two nights at each site during 2019 and 2020. Similar to our mist netting methods, we utilized a coin toss to determine which night out of the two we would deploy the lure. We placed the detector's ultrasonic microphones on extendable painter poles three meters above the ground. The detectors recorded bat data for five hours beginning at sunset to co-occur with the lure's timer. Detectors were placed at least three meters from the water's edge and vegetation; however, some sites were limited by the availability of natural openings, which are ideal for recording bat calls (Reichert et al. 2018). The acoustic lure was placed approximately one to three meters from the detector, depending on the site's vegetation characteristics, slope, ground surface substrate, and visibility to the public. We did not control the distance from the lure to the detector since the manufacturer's microphone specifications indicated that the microphones we used could pick up bat echolocations from at least 20 meters away (Agranat 2014). A camera mount secured the lure to a monopod, and then we zip tied the monopod to a fence t-post, with the height fixed at 133 cm.

We used an automatic acoustic classifier, Kaleidoscope Pro v.5.1.9 (Wildlife Acoustics, Maynard, MA), for full-spectrum acoustic data analysis in addition to manually vetting each bat pass. We used *Sonobat Echolocation Call Characteristics of Eastern U.S. Bats* v.4x key and a reference library to verify each auto-classified bat pass (Szewczak et al. 2017). We described a 'bat pass' as three or more echolocation calls (called pulses) that originated from a single species; one recorded bat pass per species equaled one recorded file (Britzke et al. 2013; Reichert et al. 2018). Bat passes included in our assessment were within the search phase and included more than three echolocation pulses per file to maintain consistency during analysis (Loeb et al. 2015; Reichert et al. 2018). Each bat pass was viewed alternatively in the 'real-time' and 'compressed' modes to verify that calls were within the search phase and originated from bats (Reichert et al. 2018), and not the acoustic lure. We conservatively removed the bat pass from analysis if we could not determine if the bat pulses observed belonged to the lure or a lower frequency bat. One author manually vetted each collected file to avoid bias.

We followed published bat species vetting protocols to complete our analysis, focusing on each bat passes' maximum and minimum frequencies, characteristic frequency, duration, pulses per second, and bandwidth (Szewczak et al. 2017; Reichert et al. 2018). We documented occurrences where multiple species were present within a bat pass file, but not those originated from multiple individuals of the same species to avoid bias and oversampling. Bat passes that were visually identifiable as originating from a bat but contained less than three pulses in the bat pass were labeled 'noise' and omitted from the analysis. We omitted unidentifiable bat passes (such as social calls) from our analyses. For bat pass sequences unidentifiable at the species level, we grouped similar echolocation calls (Reichert et al. 2018).

Vegetation Sampling

We collected vegetation data at three plots per site, with each plot measuring the vegetation within an 11.3 m radius (James and Shugart 1970). The first plot included the immediate acoustic detector area, with the detector established as the plot center. The second and third plots were 30 m away from the detector in two standardized directions, southeast and southwest. We measured our forest characteristics with a spherical convex densiometer (Forestry Suppliers, Inc, Jackson, MS), 10-factor prism, Biltmore stick and diameter height breast tape (DBH), and digital rangefinder (Forestry Pro Laser Rangefinder, Nikon, Melville, NY). Tree type was described as deciduous or coniferous. We grouped each tree's measured values into size classes based on their DBH (Table 2.1).

Table 2.1 Description and conditions between different size classes of trees.

Term	Conditions/DBH
Saplings/small trees	DBH 5-14.5 cm
Poles	DBH 14.6-27 cm
Sawtimber	DBH ≥27.1 cm

Data Analysis

We used general linear mixed models with a Poisson distribution and log link for acoustic data, although if data was overdispersed, we instead used negative binomial distributions with a log link (function *glmer* or *glmer.nb,* R package, *lme4*; Bates et al. 2020). Statistical significance ($\alpha = 0.05$) was assessed with likelihood ratio tests with the Laplace Approximation method for our acoustic results (R package, *lme4*). We included year as a random effect in our acoustic analyses because we collected acoustic data at more sites in 2020 than during 2019 and experienced different weather conditions across the two years. We used generalized linear models with a Poisson distribution and log link for our capture data, unless data was overdispersed we used negative binomial distributions with a log link (function *glm.nb*, R package, *MASS*, Dunn and Smyth 2018; Ripley et al. 2020). Statistical significance ($\alpha = 0.05$) was assessed with Wald χ^2 tests for our capture results (function *Anova*, R package, car; Dunn and Smyth 2018; Fox et al. 2020). We had few detections and low sample sizes for most species, and as such we created three species "groups" for which we then examined lure effectiveness. These groups included: 1) all bats except the two dominant species, red bats and evening bats, 2) local bat species of concern (e.g., silver haired bats, hoary bats, Seminole bats, tricolored bats, Indiana bats, gray bats, northern long-eared bats and little brown bats), and 3) all species combined. Furthermore, we compared the number of females and juveniles captured with the lure compared to without the lure.

To evaluate if vegetation characteristics impacted lure efficacy, we analyzed the total captures and total bat passes observed with the lure. Variables we tested that affect the transmission of lower frequency sounds included: number of saplings per site, average sapling height, average sawtimber height, and canopy cover (Brigham et al. 1997; Patriquin et al. 2003; Broders and Forbes 2004; O'Keefe et al. 2014). We included average sawtimber height since research suggested that the forest canopy acts as a 'noise ceiling,' which means that there may be a limit in sound transmission based on canopy density (Morton 1975).

We compared our generalized linear mixed models using Akaike's information criterion (AIC_c) adjusted for small sample sizes. We included null models (e.g., intercept only) in our comparisons, but we did not include a global model for capture results since we were limited on the number of predictors based on our sample size. We assessed each model output by evaluating models with the lowest AIC_c and model weights ω_i to compare the strength of each model (Burnham and Anderson 2002). Models with $\Delta AIC_i < \Delta AIC_c 2$ were considered to have equal support (Burnham and Anderson 2002). We checked predictor variables for multicollinearity before inclusion in the a priori models (function *chart.correlation*, R package, *PerformanceAnalytics;* Peterson et al. 2020) . We removed predictors that showed a high level of collinearity (r >0.60). We assessed model fit with pseudo-R^2, which was calculated using the lognormal observance variance option in the R package, *MuMin* (Barton 2020) with the function, *r.squaredGLMM*. We used R Studio version 1.1.463 for all statistical analyses and the package *ggplot2* to create our graphs (R Core Team 2020; Wickham et al. 2020).

RESULTS

Bat Captures

We mist netted for 33 nights between May and August 2019 and captured two red bats when the lure was used and none when the lure was not used (Table 2.2). We found no difference in the total captures of all species combined when the lure was used compared to when it was not (p=0.16). Similarly, the lure did not increase the captures of females, juveniles, red bats, evening bats, nor individuals within all tested species groups. We found no difference in our capture models when we compared forest characteristics found at sites between treatment types.

Table 2.2 Capture results observed between the two treatments in 2019. We captured 160 bats with the lure and 108 without the lure. "Escaped bats" referred to individual bats that escaped the net before processing.

		Lure Totals			No Lure Totals		
Age	**Species Captured**	**Male**	**Female**	**Unknown**	**Male**	**Female**	**Unknown**
Adult	Red bat	46	21	0	23	12	0
	Escaped red bat	2	0	19	0	0	13
	Evening bat	12	0	0	9	0	1
	Silver-haired bat	10	1	0	1	0	0
	Gray bat	4	0	0	3	0	0
	Escaped *Myotis* spp.	0	0	0	0	0	1
	Seminole bat	1	0	0	0	0	0
	Hoary bat	4	0	0	2	0	0
	Big brown bat	0	3	0	1	1	0
	Tricolored bat	1	1	0	0	0	0
	Indiana bat	0	0	0	1	0	0
Juvenile	Red bat	9	6	0	20	6	0
	Evening bat	1	0	0	0	0	0
	Totals	**109**	**32**	**19**	**75**	**19**	**14**

Acoustic Activity

Acoustic surveys occurred at ten sites for 20 survey nights in 2019, and for 60 survey nights in 2020 at 30 sites. We surveyed fewer sites during 2019 due to frequent rain events and limited equipment. We observed less acoustic activity for tricolored bats when deploying the lure, although we could not perform statistical tests on the species due to less than ten replicates observed (Table 2.3). Interestingly, our acoustic detector did not record tricolored bat passes at the time of their capture. We expected our acoustic detector microphone range to be at least 20 m (Agranat 2014). We found big brown bats (p=0.02, z=2.27, Fig. 2.3) and Indiana bats (p=0.04, z=2.04, Fig. 2.3) increased their acoustic activity with the lure compared to without the lure. We found no difference in total bat activity between the treatments (p=0.19) nor within the 40 kHz *Myotis*, local species of concern, and community of bats without red bats and evening bat groups. We found no difference in our acoustic models in our comparison of forest characteristics and the lure's effectiveness, and we were unable to reject the null hypothesis.

Table 2.3 Total bat passes recorded between treatment types during 2019-2020. Species with an asterisk represent statistically significant results.

Species	**Total Bat Passes**	
	Lure	**No Lure**
Red bat	1895	1465
Gray bat	767	558
40 kHz *Myotis* spp.	733	627
Evening/red bats	652	443
Big brown/silver-haired bats	541	230
40 kHz Bat	313	319
Evening bat	304	228
Silver-haired bat	123	110
*Big brown bat	105	37
Hoary bat	84	51
Low-frequency species	29	32
Indiana/little brown bats	17	14
*Indiana bat	16	6
Northern long-eared bat	14	9
Little brown bat	2	16
Tricolored bat	2	19
Totals	**5597**	**4164**

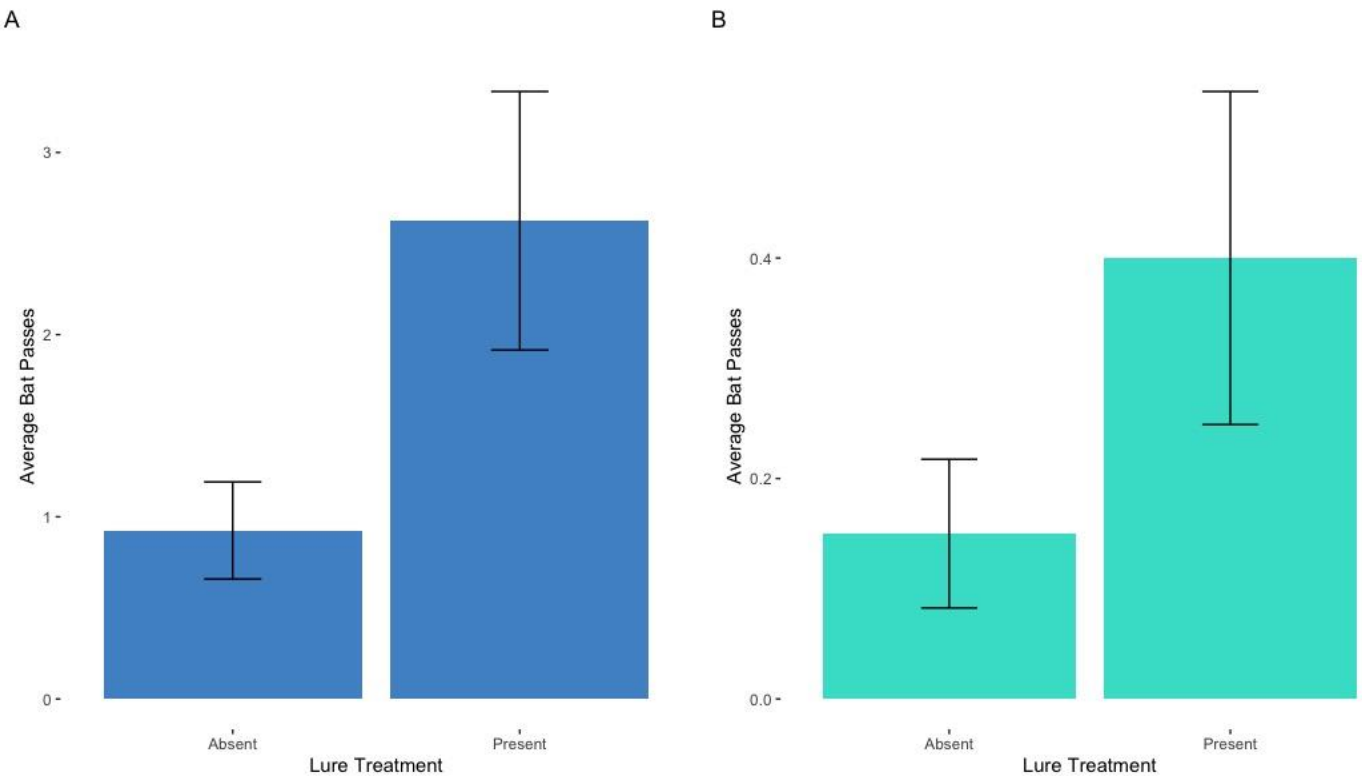


Figure 2.3 (A) Big brown bat and (B) Indiana bat acoustic activity with and without the presence of an acoustic lure. Error bars represent the standard error of the mean.

DISCUSSION

Our small sample size of tricolored bat captures and bat passes made it difficult to assess whether the acoustic lure was a reliable tool for studying the species. We cannot confirm if our tricolored bat captures were random or if individuals were responding to the lure; however, our success of capturing two individuals with the lure compared to none without the lure was encouraging.

We observed an increase of acoustic activity for big brown and Indiana bats when using the acoustic lure. Similarly, Loeb and Britzke (2010) and Quackenbush et al. (2016) observed similar patterns with acoustic lures for the two species. To our knowledge, we are only the second study in North America to document Indiana bats responding to heterospecific calls (Quackenbush et al. 2016). Although we cannot evaluate either species' capture success due to our low sample size and limited capture efforts, our acoustic results suggested that more captures would occur for the two species when using a lure if this survey was reattempted.

We did not observe a significant increase of captures when the lure was used compared to without the lure, challenging results highlighted in other lure studies (Hill and Greenaway 2005; Quackenbush et al. 2016; Braun De Torrez et al. 2017; Samoray et al. 2019). It is unclear if similar results would be observed if we used tricolored bat social calls instead of distress calls or used the two vocalizations simultaneously. Assessments in which call type would be best to use is difficult to evaluate since more information is needed to better understand the function of social echolocation in tricolored bats. Research suggests that the species does not form large social maternity colonies similar to species of *Myotis* (Veilleux et al. 2003); therefore, we do not know if tricolored bats would react strongly to conspecific social calls. Winter hibernacula surveys support a lack of social clustering structure for the species as characterized by species of *Myotis* (Langwig et al. 2012), since they disperse themselves in caves or form small clusters (Briggler and Prather 2003).

We expected our results for tricolored bats to mirror studies on their distant relative, soprano pipistrelle, where modified distress calls increased acoustic bat activity when the lure was used (Russ et al. 2005a). One significant difference between our study and Russ et al. (2005) was that the researchers used modified distress calls that were both within and outside the normal range of soprano pipistrelle echolocation. In another study, tricolored bat captures were 65% greater when a lure played an Indiana bat social or distress calls (Samoray et al. 2019). Which call type (e.g., social versus distress) resulted in a more robust response in tricolored bats was not evaluated by Samoray et al. (2019), however.

We find it particularly concerning that we captured species in two areas but did not record their echolocation at the capture time. This observation highlighted either a possible limitation in our acoustic detectors or demonstrates that bat echolocation can be highly variable when receiving information (Broders et al. 2004; Britzke et al. 2013). A lack of echolocation may be explained as a method to avoid ‘sonar jamming’ (Chiu et al. 2008) or through weak echolocation pulses known as ‘whispers’ used for approaching prey (Russo et al. 2007). Understanding the level of acoustic bat activity at sites is challenging since our detectors' known range varies with environmental conditions, and we do not know how far a bat can detect noise (Schnitzler and Kalko 2001).

Recommendations for Future Studies

Our findings suggest that future research should determine if distress and/or social calls from conspecifics or heterospecifics attract tricolored bats during mist net surveys. Areas with abundant populations or similar relatives would serve as excellent study sites. Our increased bat activity results from big brown bats and Indiana bats offer future opportunities to investigate why the species responded to the lure. Big brown bat echolocations are within a similar frequency range as our distress calls used. It is unclear what communication function the distress calls convey to the species, and further research is needed to investigate why they respond to heterospecific vocalizations. Our research contributes to other lure studies by demonstrating that Indiana bats are flexible to the vocalization call types that are broadcasted with acoustic lures. This result suggests that acoustic lures could benefit the study of this endangered species. Unlike big brown bat echolocation frequencies, our distress calls were not within a similar frequency range as Indiana bats and it is unknown why the species positively responded to the distress call. Future acoustic lure studies should explore the optimal placement of acoustic lures over ponds. Infrared cameras may assist in future studies by assessing the best location for mist net placement over ponds when an acoustic lure is used and evaluate if bats are avoiding the mist net.

CONCLUSION

In areas affected by WNS, capturing imperiled species such as tricolored bats are a game of chance with a low probability of success. Our results may not be statistically significant, but we argue that even a few additional captures were better than none, and in our research 52 additional captures occurred in one field season with the use of the lure. Researchers studying Indiana bats through mist netting would benefit through the use of an acoustic lure. We did not observe a negative effect from the acoustic lure on other species in our study, and therefore suggest there are limited downsides to deploying the lure beyond time and expense. Our results contribute to a growing number of acoustic lure studies that seek to understand acoustic lures' function and how they can be incorporated into mist net surveys. We encourage researchers to explore how to optimize future lure research to increase the capture of tricolored bats and other WNS susceptible imperiled species. Contributions from additional research extend the understanding of how acoustic lures can be easily incorporated into North American bat studies and prevent the failure to document imperiled species when they are still present.

CHAPTER III

PERCEPTIONS OF BATS AND THEIR CONSERVATION IN RURAL MISSOURI

ABSTRACT

Fourteen years have passed since white-nose syndrome (WNS) was introduced in North America. Bat conservation and educational efforts have increased as WNS-susceptible bat populations have declined, although few studies have assessed its efficacy. In this study, we assessed rural Missourian's WNS awareness, knowledge of bat natural history, attitudes towards bats, and trust level in the Missouri Department of Conservation (MDC). Goals from our research included informing the MDC of public misperceptions of bats, which could help generate strategies for improving bat education. Our research stressed that Missouri's WNS and bat ecological knowledge was limited, despite educational resources' availability. The observance of gated caves best explained our respondent's knowledge of WNS. Respondents with higher education were the only group able to identity Missouri as a WNS-positive state. The trust of the MDC was high among respondents. Respondents that perceived bats positively viewed them as a form of insect control. We suggest bat conservation efforts can be improved in southeastern Missouri by increasing the delivery of bat educational initiatives to include those in

public health messaging, normalizing bat houses, and to maintain trust in the MDC when making management decisions.

INTRODUCTION

Bats are threatened by habitat loss, wind energy development, and diseases like white-nose syndrome (WNS; Frick et al. 2019). Of the conservation issues that wildlife face, WNS is one of the most devastating wildlife epidemics (Frick et al. 2016). WNS associated bat mortality assessed by biologists during 2011 estimated that over five million bats have died from WNS since 2007 (WNS Response Team 2020). That number is likely higher today since the disease has spread to 35 states and seven Canadian provinces (WNS Response Team 2020). Federal, state, and non-governmental organizations have collaborated to create WNS working groups and a national response plan to improve the public's awareness of WNS, bat conservation, and ecosystem services provided by bats (e.g., pest control and pollination; USFWS 2011, WNS Response Team 2019*b*).

Public support for wildlife conservation is shaped in part by education. Some species require greater educational efforts than others to reverse any perceived negatives attitudes (Bexell and Feng 2013). Frightening encounters develop negative perceptions, thus requiring intensive education efforts to increase positive associations. Bat human dimension studies indicate that the public is polarized regarding bats. Some studies suggest that people are less likely to appreciate bats because they are perceived as disgusting, diseased, or damaging to agricultural economies (Kellert 1985; Rego et al. 2015; Kingston 2016; Aziz et al. 2017), while others suggest that bats are positively perceived as taxa that provide ecosystem services for humans (Sexton and Stewart 2007; George et al. 2016; Fagan et al. 2018). Educational efforts, when designed effectively, can improve positive values towards bats. For example, bat knowledge and values were assessed through focus groups at the Great Lakes Bat Festival, held annually in Detroit, Michigan (Hoffmaster et al. 2016). Festival attendees retained educational knowledge they learned during the event, and their overall values of bats increased, thus deepening their desire to protect bats (Hoffmaster et al. 2016). Attempts made by agencies to fulfill deficiencies in the public's knowledge of bat natural history and WNS still need to be evaluated if they are successful for increasing bat conservation (Sexton and Stewart 2007; Hoffmaster et al. 2016; Fagan et al. 2018).

The current understanding of bat natural history and WNS awareness in southeastern Missouri is unknown. Bat conservation in rural areas such as southeastern Missouri may be challenged by the need for natural resources as a source of income in an impoverished area (U.S. Census Bureau 2018). Timber harvests are a valued economic activity in southeastern Missouri. Timber harvests were estimated to contribute 9.7 billion dollars to Missouri's 2018 economy (Treiman 2019). Two threatened bat species in Missouri are currently affected by forest management: Indiana bat *Myotis sodalis* and northern long-eared bat *Myotis septentrionalis*. Harvest limitations are in place to protect these species, such as a 300-acre no-harvest buffer around known Indiana bat maternity roosts (Ziehmer and Draper 2016). If southeastern Missouri residents are unaware of bat natural history or ecological benefits, they may not understand why logging restrictions are in place to protect some bat species. This misunderstanding may lead to a lack of trust and support for the Missouri Department of Conservation (MDC) when timber management decisions are made to benefit bat conservation. Measuring the level of trust in management agencies from the public is critical to assess before implementing management changes that impact local economies. If timber harvest stakeholders perceive the MDC as a trustworthy agency, they would be more likely to change their behaviors or respect the agency's recommendations.

In this study, we first assessed respondents' knowledge of bats and WNS in Missouri. Results from this inquiry provide the MDC with an awareness of gaps in bat-associated knowledge, which could help suggest methods that improve the public's perceptions of bats. Secondly, we determined respondents' negative or positively held perceptions of bats (e.g., values towards bats). Knowledge of bat associated values in rural Missouri allows the MDC to assess how likely it is that the public would support bat conservation. If it were discovered that most respondents held negative bat values, future research would need to focus on initiatives that promote positive attitudes. Enhancing the perceived perceptions of bats requires not only an understanding of where educational gaps in bat knowledge exist but input from the beneficiaries and stakeholders of the MDC areas who are impacted by bat declines, potentially unbeknownst to them. Finally, we measured public trust in the MDC. Before the MDC can recommend initiatives that increase positive perceptions of bats or related knowledge, we must ensure that respondents would be willing to listen to MDC officials.

METHODS

Study Area

Within Shannon, Carter, and Reynolds counties, we selected two towns per county and distributed the sites as far apart from one another as possible to increase the spatial distribution of the survey (Fig. 3.1). These counties were selected due to their similarity in population demographics, proximity to MDC areas, and the presence of imperiled bat species. We focused on grocery stores within each survey town as the primary location to interview individuals, but we did include two gas stations in towns where grocery stores were absent. Public land owned by entities such as the National Park Service, Army Corps of Engineers, Missouri Department of Natural Resources, or the U.S. Forest Service were near our survey sites. We assumed we would receive surveys from people who spend time outdoors and possibly interact with MDC properties by distributing our surveys in towns that were nearby public land.

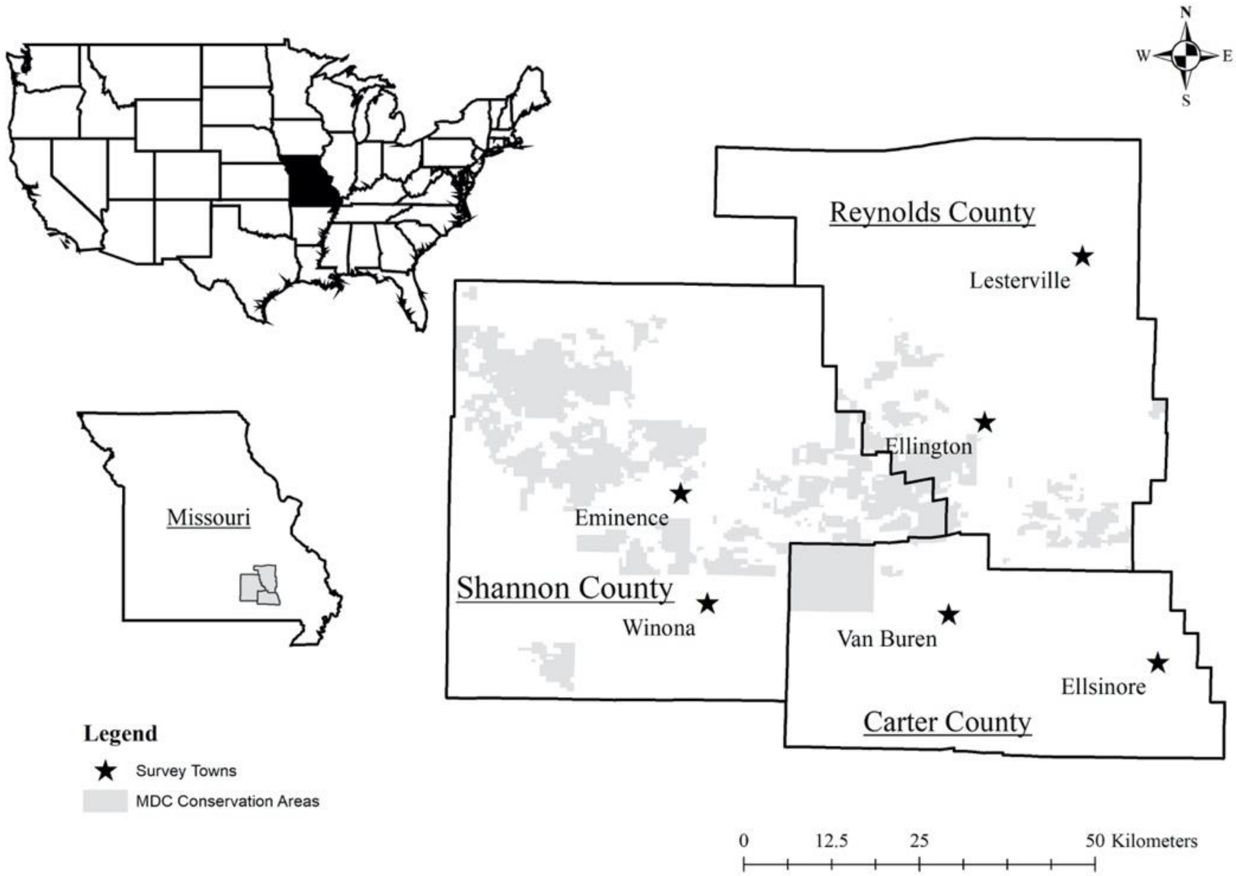


Figure 3.1 Map of survey sites selected for a human dimension study on bats in southeastern Missouri.

Sampling Methodology

Survey data was collected between June 6 and August 9, 2019. Our targeted stakeholders included Missouri residents 18 years or older. We standardized our survey effort at each site by administering surveys at random times between noon and 20:00, with each attempt lasting two hours. Our goal was to survey 100 individuals, and we divided our surveys between the six sites. If we were unable to complete our survey quotas at a location after our two-hour survey, we finished data collection later.

Surveys were conducted alongside building entryways or as directed by the property's business owner. We conducted convenience sampling for any person visiting the sample site while also randomizing our sampling effort by asking every third person over 18 years old and a Missouri resident to participate in the survey. Individuals that agreed to participate in our survey completed surveys on site. We monitored traffic flow into buildings with a tally counter. When asked if they would participate in our survey, we recorded individual responses, such as the number of 'Yes and No' answers to Missouri resident inquiries and if they were at least 18 years old. If our inquiry resulted in a survey refusal or a non-Missouri resident response, we reset our count interval until the next third person entered the building.

Bat Human Dimensions Survey

All of our objectives included socio-demographic components to understand if they influenced respondents' survey answers. We assessed responses to reflect a respondent's education level, sex, age, income, and hometown population size. Our income, population, and age demographics were grouped as those that appear within the U.S. Census. We evaluated how much respondents knew about WNS, WNS in Missouri, and bat natural history. We assessed respondents' answers by evaluating relationships between socio-demographic criteria and the use of educational materials.

We measured our respondents' motivations towards activities that improve bat conservation, such as creating bat houses, landscaping for bats, donating money, sharing knowledge with others, participating in educational activities, and using less energy. We assessed their associated bat values by asking respondents if they could write at least three benefits bats provided people. This assessment allowed us to identify the known ecosystem benefits bats provided humans while highlighting less known ecosystem benefits that can be enhanced through improved future educational messaging. We measured positive and negative perceptions of bats by asking respondents the question, “What comes to mind when you think of bats?” We categorized responses into three groups: neutral, positive, or negative. Positive responses included words that were associated with ecosystem services or attractive physical appearance, whereas negative responses included words that were associated with a disease or frightening images. Neutral responses included words that could not be identified as positive or negative. Some respondents selected two words for what bats brought to mind; we separated the responses into two different values.

We measured the level of trust respondents held regarding the MDC using a Likert scale (1=Strongly agree, 2=Somewhat agree, 3=Neither agree nor disagree, 4=Somewhat disagree, 5=Strongly disagree). We evaluated whether respondents were willing to recommend the MDC to others on a Likert scale (0=Not likely at all to 10=Extremely likely). We included open-ended questions for why respondents were or were not likely to recommend the MDC and other comments they wanted to share.

Data Analysis

Our data analysis included descriptive statistics for our survey results, such as the percentages of ‘Yes and No’ questions. If a respondent did not answer a question, we omitted it from the analysis. We only included ‘Yes or No’ responses from individuals and omitted any answers that were less informative such as, ‘Maybe.’ We kept answers for knowledge of WNS in Missouri as 'Yes, No, or I do not know.' We determined the consistency between each respondent's 'Yes or No' responses through Cronbach's alpha (Appendix II; Section 2 questions #11-13, 15, 19-21, 26). We used Fisher's exact test of independence with two-tailed results to determine relationships between selected respondent answers and socio-demographic groups. For the two questions that used a Likert scale as a response, we used Spearman's rank correlation for ordinal demographic predictor variables and Kruskal-Wallis test for nominal demographic predictor variables. In these analyses, we ranked the MDC trust scores and willingness to recommend the MDC with demographic variables.

RESULTS

Survey Effort

We spent 24.5 hours over 14 days collecting survey data. Out of the 260 individuals asked to participate in our survey, 32.2% complied, 39.1% declined, 0.8% were under 18 yrs. old, and 11.2% were not Missouri residents. Additionally, 5.9% volunteered to participate in the survey before our inquiry, 3.1% requested surveys be read to them, and 6.5% ignored surveyor requests. We omitted two surveys that included answers from people that did not fit our age criteria for a total sample size of 98 individuals.

Demographic Distribution

The survey respondents' socio-demographics were primarily middle-aged men (n=57, average 52 years old, SD±17 58% of respondents) and women (n=41, average 50 years old, SD±16, 41.8% of respondents). Six male respondents did not provide their age. The majority of survey respondents lived within the study area's counties (65.8%), with most respondents residing within Shannon County (27.8%). The level of education held by our respondents was similar between a high school diploma (25%), attending some college (24%), and a college degree (25%). Only 10.4% of respondents had a graduate degree or professional certificate. Most respondents (29%) had a low household annual income, between $0 –24,999.00, followed by a tie (23% for each category) for respondents that had a household annual income of $25,000.00 – $49,000.00 and $75,000 – 100,000.

Bat Natural History and White-nose Syndrome Awareness

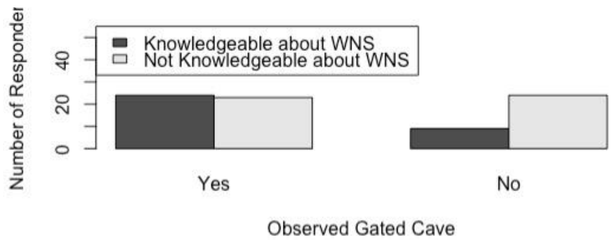
Respondents answered questions on their knowledge of bat natural history and myths consistently (Cronbach's alpha= 0.75). Only 35.4% of respondents were able to correctly identify a bat species found within Missouri. The most frequently correctly reported bat species was the brown bat (27%), followed by the gray bat (4.6%). Two individuals separately wrote tricolored bat *Perimyotis subflavus* and Indiana bat for their answers. Two species of 'brown bat' live in Missouri, commonly known as the big brown bat *Eptesicus fuscus* and the little brown bat *Myotis lucifugus.* It is unclear if respondents are referring to one, both or a different local species interpretation. We treated 'brown bat' as a correct answer in our analysis. Respondents listed a few species of bats that were exotic or non-existent; 14% of respondents recognized 'fruit bat' as a species of bat, while two non-existent species, 'black' and 'cave bat' were written. The only variable that predicted local bat species' knowledge was if they read MDC signs ($p=0.01$); however, this relationship was only significant for individuals who either incorrectly identified a species or could not identify a species of bat in Missouri. Socio-demographic comparisons for species knowledge such as census age group, sex, and the highest level of education were not associated with bat knowledge

Survey respondents were not knowledgeable of the correct answers for our three bat myths. Almost half of the Missouri respondents knew bats were not blind (49%). One respondent believed that only some bats were blind, and one respondent did not know the answer. Over half of the respondents incorrectly believed that bats could get tangled in human hair, while most people knew Missouri bats do not drink blood (53.8% and 88.5%, respectively). Three respondents answered ‘Yes and No’ for whether they believed Missouri bats drank blood. Three respondents clarified their answer by indicating that bats preferred cow blood or blood that did not belong to humans. Cumulatively between respondents, only 25.2% correctly identified all three myths, 3.2% guessed incorrectly for all myths, and 71.8% only knew the correct answer for one or two myths. The sex of respondents and education level were not significant indicators of bat myths ($p>0.05$). Missouri Department of Conservation educational material use and media preferences (e.g., social media, websites) did not influence the respondent’s knowledge of myths. Our results suggested that 68% of respondents used MDC educational media materials.

Respondents in our study were misinformed about bat species and, unexpectedly, about WNS. Most respondents never heard of the disease (50.6%), while only 39.1% of respondents correctly identified that WNS was a fungal bat disease. When asked whether WNS was in Missouri, 67.7% of respondents were unsure if it was found in Missouri. The respondent’s level of education, use of MDC educational materials, or MDC brochures, signs, word of mouth (e.g., information from MDC employees) did not improve their knowledge of WNS. We found no support for respondents' knowledge of WNS by those who preferred websites or social media. Out of 83 respondents who responded to whether they observed bat declines, 63.9% of people did not notice bat declines, and 36.1% of respondents noticed bat population declines.

Respondents were aware that bats used caves for habitat. Respondents that observed gated caves correctly identified WNS ($p=0.04$) and understood that caves were gated to protect bats ($p=0.02$) (Fig. 3.2). The majority of respondents (57.6%) observed a gated cave or closure. Some respondents were aware that gated cave closures were to protect bats (27.3%), although other respondents thought that the caves were closed due to vandalism (26%) or to protect cave formations (20.2%). Some respondents were not sure why the caves were closed (14%).

A



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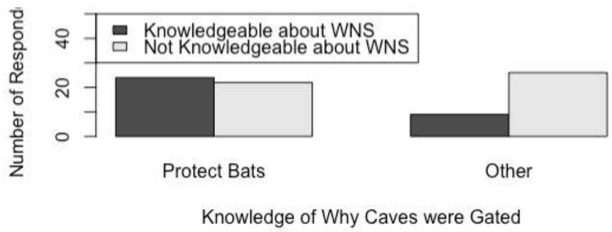

Figure 3.2 Knowledge of WNS through the observance of a gated cave (A) and understanding that caves were gated to protect bats (B).

We evaluated factors that could influence respondents' correct answers about general WNS awareness and knowledge of WNS in Missouri. We found that the difference between the WNS awareness groups (e.g., Yes, No, vs. I don't know) was significant, and the greatest representation was within the 'I don't know' group ($p<0.001$; Fig. 3.3). Respondents within the 'I don't know' group had correctly identified WNS but were unsure if it occurred in their state. Only one respondent selected 'No' WNS was not in Missouri, although they could correctly identify WNS as a bat disease. The analyses included in this section only included respondents who knew the correct general knowledge of WNS and that WNS was in Missouri. The use of any educational materials, observance of gated caves, or knowledge of bat declines did not improve knowledge of WNS in Missouri. Level of education was the only variable that differed across respondents regarding their answers, with individuals with a college degree or higher were more knowledgeable about WNS in Missouri (p=0.05; Fig 3.4).

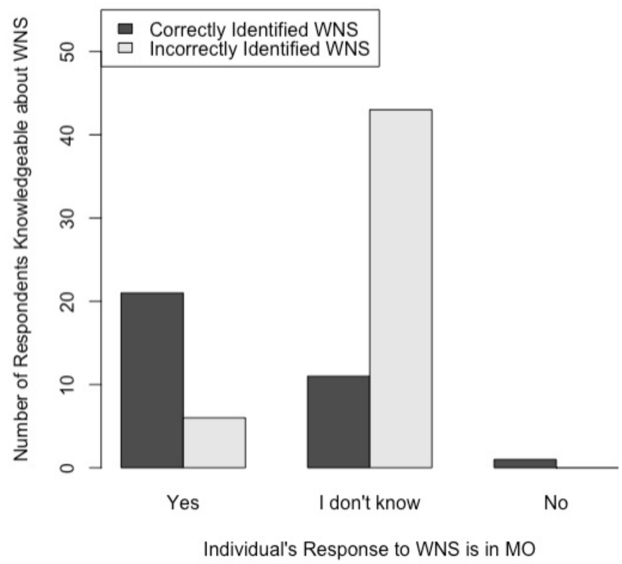

Figure 3.3 Respondent answers for correctly and incorrectly identifying WNS, and selecting ‘Yes, No, I do not know’ for if WNS was in Missouri.

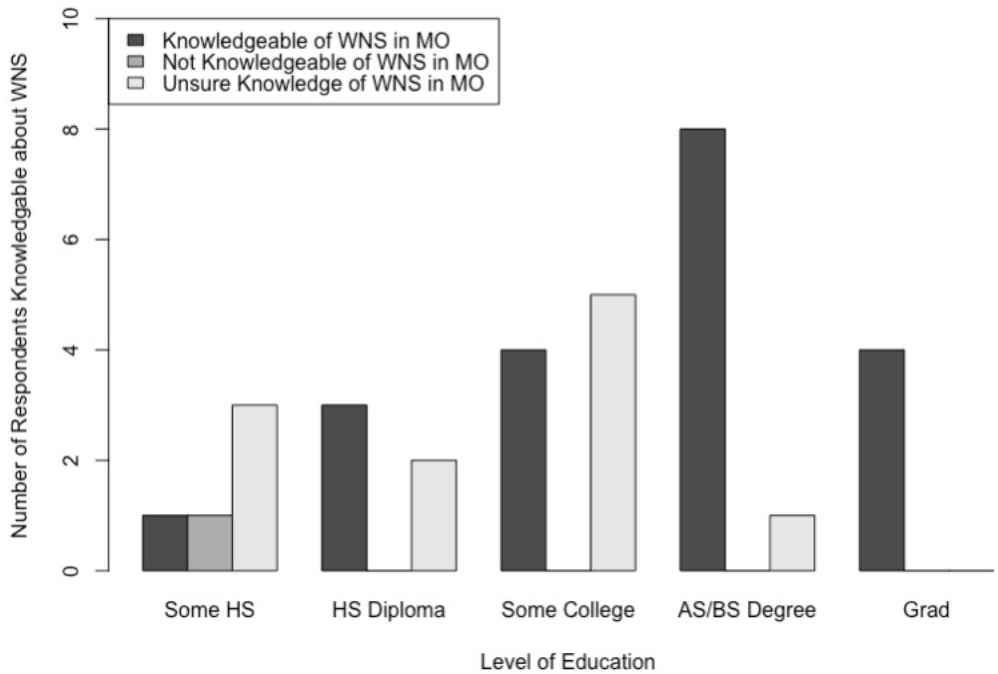

Figure 3.4 Knowledge of WNS and its occurrence in Missouri compared to education status.

Attitudes for Bat Conservation

Respondent answers to “What comes to mind when you think of a bat?” varied in their core attitudes, either positive, negative, or neutral. Respondents were more likely to use neutral (32.6%) or positive (30.6%) words compared to negative words (21.3%). Only one individual wrote that they could not think of a word associated with ‘bat.’ Insect control or eating bugs was the most commonly used positive word or phrase (25% of the words used, Appendix III). The most frequently used negative word choice included those associated with disease (9.3% of the words used). Some respondents referenced negative popular culture answers that evoked fearful emotions, such as

Dracula and vampires (4.6% of the words used). Neither census age group, sex, the highest level of education, or the amount of time a respondent visited MDC areas explained bats' perceived attitudes.

The majority of survey respondents (67.3%) could think of at least one benefit that bats provided humans. Fewer individuals could think of a second (19.4%) and a third (8.2%) benefit provided by bats. Out of all of the first benefits written by respondents, most respondents listed insect control or eating insects (91%). The most common written benefit of the second listed benefit included the use of guano as fertilizer or in cosmetics (57.9%). The third most listed benefit mentioned was ecosystem health (37.5%). Respondents, in turn, were willing to participate in some activities that benefited bat conservation. Of the activities listed, respondents were willing to build bat houses (24.3%), share learned knowledge with friends and families (23%), and reduce energy use (18.4%).

Missouri Department of Conservation Use and Values

Belief in MDC's trustworthiness as an agency was mostly positive among respondents; over half of respondents strongly agreed the MDC was a trustworthy agency (55.7%). Belief in MDC trustworthiness did not differ between age, income, sex, or level of education. Respondents were likely to recommend the MDC as an organization to other individuals (N=84; average score: 8.1 ± 2.3 SD with ten being "extremely likely" and zero as "not likely to recommend"). According to respondents' explanations of their provided scores, some reported negative scores based on recent negative experiences on MDC owned land (see scores and associated comments in Appendix IV). Sixteen individuals did not provide any comments for why they ranked their response. We compared respondents' willingness to recommend the MDC to others against socio-demographics, including age, sex, and income, but they did not differ in their recommendations of the MDC to others. The education level indicated a weak positive relationship (ρ=0.28, p=0.01) for willingness to recommend the MDC. Individuals with a higher education level were more likely to recommend the MDC than other educational groups (Fig. 3.5). Additional comments about the MDC and wildlife from respondents are included in Appendix V.

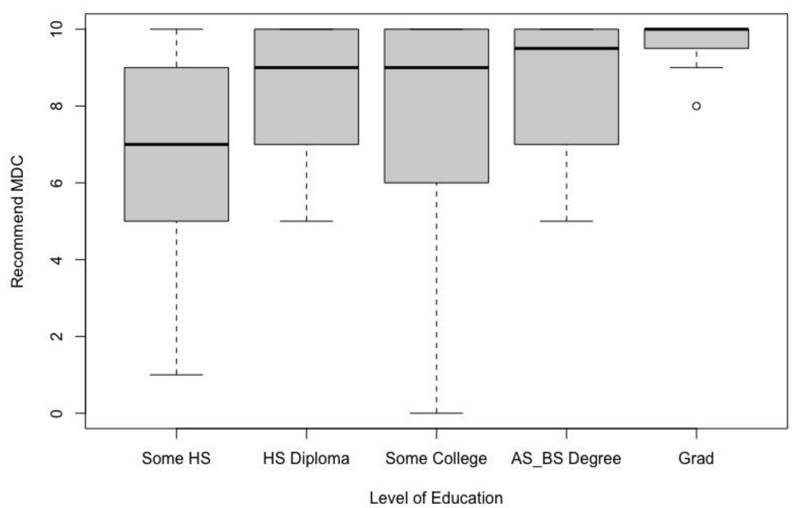

Figure 3.5 Level of education and willingness to recommend MDC. A rank of 0 demonstrated that respondents were not likely to recommend the MDC, whereas a rank of 10 indicated that respondents were extremely likely to recommend the MDC.

DISCUSSION

Bat Knowledge

Missouri respondents knew little about bat natural history and WNS, regardless of using MDC educational materials and the respondent's level of education. These results contrasted our expectations. We anticipated respondents would have had greater bat knowledge if they engaged in the MDC's educational materials or had more education (Kellert 1985). Knowledge of bat resources went down when our respondents read signs. This awareness underscores the MDC's lack of communicating bat threats and natural history to the public when they visit MDC areas. Research on bat-related knowledge is limited. Most bat human dimension assessments include those within similar age groups (e.g., children) or locations where the highest level of education was elementary or high school (Kahn et al. 2008; Prokop and Tunnicliffe 2008; Musila et al. 2018). The respondents' education level did not increase the general knowledge of WNS, although it did influence respondents' knowledge of WNS occurring within Missouri. We suggest that our study respondents either struggled to retain the information they read about bats or were disinterested in bats. Deficiencies in bat-related knowledge are reported in other human dimensions studies. For example, Sexton and Stewart (2007) found that respondents knew little about bat ecology, despite the respondent's attempts to learn about bats from magazines/newspapers, television, and non-experts (e.g., friends and family). When asked in Great Smoky National Park, only 39% of visitors knew about WNS (Fagan et al. 2018).

We discovered that respondents were more likely to know of WNS if they observed a gated cave, a result not evaluated in other bat human dimension studies. Two percent (e.g., 120) of Missouri's 6,000 caves are gated, with a few gated caves in Shannon County (Elliott et al. 2010). Our findings indicate that respondents knew the purpose of gated caves was to protect bats. It is unknown if the respondents' previous observance of a gated cave sparked their curiosity or if they had read a sign at a gated cave that explained the closure. In Shannon County, Powder Mill and Round Spring Caves are two local gated caves and are visible to the public. Neither caves had signs that provided information about WNS. A small sign located inside of Powder Mill Cave labeled the cave as a refuge for cave-dwelling organisms. Round Spring Cave had an interpretive sign outside of the cave that informed visitors about endangered bats but not about WNS. Adding WNS information on signs may improve overall knowledge of WNS and the ecology of bats that use the caves. The generalization of bats as a taxon was highlighted in our survey by words used to describe bats species, such as 'the bats that live in caves,' or 'brown bat.'

Timber harvesting is a significant economic contributor to the area. Information provided by the MDC highlights the use of caves as bat habitat, but little about bat's habitat use in the forest. Respondent's knowledge of bat's roles in the forest can be amplified in the future by providing additional information about forest management techniques and how various management activities benefit bats. Educational materials can provide more detail on how bats rely on different forest habitats to rear young. Forest management activity that benefit bats include non-linear forest openings (O'Keefe et al. 2009) for the tricolored bat *Perimyotis subflavus*, and prescribed fire activity benefits northern long-eared bats *Myotis septentrionalis* (Lacki et al. 2009). Timber management techniques not only benefit bats, but benefit other wildlife populations, such as white-tailed deer *Odocileus virginianus* (Lashley et al. 2011). Several respondents indicated that hunting was an activity they participated in on MDC lands, and so advocating timber management for bats may obtain more support from the public as a means for improved hunting experience.

Printed educational materials about bats are available to Missouri residents within some MDC areas and nature centers; however, this information's availability did not influence the respondents' knowledge of WNS. Educational resources about bats are abundant on digital MDC platforms such as websites and affiliated agency social media pages (MDC 2020a), compared to printed materials. Access to digital media educational resources may be limited for southeastern Missouri residents, especially given the poverty rate and isolation from urban areas. The Missouri Department of Economic Development indicated that the three research counties in this study were underserved in broadband internet access, especially in Shannon and Carter Counties (MODED 2020). Available printed MDC literature and educational programs in the study areas need to be improved to increase knowledge about bats. Educational efforts need to be expanded beyond identifying species of bats, or some common facts about bats, and instead strengthen the benefits bats provide local respondents and inform about bat conservation threats in Missouri. State public health messaging can contribute more ecological information when talking to the public about zoonotic diseases, such as rabies, by discussing One Health language. One Health language, an approach that recognizes the interaction of humans and the natural world as a reflection of the health of the environment and people (CDC 2020).

Future human dimension bat studies should evaluate if respondents learned about WNS through a guided cave tour. We recommend that gated caves have more information about WNS and bat natural history through interpretive signage. Alternative survey methods such as focus groups and social-learning activities help understand more effective educational media techniques. Bat-featured events such as festivals or those during National Bat Week in October help cultivate bat conservation and dissolve some of the misconceptions commonly held of bats (Bexell and Feng 2013). These initiatives include some of the first steps that help increase knowledge. Activities that respondents could participate in during special events could include those most popular in our survey, such as building bat houses, sharing knowledge with their families, and reducing energy use. After these events, respondents could participate in follow up questions after the event to understand which methods were most efficient for retaining knowledge.

Attitudes Towards Bats

Recognition that bats control insects contributed to more positively bat associated values in our study, similar to other bat human dimension research (Sexton and Stewart 2007; Fagan et al. 2018). When people feel positively towards a resource, they are more likely to support the resource (Tarrant et al. 2016). The recognition that bats benefit humans through insect control could be expanded to include benefits of which respondents may not be aware. Installing a bat house not only benefits bats in a person's backyard but helps insect control around their homes by supporting a colony of bats. One particular species of bat that consumes a lot of mosquitos is the little brown bat *Myotis lucifugus* (Wray et al. 2018). The consumption of mosquitos can mitigate the transmission zoonotic diseases such as West Nile Virus (NIOSH 2018). Little brown bats have been hit hard from WNS both range wide and in Missouri (Ingersoll et al. 2016;

Colatskie 2017). The species uses attics for maternity roosts, but can successfully occupy bat houses (Thomas and Jung 2019). The installation of a bat house can shift occupancy from people's homes to a more desirable location outside. Bat populations this way can remain sustained while providing humans with beneficial ecosystem services. One respondent elaborated on their frustration from bats occupying their homes by expressing, "They crap all over my barn!" Respondents specifically listed mosquito control as a benefit, instead of the control of larger insects that damage agricultural crops such as moths and beetles (Boyles et al. 2011). Insect control provided by bats within the corn industry is estimated to save farmers one billion U.S. dollars in pesticide use and associated insect damage crop losses (Maine and Boyles 2015). Consideration of these additional benefits in educational messaging provides the public with relatable reasons of why they should care about the bats living in their communities.

Trust in the Missouri Department of Conservation

Trust in the MDC was overall high among respondents. Of the respondents we surveyed, most respondents viewed the MDC positively and were willing to recommend the agency to others. The level of trust in an agency is vital in natural resource conservation. Previous studies in conservation and social trust in agency management decisions have found more generous support from the public when individuals trust the agency implementing management actions (Cvetkovich and Winter 2003; Sponarski et al. 2014). Successful conservation of imperiled bat species in southeastern Missouri relies on the public's support and inclusion in the MDC's management decisions. Recent management decisions in the area, such as feral hog control and elk reintroduction, resulted in a lack of trust from some respondents, who felt misunderstood as indicated by their responses and ranked scores (Appendix IV & V). The public must be informed of management decisions and be provided with ecological knowledge of why bat's conservation is necessary to maintain a healthy ecosystem while sustaining timber harvest economies. Missouri residents unfamiliar with bat ecology may be angered by increased conservation measures for imperiled bat species that limit logging activity. Individuals may believe any bat is 'just a bat' and may not comprehend that species require different habitat needs or why species are imperiled. As WNS progresses in southeastern Missouri, additional timber harvest activity restrictions may be implemented to protect populations. The MDC's best interest to provide appropriate educational messaging about bat ecology and WNS to southeastern Missouri residents when proposing new forest management objectives. Assessments about how respondents feel about bat management and timber harvesting should occur.

CONCLUSION

Respondents positively viewed bats for their insect control services; however, they were unaware of WNS, bat species in Missouri, or myths associated with bats. Our respondents' lack of bat knowledge meant that they were less likely to recognize bat conservation threats in their area. Connecting the public to forest management techniques that benefit bat populations and humans for hunting opportunities create a desirable experience that fosters bat conservation. Expanding the public's knowledge of bat insect control to agricultural pest and zoonotic disease maintenance strengthens bat' benefits beyond eliminating insects that are considered annoying, such as mosquitoes. The

MDC’s educational materials were available to respondents; however, these materials’ use did not improve their ecological bat knowledge or WNS awareness. Our results suggest that respondents who correctly identified WNS knew that it was a reflection of an observed gated cave. We are unaware of any other studies that have evaluated the efficacy of gated caves as a measurement for WNS awareness. Communities within our study areas may not receive equal opportunities to learn about bats since the agency primarily focuses on educational efforts on digital platforms. We suggest that future outreach efforts in the study focus on improving educational opportunities for the public to determine the most effective method to share bat conservation issues. Normalizing bat houses and increasing social learning efforts shift current perspectives from diseased animals to ecologically valuable animals. Changes created within these levels stimulate empathy and compassion for bats at a time where the public’s support is necessary, unbeknownst to them.

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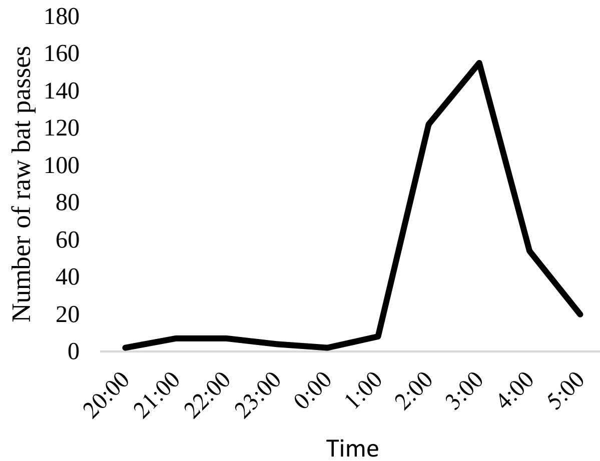
APPENDIX



Appendix I: Total number of tricolored bat passes by time.

Appendix II: Survey distributed to rural Missouri residents

Missouri Resident Questionnaire: **Missouri Wildlife and Public Land Use**

Dear Missouri Resident:

Thank you for participating in this survey. This survey will help a graduate student at Pittsburg State University understand Missouri residents' attitudes toward wildlife and public land. From this survey, we will understand if available educational materials within Missouri Department of Conservation Conservation Areas are sufficient and enjoyable for Missourians. **This survey does not reflect the views or attitudes of the Missouri Department of Conservation. Results from this survey will not guarantee any management changes or production of new educational materials from the Missouri Department of Conservation.**

Risks and Confidentiality
We will not collect names or any other personal information. All collected data will be used for research purposes.

Contact
If you have questions at any time about this questionnaire, you may contact the graduate student, Amy Hammesfahr, at ahammesfahr@gus.pittstate.edu. For additional information regarding participation of this research, please contact the Pittsburg State University's Human Subjects Committee officer, Brian Peery, at 620-235-4175.

Instructions
This survey will take less than 10 minutes to complete. Please answer all questions to the best of your ability. Completing the survey indicates your consent. You may withdraw your consent at any time. Please return this survey to the individuals distributing this survey.

Thank you for your time,

Amy Hammesfahr
Graduate Research Assistant
Department of Biology

Missouri Resident Questionnaire:
Missouri Wildlife and Public Land Use

SECTION ONE: DEMOGRAPHICS

In this section, we would like to get a profile of the individuals taking this survey.

1. What year were you born? ________

2. Are you **male** or **female**?
   If you identify otherwise, write it here: ______________________

3. Are you a Missouri resident?
   Circle **YES** or **NO**

4. What county do you live in?
   ____

5. What is the population size of your hometown?
   a. Less than 2,500
   b. 2,501 – 10,000
   c. 10,001 – 50,000
   d. Over 50,000

6. What is the highest level of school that you have completed?
   a. Some high school, no diploma
   b. High school or equivalent (GED)
   c. Some college, no diploma
   d. Associates or Bachelor's degree
   e. Graduate or professional degree

7. What is your household annual income range?
   a. $0 – $24,999
   b. $25,000 – $49,999
   c. $50,000 – $74,999
   d. $75,000 – $100,000
   e. $100,000 or more

SECTION TWO: MISSOURI WILDLIFE & CONSERVATION

In this section, we want to determine the natural history knowledge and values of Missourians.

8. Name one species of bat that lives in Missouri:___________________

9. Where have you seen bats?
   Check off all that apply.
   ___My home or attic
   ___Barn or outdoor storage building
   ___Around ponds or waterways
   ___Around open fields
   ___I haven't seen any bats
   If other, describe:
   ____
   ____

10. What comes to mind when you think about bats?
    ____
    ____
    ____

11. Are bats blind? Circle **YES** or **NO**

12. Can bat wings get tangled in your hair? Circle **YES** or **NO**

13. Do Missouri bats drink blood?
    Circle **YES** or **NO**

2

Missouri Resident Questionnaire:
Missouri Wildlife and Public Land Use

14. List 3 ways that bats are beneficial to humans.

    1.___________________________
    ____

    2.___________________________
    ____

    3.___________________________
    ____

15. Have you noticed a decline of bats in your area within the last ten years? Circle **YES** or **NO**

16. What things would you be willing to do for bat conservation? Check all that apply.
    ___ Build a bat house
    ___ Landscape to attract bats
    ___ Teach your friends and family about bats
    ___ Donate to organizations that protect bats
    ___ Participate in educational activities about bats
    ___ Use less energy and water at home

17. What is white-nose syndrome? Check off all that apply.
    ___ A contagious disease bats can spread to humans
    ___ A type of frostbite found on bats
    ___ A contagious, deadly fungal disease bats get in caves
    ___ I've never heard of white-nose syndrome before

18. Is white-nose syndrome in Missouri?
    a. Yes
    b. No
    c. I don't know

19. Do you consider yourself a technical caver, and a member of a grotto group? Circle **YES** or **NO**

20. Do you explore caves outside of cave tours that does not require technical equipment (ropes, harnesses, etc)? Circle **YES** or **NO**

21. Have you noticed any caves gated off, preventing access? Circle **YES** or **NO**

22. Why are the caves gated? Check off all that apply.
    ___ To protect endangered salamanders
    ___ To prevent access due to vandalism
    ___ To protect sensitive formations inside the caves
    ___ To protect bats
    ___ I'm not sure why

23. How does it make you feel when you find a cave that is gated, preventing your access? Check off all that apply.
    ___ Angry
    ___ Indifferent
    ___ Appreciative
    ___ Confused
    If other, describe:
    ____
    ____

3

Missouri Resident Questionnaire:
Missouri Wildlife and Public Land Use

SECTION THREE: MISSOURI DEPARTMENT OF CONSERVATION

In this section, we want to determine how Missourians enjoy recreating in Missouri Department of Conservation's Conservation Areas and how they seek natural history knowledge when visiting.

24. How often do you visit Missouri Department of Conservation Conservation Areas?
    a. Never
    b. A few times a year
    c. Frequently, several times a year
    d. Often, at least twice a month

25. Check off all the things you like to experience when visiting Missouri Department of Conservation Conservation Areas:
    ___ Hunting opportunities and scouting
    ___ Camping
    ___ Solitude
    ___ Enjoying time with my family
    ___ Fishing
    ___ Hiking
    ___ Use off-road vehicles like UTVS and ATVS
    ___ Tour and drive a natural area
    ___ Look for wildlife
    ___ Look for caves
    ___ Forage for mushrooms or other wild edibles
    If other, describe:
    ____
    ____

26. Do you use Missouri Department of Conservation educational materials to learn more about the wildlife of an area? Circle **YES** or **NO**

27. If you noticed or used Missouri Department of Conservation educational materials, what form do you see/use most often? Check off all that apply.
    ___ Brochures or other handouts
    ___ Roadside or trail signs
    ___ By word of mouth from Missouri Department of Conservation employees
    ___ Missouri Department of Conservation Website before or after my visit
    ___ Social media posts
    ___Missouri Department of Conservation mobile apps
    If other, describe:
    ____
    ____

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Missouri Resident Questionnaire:
Missouri Wildlife and Public Land Use

28. What type of information do you read while visiting Missouri Department of Conservation Conservation Areas? Check off all that apply.
    ___ Signs
    ___ Brochures and other handouts
    ___Missouri Department of Conservation mobile apps
    ___ I don't read anything when visiting
    ___ I didn't notice any educational materials during my visit
    If other, describe:
    ____
    ____

29. Check off the best ways you like to learn about wildlife:
    ___Social media posts
    ___Videos
    ___Radio
    ___Websites
    ___Books
    ___Brochures and other short handouts
    ___Signs
    ___Word of mouth
    ___Magazines
    ___Newspapers
    ___Missouri Department of Conservation mobile apps
    If other, describe:
    ____
    ____

30. The Missouri Department of Conservation is a name I can trust (Select the best answer):
    a. Strongly agree
    b. Somewhat agree
    c. Neither agree nor disagree
    d. Somewhat disagree
    e. Strongly disagree

5

Missouri Resident Questionnaire:
Missouri Wildlife and Public Land Use

31. On a scale from 0-10, how likely are you to recommend the Missouri Department of Conservation to a friend of colleague? 0 is Not At All Likely, to 10, Extremely Likely. Select the appropriate number:

0	1	2	3	4	5	6	7	8	9	10

What are the primary reasons you gave that score?

____

____

____

____

32. Please write any additional comments you may have about Missouri's wildlife and public land use below:

____

____

____

____

Thank you for taking the time to complete this survey. Please return the completed survey.

--END OF SURVEY--

6

Appendix III: Respondents' (N = 85) answers to the question "What comes to mind when you think of bats?" and their answer's associated attitude score of positive, negative, or neutral.

Viewpoints of Bats	**N**	**Percentage**	**Positive**	**Negative**	**Neutral**
Bug eaters	18	21.2	X		X
Bug control	10	11.8	X		
Caves	9	10.6			X
Rabies	6	7.1		X	
Bugs	5	5.9			X
Vampires/Dracula	5	5.9		X	
Ecosystem benefit	4	4.7	X		
Blind	2	2.4			X
Disease	2	2.4		X	
Ace Ventura	1	1.2			X
Austin TX bat cave	1	1.2			X
Bat	1	1.2			X
Beautiful night flyers	1	1.2	X		
Biting	1	1.2		X	
Black	1	1.2			X
Blood suckers	1	1.2		X	
Bug catchers	1	1.2	X		
Crap all over my barn	1	1.2		X	
Cute	1	1.2	X		
Darkness n' caves	1	1.2			X
Don't close the caves	1	1.2		X	
Don't hurt them, don't touch them	1	1.2			X
Environ. Ed State Park	1	1.2			X
Flying	1	1.2			X
Flying rats	1	1.2		X	
Guano	1	1.2			X
Hey look a bat	1	1.2			X
Just fine	1	1.2			X
Make-up	1	1.2			X
Night	1	1.2			X
Nightbirds	1	1.2			X
No choice	1	1.2			X

Scary in dark	1	1.2		X	
Softball	1	1.2			X
Stay away	1	1.2		X	
They catch bugs so they're ok	1	1.2	X		
Ugh!	1	1.2		X	
White-nose syndrome	1	1.2		X	
Missing	13				

Appendix IV: Respondent answers for why they provided their score for how trustworthy the MDC was.

Score	Reason for provided score
10	Because I believe in them and watched their work through my years as a ranger.
10	love MDC
10	Trustworthy, informative
10	Our family love the outdoors. We all love hunting and fishing.
10	I have family who work for the MDC, I love the outdoors and the area I live in.
10	We use their programs in our school as well as the grant for supplies and field trips.
10	That's where I get my info on wildlife in my area.
10	They know a lot about outdoor
10	Love Missouri
10	If they need to know, I'm sure you have the answer
10	Highly respect what they do!
10	They do a good job protecting and improving habitat
10	I enjoy going to the parks to sight see, family picnics and hiking.
10	Great outdoor activities
10	I have always enjoyed visiting the conservation areas.
10	Mo Conservation areas are really pretty, we use them a lot.
10	All areas we have visited have been well marked, well maintained, and beautiful
10	Always have had positive experience.
10	One of the best in the US
10	My love for wildlife and nature
10	Information is great!
10	Because of score it is important to learn
10	Because they do a lot to help and inform our community about wildlife and nature
10	It’s always been a positive experience and trusted source for up-to-date accurate information.

10	They do great work
10	Very educational to all!
10	They can make their own assessment I don't have to agree about.
10	Good info
10	I'm very proud of MDC in our state we are way ahead of other states.
10	Good info, agents are easy to talk with
9.5	People that love the outdoors are a blessing to our world.
9	It hear a good reputation. It attempts to be non political.
9	I haven't been to any locations provided by them but the care and maintenance of all the land tells all.
9	Because my family likes being outside.
9	You call they come
9	It is the primary source
9	Have always had great experiences with MDC
9	They are very informative
9	I believe the MO Conservation is important to our community.
9	I use Twin Pines for Educational activities. I also visit it with my grandchildren Conservation Magazine good.
9	Read the magazine & participated in outdoor activities every weekend.
8	Useful info & products
8	I have friends who work for the conservation department.
8	A very good dept to serve the public
8	As an environmental agency, it's top notch and assists with other educational institutes to further understand local wildlife.
8	Just like some of their efforts
8	For the most part all the areas I have been are well kept and well patrolled.
8	I enjoy all the wildlife
8	Don't know
8	I can see myself say, "Go ask them"
8	Dept of Con is good!

7	I think the workers could do more cleanup instead of just standing around.
7	There is a lot to do and a lot to see and learn
7	Educational
7	I have a friend that works for the MDC and he has really told us a lot of MO, trees, plants wildlife & Ponds
7	They are always friendly when talking to someone.
6	Cause it depends on my mood and the other person
6	I feel they do a good work
5	Personal experience
5	Unsure
5	I don't like what they do about the hogs.
5	Not sure I trust MO Conservation
5	Worked for the Conservation force
5	Some parts of the agency are doing great things for wildlife. Other parts are a complete waste of money.
5	Don't know enough about it?
2	I feel as a member of the area that the conservation abuses what power it has. Local residents are not trying to hurt the area.
1	Theres good people who live here.
0	Don't like the stands on hog hunting. Spend money wiser.

Appendix V: Additional comments respondents provided for their thoughts on wildlife and public land.

Additional Comments about MDC lands and wildlife
Keep up the good work. It is needed.
It provides us with abundant unlimited opportunities for recreation, hunting, fishing, and learning about nature and wildlife.
We have too much tourism. They are not informed of the various diseases that ticks can give off. If you advertise MO warn them what we have to live with.
Great director of commissioner do{es} their job.
It's a wonderful place to live.
More is better
I enjoy hunting on conservation land
Ya'll rock for doing this. Gonna have to research some info I wasn't aware of how uneducated I was in my own backyard.
Open hog hunt for everyone.
I'm happy to see they are all well maintained.
MDC is doing a great job wisely using the tax money we designated for them.
We love MO Conservation Areas. PS Amidon is our favorite.
Pick up the trash
Follow the rules! Vacationers come and leave a mess. {We} Need more field officers to enforce.
Missouri{'s} wildlife is important to have around
I enjoy going to see the wildlife at the Peck Ranch area in my county.
We do not want {a} widespread shut down of our natural areas. We realize we live in "Gods Country." We love it here.
It would be nice if they would ask the community before deciding something.
Some of the state's public land is beneficial to wildlife habitat. Other land such as small accesses are just a magnet for jobless drug users and all-around criminals.
Elk
Enjoy Twin Pines. We need to keep some areas open to public use- conservation is good but access to it is good too.
To restrict on some things.
Conservation--keeps areas available for all the people---important