A Survey of Potential Non-Traditional Hibernaculum of Bats within Boone Cliffs State Nature Preserve

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ABSTRACT

Bats are an integral part of the forest ecosystem providing insect control, pollination, seed dispersal, and forest health indication. The following study was conducted in October and November 2023 to determine what species of bats are present in Boone Cliffs State Nature Preserve, an old-growth mixed mesophytic forest, in Boone County, Kentucky at a time when many species migrate to warmer temperatures. Stationary acoustic monitoring of echolocation calls detected a total of seven species: big brown bat (*Eptesicus fuscus*), eastern red bat (*Lasiurus borealis*), evening bat (*Nycticeius humeralis*), hoary bat (*Lasiurus cinereus*), silver-haired bat (*Lasionycteris noctivagans*), tri-colored bat (*Perimyotis subflavus*), and gray (*Myotis grisescens*). Understanding species diversity is utilized for land use decisions and ecosystem conservation.

INTRODUCTION

Over winter, bats hibernate, or enter torpor, in various locations including caves, manmade structures, hollow trees, or leaf litter (Vowels 2018). For example, Indiana bats (*Myotis sodalist*) like to roost in trees for the winter but other bats, like gray bats, prefer to hibernate in caves and sinkholes.

A site where one or more bats hibernate in the winter is called a hibernaculum (WHF Field Guide 2023). The purpose of hibernacula is to provide a more consistent temperature and protection through the winter. Bats slow their breathing rate, metabolic rate, and heart rate and decrease body temperature in closer alignment to the air temperature. Most wake from torpor

frequently through the winter (WHF Field Guide 2023). The traditional hibernacula for many species are caves and mines.

Currently sixteen species of bats have a documented presence in Kentucky (Kentucky Fish and Wildlife Resources 2023). Boone Cliffs State Nature Preserve is 29.9 hectares of mainly maple-oak forest with cliffs formed from glacial deposits. There was some logging prior to the property's purchase in 1975 by The Nature Conservancy. Based on historical research by Bryant et. al, the Sugar Maple (*Acer saccharum)* was noted as the most abundant species (2019 data Boone Cliffs, Table 5, 6, 7, Appendix). The water source for this location is a tributary to Middle Creek, and the Cliffs are notable in Boone County for its Quaternary glacial conglomerates. Elevations within the preserve range from 198 to 259 m above sea level (Bryant 1978). Soils include silt loam and clay loam with conglomerates of sand, gravel, limestone, and crystalline rocks (Bryant 1978).

There are two main types of rock in Northern Kentucky: Ordovician formations and Quaternary glacial deposits. Ordovician formations contain fossils like trilobites and brachiopods. Formations can be distinguished based on their composition and fossil makeup. There are four major formations of limestone and shale in the area: the oldest, Kope formation (high shale content); Fairview Formation (more limestone with thinner beds and more lateral flow); Grant Lick (lays over Fairview and is prone to sinkholes, shells, and bryozoans); and the youngest, Bull Fork (rolling hills/moderate slopes). These limestone formations hold the potential to form sinkholes due to their water permeability. The second type of rock is Quaternary glacial deposits. The formation present at the Cliffs is pre-Illinoian glacial gravel remains, and the outwash is well exposed at the Cliffs. The glacial deposits experience wind and

water erosion, causing them to form the steep cliffs at the State Preserve, and cycles of freezing and thawing cause cracks and cavities for further water intrusion.

The purpose of this study was to survey the potential hibernacula of bat species within Boone Cliffs State Preserve. Habitat characteristics selected by cave dwelling bat species - including glacial deposits, limestone, and rock caves - are present at this location. This is part of ongoing research into the overall forest health in the county and nearby areas through a better understanding of species diversity throughout the year.

Figure 1. Kentucky County Map, identification star at Boone Cliffs State Nature Preserve. https://gisgeography.com/kentucky-county-map/
accessed December 9, 2023

Figure 2. Photo by Paul Ankenbauer. Unidentified live bat found in Boone County on November 1st, 2023.

METHODS

First, researchers used the geologic map of the area (W. C. Swadley 1971) to predict the location of sinkholes. Sinkholes form when a permeable soil or rock layer is overlain by a more solid formation – as the permeable layer erodes due to subsurface water movement, the overlying material can no longer support the stresses it is subjected to and falls away into the empty space. It was determined that due to the permeability of the local limestone formations, the Grant Lick formation was the most likely to cause sinkhole formation.

In Figure 3, the legend shows that brown represents the Kope formation. Researchers climbed out of the Kope, into the Fairview formation, and then checked the Grant Lick. This formation is only present on the surface in one small area near the origin of the tributary creek. Researchers walked the area in a line transect and looked for visible signs of “seepy” areas through depressions in the ground, cracks in the soil, or unusual vegetation patterns.

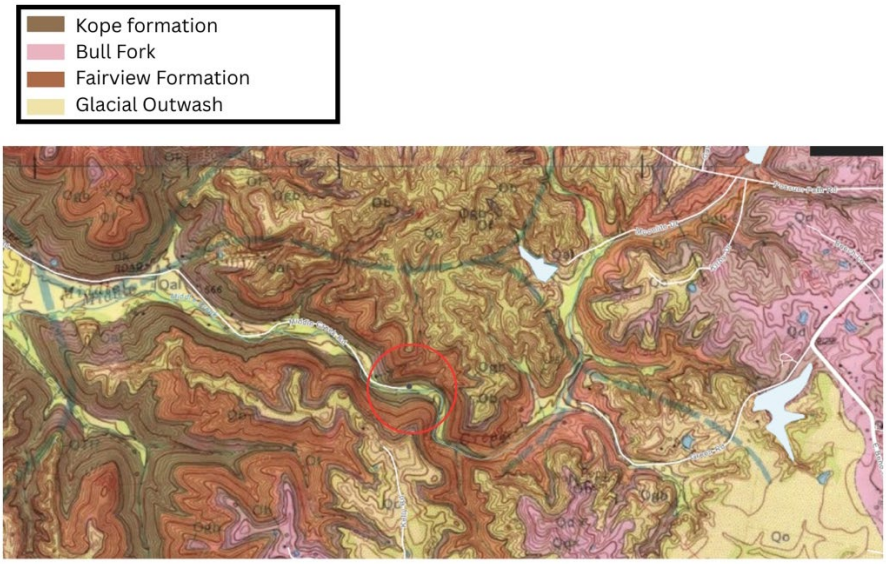

Figure 3. Topographical map of Boone County Kentucky, Geologic Quadrangle Map GQ-929, U.S. Geological Survey https://ngmdb.usgs.gov/Prodesc/proddesc_2229.htm accessed December 9, 2023 and adapted by Lillie Daniel.

A grid pattern (6m x 6m) was formulated and implemented with researchers in each square searching for sinkholes to effectively cover the entire highlighted section of property. Sinkholes would be marked, and areas monitored with an acoustic detector (Figure 15, Appendix) over the months of October and November. The Anabat Swift is a full spectrum passive detector from Titley Scientific that records echolocation calls in .wav files.

Four researchers went to Boone Cliffs State Nature Preserve on August 29th, 2023, to scout out the determined area. No sinkholes were marked. One moderately sized rock shelter was identified in a conglomerate cliff, and the acoustic detector was placed nearby (38.99538° N, 84.78230° W). One researcher surveyed inside the small cave, and although no bats were observed, there were many crevices and holes that stretch far back providing potential for a valuable roost site.

Materials included an Anabat Swift Acoustic Monitoring Stationary Device by Titley Scientific, Omni-directional Ultrasonic Microphone US-O V3, Master Python Cable Locks, headlamp, GoPro, knee pads, and gloves. Researchers used a Geologic map of part of the Rising Sun quadrangle, Boone County, Kentucky Geologic Quadrangle 929 by: W. C. Swadley (1971) and Kaleidoscope Pro Analysis Software by Wildlife Acoustics (version 5.6.4).

Figure 4. Photos by Lillie Daniel. Cave and rock formations Boone Cliffs State Nature Preserve.

Figure 5. Photo by Lillie Daniel. Anabat Swift placement Boone Cliffs State Nature Preserve.

RESULTS

Acoustic devices generate .wav sound files containing echolocation pulses. All data collected were analyzed through the Kaleidoscope software (version 5.4.1), an acoustic analysis program, to determine the species. Each bat species is assigned a six-character code in the software (Table 8, Appendix) and software is set to “Bat Mode” with default signal parameters and Auto ID set to North America and the state of Kentucky. Kaleidoscope provided an onscreen I.D. (Figure 16, Appendix) and spreadsheet of species based on the detection of echolocation calls that are consistent with the structure of the species within an acceptable confidence level (pulse match ratio). The software additionally provides on screen spectrograms of the echolocation calls (Figure 17, Appendix).

Table 1 details the average high, low, and mean temperatures for the months of October and November (the study period). Daily temperatures are shown in Figures 6 and 7. Many days in both months fall above the historical averages for this time period.

Table 1. Average Boone County Temperatures in October and November 2023 as reported by datetime.com.

October Climate & Weather Averages in Boone County (°C)	
High Temperature	19.4
Low Temperature	6.1
Mean Temperature	12.8

November Climate & Weather Averages in Boone County (°C)	
High Temperature	12.8
Low Temperature	1.1
Mean Temperature	7.2

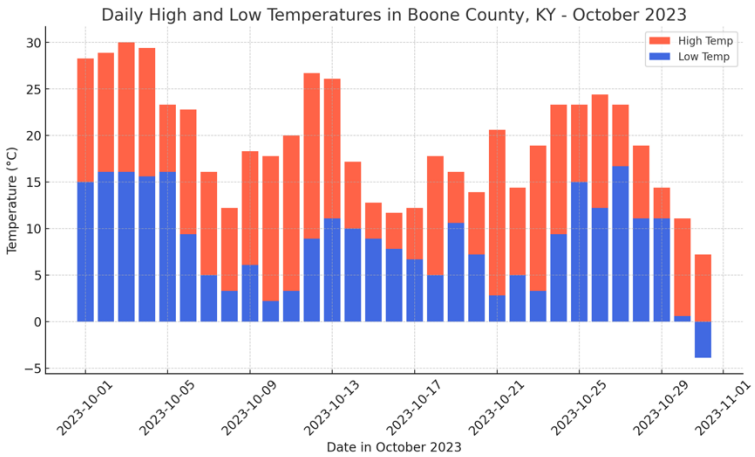

Figure 6. Daily Temperatures in Boone County Kentucky from October 2023. (https://www.accuweather.com/en/us/boone/40403/october-weather/2177592?year=2023)

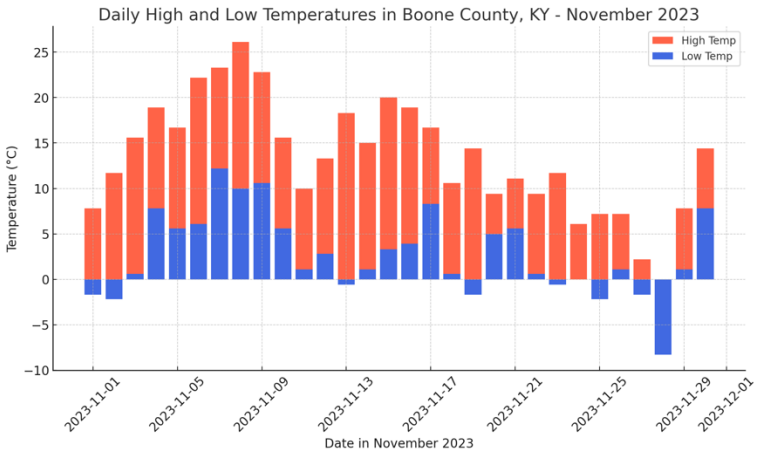

Figure 7. Daily Temperatures in Boone County Kentucky from November 2023. (https://www.accuweather.com/en/us/boone/40403/october-weather/2177592?year=2023)

Table 2 presents the eleven species detected at Boone Cliffs State Nature Preserve in June and July of 2023.

Table 2. Bat species identified with passive acoustic monitoring at Boone Cliffs State Nature Preserve during the months of June and July 2023.

Bat Species
Big Brown *(Eptesicus fuscus)*
Eastern Red *(Lasiurus borealis)*
Evening *(Nycticeius humeralis)*
Hoary *(Lasiurus cinereus)*
Indiana (*Myotis sodalis*) *
Little Brown (*Myotis lucifugus*)
Northern Long-Eared *(Myotis septentrionalis)*
Silver-Haired *(Lasionycteris noctivagans)*
Small-Footed *(Myotis leibii)*
Southeastern (Myotis austroriparius)
Tricolored *(Perimyotis subflavus)*

*Currently listed as endangered (Kentucky State and Federal)

Table 3 presents the seven species detected at Boone Cliffs in October and November of 2023.

Table 3. Bat species identified with passive acoustic monitoring at Boone Cliffs Nature Preserve during the months of October and November 2023.

Code	Bat Species	Number of Files	Date Occurrences
EPTFUS	Big Brown *(Eptesicus fuscus)*	46	10
LASBOR	Eastern Red *(Lasiurus borealis)*	5	4
LASCIN	Hoary *(Lasiurus cinereus)*	93	27
LASNOC	Silver-Haired *(Lasionycteris noctivagans)*	158	26
MYOGRI	Gray (*Myotis grisescens*) *	4	3
NYCHUM	Evening *(Nycticeius humeralis)*	42	13
PERSUB	Tricolored *(Perimyotis subflavus)*	12	7

*Currently listed as endangered (Federal)

Table 4 presents a standard species richness, a total count of the bat species observed in the biological community of Boone Cliffs State Nature Preserve.

Table 4. Species richness at Boone Cliffs during June-July and October-November passive acoustic monitoring.

Species Richness	
June-July	11
October-November	7

Figures 8 – 14 show the pulse activity per date for each of the observed species.

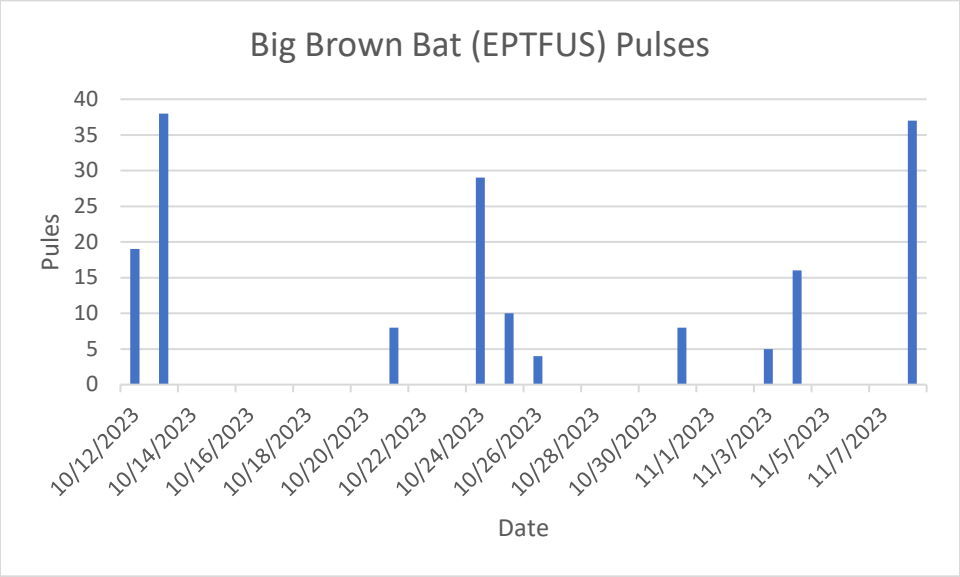

Figure 8. Big brown bat (*Eptesicus fuscus*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

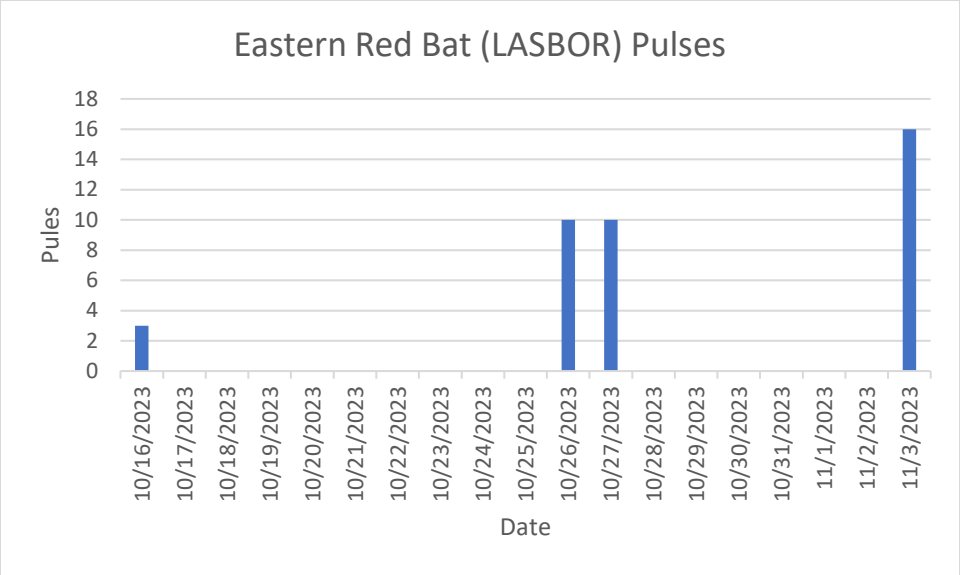

Figure 9. Eastern red bat (*Lasiurus borealis*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

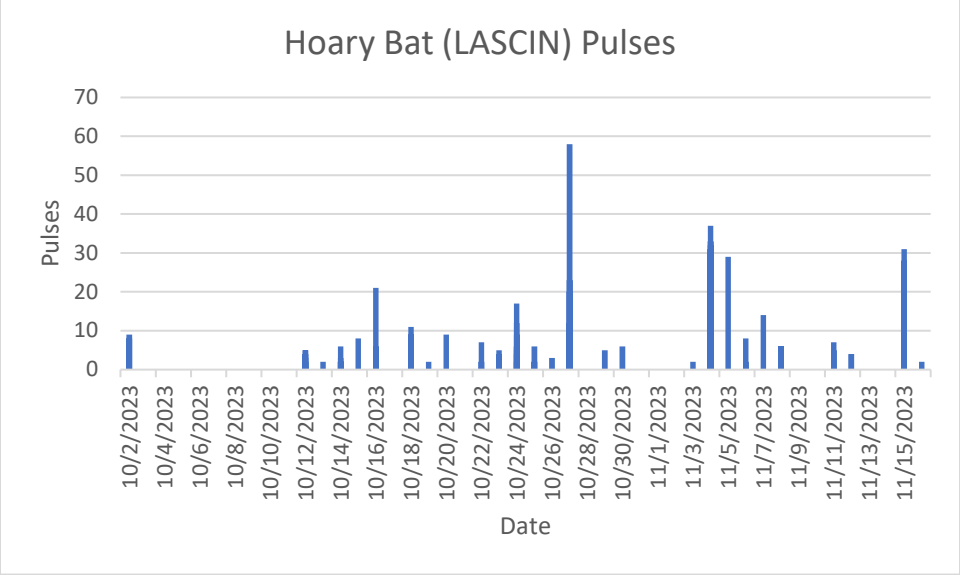

Figure 10. Hoary bat (*Lasiurus cinereus*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

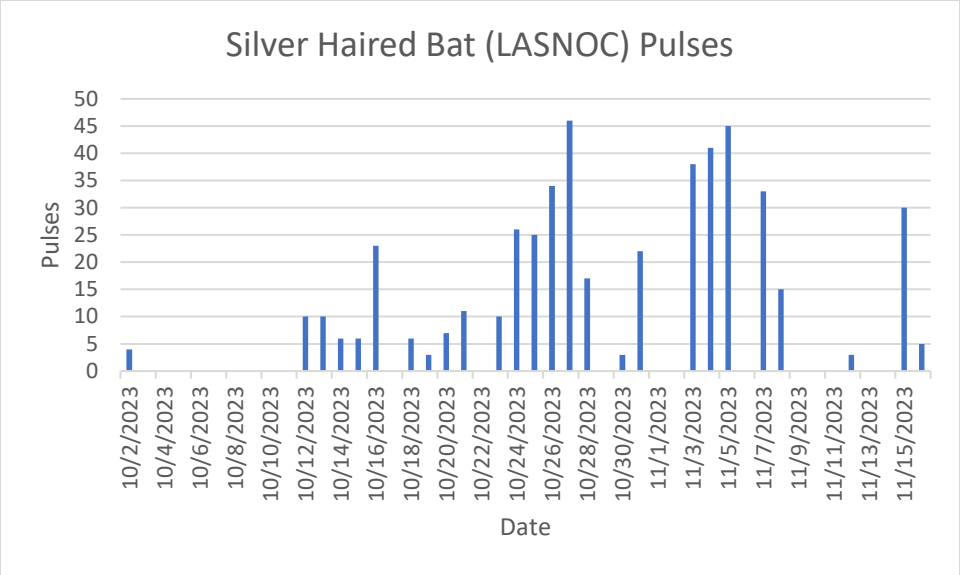

Figure 11. Silver-haired bat (*Lasionycteris noctivagans*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

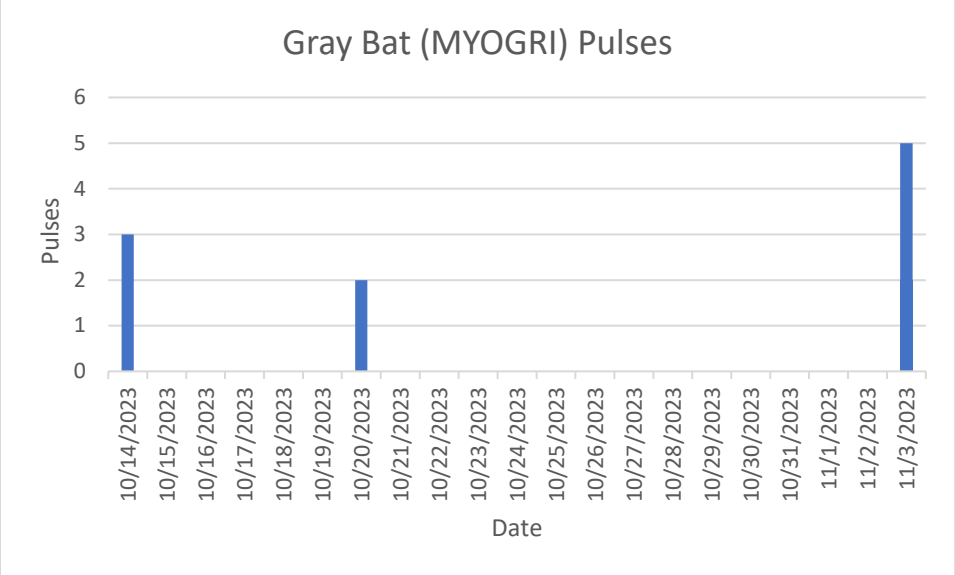

Figure 12. Gray bat (*Myotis grisescens*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

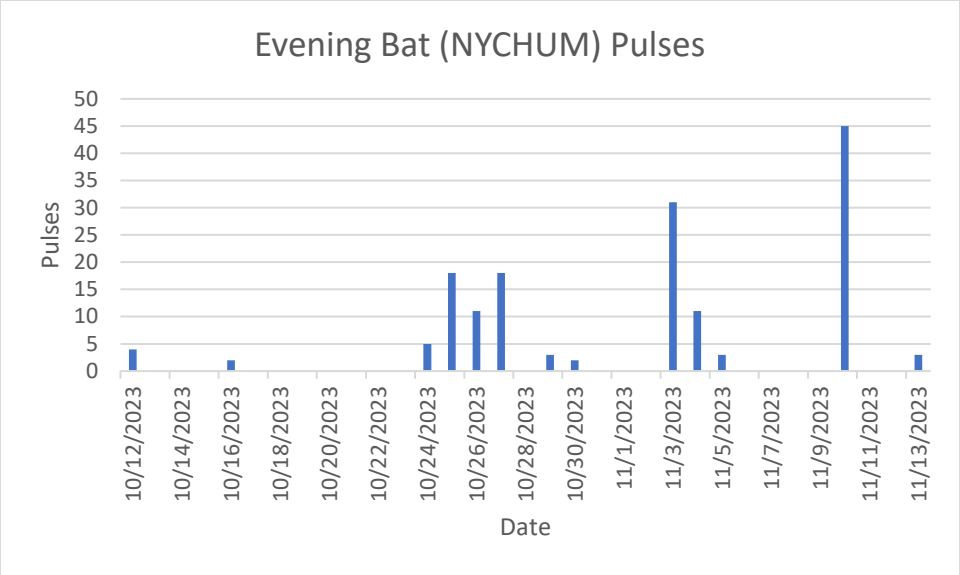

Figure 13. Evening bat (*Nycticeius humeralis*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

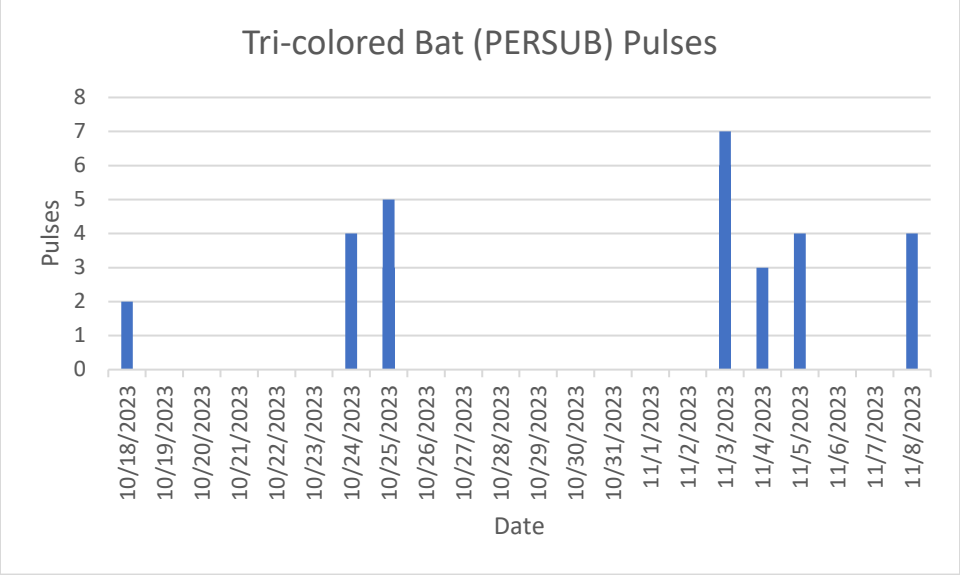

Figure 14. Tri-colored bat (*Perimyotis subflavus*) pulse activity collected by stationary acoustic monitoring for October and November 2023, Boone Cliffs State Nature Preserve.

The overall species richness decreased from the June-July 2023 observation period to the October-November 2023 period. The silver-haired bat registered 158 call files, the highest

call volume observed. This bat species saw a mid-October/early November peak. The hoary bat registered 27 date occurrences (10/27/2023 highest call volume), the highest frequency of date occurrences. An endangered gray bat was registered (no distinct pattern). Additional observations of highest call volume: big brown (10/13/2023 and 11/8/2023), eastern red (11/3/2023), evening (11/10/2023), and tri-colored (11/3/2023).

DISCUSSION

During the months of October and November 2023, stationary acoustic monitoring was conducted to identify the bat species present in Boone Cliffs State Nature Preserve. Boone Cliffs provides habitat characteristics chosen by cave dwelling bat species including glacial deposits, karst formations, and rock caves. Data was analyzed through the Kaleidoscope software identifying a total of seven species in the surveyed area: big brown bat (*Eptesicus fuscus*), eastern red bat (*Lasiurus borealis*), evening bat (*Nycticeius humeralis*), hoary bat (*Lasiurus cinereus*), silver-haired bat (*Lasionycteris noctivagans*), tri-colored bat (*Perimyotis subflavus*), and gray bat (*Myotis grisescens*). Overall behavior patterns for most of the species indicate several days with limited to no activity followed by a day of activity. The silver-haired bat and hoary bat were the most activity with less gap between echolocation call observation. These activity pattern observations are supported by prior knowledge of bat hibernation - torpor with frequent waking through the season.

Traditional winter habitat for the big brown bat includes caves, and the tri-colored bat also prefers caves, mines, and rock shelters. The eastern red bat selects tree hollows or leaf litter. The silver-haired bat has the potential to migrate seasonally but can choose to remain in

the area selecting small tree hollows, loose tree bark, wood piles, cliff face crevices, cave entrances, and buildings (Kentucky Department of Fish and Wildlife Resources 2023). During October and November 2023, the silver-haired bat registered 158 call files, the highest call volume observed, indicating this species tended to remain in the area early Fall. With the early November peak, additional data collection in the winter months is required to determine if migration may have occurred later in the season. The hoary bat and evening bat also typically migrate to a warmer area (Batcon.org 2020), but both still showed a presence near the end of the data collection period indicating the species had not yet migrated. The endangered gray bat typically looks for limestone karst sites to roost. The species was registered with no distinct pattern in date occurrence, so further study is recommended as this species was not identified during the June/July 2023 survey of the area. Four species that were present in June-July 2023 were not detected in the October-November 2023 survey. The Indiana bat (*Myotis sodalis*), which prefers a limestone cave with a consistent temperature, likely migrated to a more hospitable area. The little brown bat (*Myotis lucifugus*) also prefers warmer caves, likely migrating as well. The northern long-eared bat (*Myotis septentrionalis*) and small footed bat (*Myotis leibii*) will hibernate in caves but do sometimes use alternate roosts like rock shelters and fissures (Kentucky Department of Fish and Wildlife Resources 2023). In a study of similar purpose, Joseph L. Pettit and Joy M. O'Keefe of Indiana State University conducted an extensive long term 17-year study on the timing of migration of the indiana bat (*Myotis sodalis*). The basis of the study was to better understand the cues used for migration to guide area management strategies including reducing disturbance during peak migration periods. Cues can include changes in prey availability, temperature, precipitation, wind speed, and lunar illumination.

Modeling data for the Pettit and O'Keefe study included: temperature, precipitation, average daily wind speed, mean sea level pressure, daily lunar illumination, and daily Pacific North American Pattern index (associated with temperature and wind patterns). Results indicated that beyond day of the year, temperature, precipitation, and wind were the next most important cues. Understanding bat migration ecology can assistant in development effective area management strategies to reduce impact in these periods (Pettit and O'Keefe 2017).

Temperature patterns recorded during the October – November 2023 study were above the historic average. This is a general trend and may represent cause for concern of the climate change impact. Increased temperatures can disrupt migration patterns and cause bats to wake from torpor earlier or more frequently. This can result in bats using stored energy that is required for them to survive the winter. Based on the trend in changing weather patterns, a longer study period is required to determine if bats are migrating later or remaining in the area.

Future research should include monitoring temperature, precipitation, and wind patterns throughout the winter months and into spring to fully understand roosting and migration patterns in the area

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https://www2.gov.bc.ca/assets/gov/environment/natural-resource-policy-legislation/legislation-regulation/frpa-pac/wildlife-habitat-features/whf_field_guide_kootenay_boundary_bat_hibernaculum.pdf

APPENDIX

App. Table 5. Diversity Indices: Richness, Shannon-Weiner Index, and Evenness for sites at Boone Cliffs calculated with data provided at Thomas More University, Department of Biology, BIO 318 Ecology 2023.

Site	Year	Richness # of Species (s)	Shannon Index (H')	Evenness (J)
Boone Cliffs				
	1972	18	2.23	0.77
	1997	15	2.06	0.76
	2019*	14	1.86	0.70

App. Table 6. Basal area (m^2/ha) and Density (N/ha) for the tree species sampled at Boone Cliffs calculated with data provided at Thomas More University, Department of Biology, BIO 318 Ecology 2023.

Site	Year	Basal area (m^2/ha)	Density N/ha
Boone Cliffs			
	1972	Unavailable	645.8
	1997	21.55	283.3
	2019*	28.87	375.0

*Not the entire forest

App. Table 7. The Number (N) and Importance Value (IV) for all tree species at sampled at Boone Cliffs calculated with data provided at Thomas More University, Department of Biology, BIO 318 Ecology 2023.

Species	Common Name	N 1972	N 1997	N 2019	IV 1972	IV 1997	IV 2019
Acer saccharum	Sugar maple	51	23	27	61.4	69.16	75.07
Quercus muhlenbergii	Chinkapin	23	14	7	43.8	54.17	26.35
Celtis occidentalis	Common hackberry	10	5	32	18.0	26.81	64.81
Quercus rubra	Northern red oak	8	1	3	28.3	4.92	6.97
Quercus alba	White Oak	1	0	0	5.2	0.0	0.0
Carya cordiformis	Bitternut hickory	2	1	2	12.0	10.71	11.71
Juglans nigra	Black walnut	9	0	0	22.3	0.0	0.0
Ulmus rubra	Slippery elm	18	9	6	25.6	39.96	28.01
Aesculus glabra	Ohio Buckeye	1	0	0	2.6	0.0	0.0
Fagus grandifolia	American beech	0	1	1	0.0	6.41	5.79
Acer negundo	Box elder	1	3	4	2.8	12.17	18.81
Asimina triloba	Pawpaw	0	1	0	0.0	4.95	0.0
Carpinus caroliniana	America hornbeam	1	0	0	2.7	0.0	0.0
Carya ovata	Shagbark hictory	0	1	1	0.0	5.39	5.06
Cercis canadensis	Eastern redbud	7	0	0	9.6	0.0	0.0
Fraxinus americana	American white ash	8	3	1	21.6	13.52	5.29
Fraxinus quadrangulata	Blue ash	1	3	3	2.7	18.46	18.00
Gymnocladus dioicus	Kentucky coffeetree	0	1	1	0.0	11.35	11.06
Liriodendron tulipfera	Tuliptree	6	1	1	16.3	6.22	6.65
Platanus occidentalis	Sycamore	1	1	1	5.1	15.80	16.43
Robinia pseudo-acacia	Black locust	2	0	0	6.2	0.0	0.0
Ulmus americana	American elm	5	0	0	13.7	0.0	0.0
	Totals	155.0	68.0	90.0	300.0	300.0	300.0

App. Table 8. Kaleidoscope Codes for the Bat Species in Kentucky

Common/Scientific Name	Kaleidoscope Code
Big Brown Bat *(Eptesicus fuscus)*	EPTFUS
Eastern Red Bat *(Lasiurus borealis)*	LASBOR
Evening Bat *(Nycticeius humeralis)*	NYCHUM
Gray Bat *(Myotis grisescens)**	MYOGRI
Hoary Bat *(Lasiurus cinereus)*	LASCIN
Indiana Bat *(Myotis sodalis)**	MYOSOD
Little Brown Bat *(Myotis lucifugus)*	MYOLUC
Northern Long-eared Bat *(Myotis septentrionalis)*	MYOSEP
Rafinesque's Big-eared Bat *(Corynorhinus rafinesquii)*	CORRAF
Southeastern Bat *(Myotis austroriparius)*	MYOAUS
Silver-haired Bat *(Lasionycteris noctivagans)*	LASNOC
Small-footed Bat *(Myotis leibii)*	MYOLEI
Tricolored Bat *(Perimyotis subflavus)*	PERSUB
Virginia Big-eared Bat *(Corynorhinus townsendii)**	CORTOW

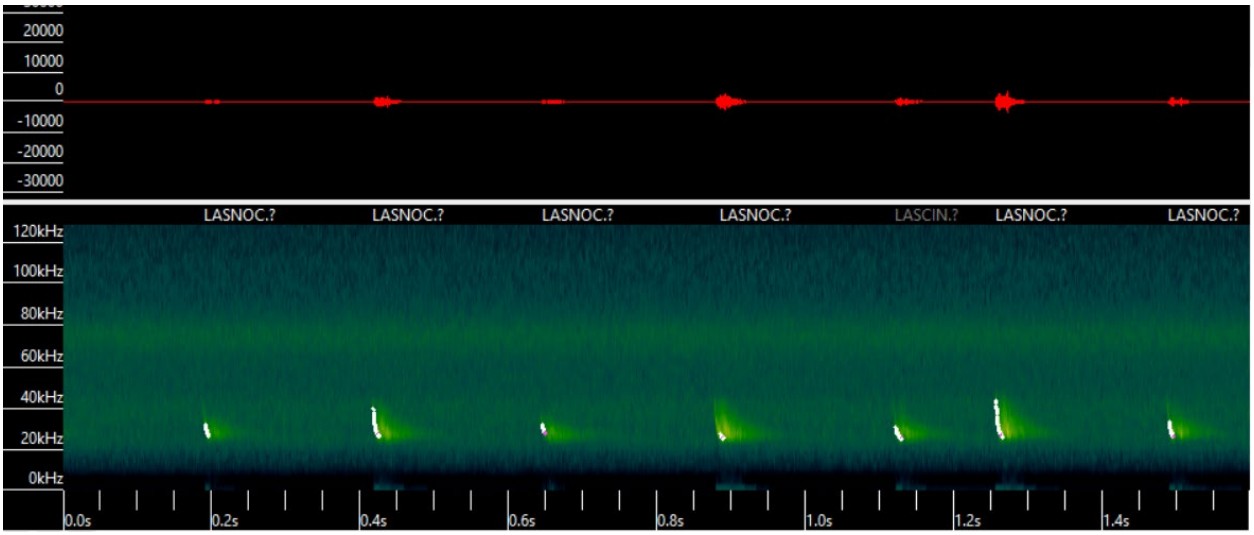
*currently listed as endangered

App. Figure 15. Anabat Swift Acoustic Monitoring Stationary Device by Titley Scientific, Omni-directional Ultrasonic Microphone US-O V3, Master Python Cable Lock (September 2023, Conservancy Park Belleview)

File Help

	FOLDER	IN FILE	OUT FILE FS	OUT FILE ZC	AUTO ID	PULSES	MATCHING	MATCH RATIO
6		546412 2023-10-24 15-29-55.wav			LASCIN	6	4	0.667000
7		546412 2023-10-24 20-18-33.wav			LASCIN	6	3	0.500000
8		546412 2023-10-24 15-29-35.wav			LASCIN	9	3	0.333000
9		546412 2023-10-24 14-50-25.wav			LASCIN	2	2	1.000000
10		546412 2023-10-24 20-01-11.wav			LASCIN	4	1	0.250000
11		546412 2023-10-24 19-30-18.wav			LASNOC	26	17	0.654000
12		546412 2023-10-24 14-48-24.wav			LASNOC	16	11	0.688000
13		546412 2023-10-24 21-30-03.wav			LASNOC	15	10	0.667000
14		546412 2023-10-24 15-29-25.wav			LASNOC	25	8	0.320000
15		546412 2023-10-24 14-48-13.wav			LASNOC	13	7	0.538000
16		546412 2023-10-24 15-30-07.wav			LASNOC	16	7	0.438000
17		546412 2023-10-24 20-01-24.wav			LASNOC	7	6	0.857000
18		546412 2023-10-24 20-00-47.wav			LASNOC	5	5	1.000000
19		546412 2023-10-24 14-50-14.wav			LASNOC	6	4	0.667000

App. Figure 16. Sample id. Output file by the analytical software Kaleidoscope.



App. Figure 17. A spectrogram of silver-haired bat call generated by the analytical software Kaleidoscope.