RISON OF ACOUSTIC MONITORING STRATEGIES FOR DETERMINING BAT POPULATIONS THROUGHOUT LAND BETWEEN THE LAKES NATIONAL RECREATION AREA

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Comparison of acoustic monitoring strategies for determining bat populations throughout Land Between the Lakes National Recreation Area

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DEDICATION

I would like to dedicate this thesis to my mother, Vivian McCormick. It is because of her influence and unwavering support that I made it this far. Thank you.

Table 11: Transect 3 for 2009-2011 was compared to historical mist-netting data. Mist-netting captured all species except EPFU, LACI, and LANO. The acoustic transects did not record LANO and MYSE 36
Table 12: Each stationary site was surveyed by using two detectors. Anabat and Avisoft both detected various bat species. Names indicate which recording technique recorded a particular bat species. 37

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ABSTRACT

RILEY SETH MCCORMICK. Comparison of acoustic monitoring strategies for determining bat populations throughout Land Between the Lakes National Recreation Area (under the direction of DR. ANDREW N. BARRASS).

The use of ultrasonic acoustic bat detectors, used to detect echolocation calls, has increased our knowledge on bats and has provided new techniques for species monitoring. The acoustic monitoring of bats in the U.S. Forest Service, Land Between the Lakes began summer 2009-2011. The sampling includes a total of three driven transects and five stationary monitoring sites. Acoustic monitoring of bats was performed during June and July. Each transect was driven for approximately 30 miles at 15-20 miles/hr using an ANABAT© high frequency recording unit attached to the roof of each vehicle. Acoustic monitoring of bats was also performed at stationary sites after transects were recorded using two recording units, Avisoft Ultrasound© gate and ANABAT©. The transects and stationary sites were entered into a GPS project map for further investigation. Field acoustic samples were analyzed and identified in the lab using proprietary software. The total number of calls, or sounds, collected for all three transects were 4350 and for all five stationary sites equaled 622. A total of nine species were recorded and identified throughout Land Between the Lakes during the three years of study. The number of bat calls and species identified for each year varied among each stationary and transect site.

Table of Contents

Chapter I: Introduction	1
Ultrasonic acoustic bat detectors	6
Identifying bats by their call	9
Chapter II: Materials and Methods	12
Study Site	12
Species of bats to be considered for study	12
Collection Methods	13
Driven transects	14
Stationary Sites	15
Geographic Information System mapping	17
Analysis of acoustic data	19
Chapter III: Results	20
Chapter IV: Discussion	38
Transects and Stationary Sites species occurrence	38
Transects versus Mist-netting	39
Anabat versus Avisoft	40
Bat calls recorded and Unidentifiable files	40
Transects and Stationary Sites	41
White-nose syndrome and management	43
Literature Cited	45
Appendices	51
Vitao	56

List of Tables

Table 1: In 2009 a total of 1,315 files were recorded for all three transects. The totals for species calls and other files are represented in this table for each sample period and for each transect. The LABO, PESU, unidentified and noise were the most abundant files recorded during 2009
Table 2: In 2009 a total of 158 files were recorded for all five stationary sites. The totals for species and other files are represented in this table for each sample period and for each individual stationary site. The LABO, PESU, and unidentified were the most abundant files recorded during 2009. 24
Table 3: In 2010 a total of 680 files were recorded for all three transects. The totals for species and other files are represented in this table for each sample period and for each transect. The LABO, PESU, unidentified and noise were the most abundant files recorded during 2010.
Table 4: In 2010 a total of 149 files were recorded for all five stationary sites. The totals for species and other files are represented in this table for each sample period and for each individual stationary site. The PESU and unidentified were the most abundant files recorded during 2010.
Table 5: In 2011 a total of 2,355 files were recorded for all three transects. The totals for species and other files are represented in this table for each sample period and for each transect. The LABO, PESU, NYHU, unidentified and noise were the most abundant files recorded during 2011
Table 6: In 2011 a total of 355 files were recorded for all three transects. The totals for species and other files are represented in this table for each sample period and for each transect. The LABO, PESU, unidentified and noise were the most abundant files recorded during 2011
Table 7: The total number of files recorded for the entire study was accessed for usability for both transects and stationary sites. Unidentifiable was more that 50% for both34
Table 8: A comparison of total years of species specific calls for transects and stationary sites 35
Table 9: Transect 1 for 2009-2011 was compared to historical mist-netting data. Mistnetting captured one more bat species that the acoustical transects, the MYSE35
Table 10: Transect 2 for 2009-2011 was compared to historical mist-netting data. Mist-netting captured all species except EPFU. The acoustic transects did not record LACI, LANO and MYSE

List of Figures

Figure 1: The Land Between the Lakes study area, counties represented by yellow. The counties represented by dark blue are confirmed cases of WNS and the counties represented by light blue with hashes are suspected of having the WNS fungus infected bats. 3
Figure 2: A map of LBL (shaded in dark green). The map shows the two rivers, Tennessee and Cumberland, which encompasses LBL and the three counties it resides within, Stewart county TN, Trigg and Lyon, counties KY
Figure 3: Photograph of the homemade protective box with the Anabat Microphone protruding from the surface
Figure 4: A GIS map of LBL including driven transects and stationary acoustic sampling sites. The different colors (road numbers) for each transect represents the road traveled or route for a specific transect. The green dots represent each stationary sampling site16
Figure 5: The data collected for each Anabat file recorded was compiled for each transect to illustrate for the entire study what files were most abundantly recorded32
Figure 6: The data collected for each Anabat and Avisoft file recorded was compiled for each stationary site to illustrate for the entire study what files were most abundantly recorded

CHAPTER 1

INTRODUCTION

Successful conservation and preservation of bat populations depends on the different techniques used to monitor bat populations (Lewis, 1995). It is important to determine what techniques are most appropriate for surveying bat communities, especially since a newly discovered fungal disease, called the White-nose Syndrome (WNS), is killing millions (>5.5) of bats in the Eastern United States (U.S. Fish and Wildlife, 2012).

The WNS is linked to a newly described fungus, Geomyces destructans (Gargas et al., 2009). The scientific community has little information on this fungus, but the fungus is associated with the death of cave dwelling bats during their winter hibernation. The disease is termed WNS because the white fungus appears around the muzzle of the bat and on its soft tissue membranes (Blehert et al., 2008). The disease was first recognized in 2006 at Howe's Cave, New York, and has since spread throughout the Eastern United States (Blehert et al., 2008; USGS National Wildlife Health Center, 2010). In response to the increasing encroachment of WNS, the U.S. Fish and Wildlife (USFWS) prompted the state of Tennessee (TN) to create the White-nose Syndrome Cooperative Monitoring and Response Plan for TN in 2009 to brace for WNS (Arnold Air Force Base, 2009). The USFWS decided to monitor bat populations by developing acoustic transects throughout TN during each summer (Arnold Air Force Base, 2009). These transects are driven once in June, using acoustic monitoring devices, at a time

before females give birth to their young, and once in July when pups have left roost sites for the first time (Arnold Air Force Base, 2010). The purpose of the acoustic monitoring transects is to acquire data on densities of various bat populations each summer during the bats normal feeding activities or flight patterns within varying habitat conditions (Arnold Air Force Base, 2009; Russ et al., 2003). The data collected are used to compare survey years and assess any patterns of potential decline. If density numbers for bat populations decline from year to year, then other means of monitoring will most likely be implemented to determine the cause for the decline. This data also will give researchers a history of what the bat densities were in the past if WNS kills an entire population.

Simply the presence of WNS in TN (Figure 1) demands that questions regarding population status for various bats be answered, and the White-nose Syndrome

Cooperative Monitoring and Response Plan for acoustic monitoring creates opportunities for research on bat populations.

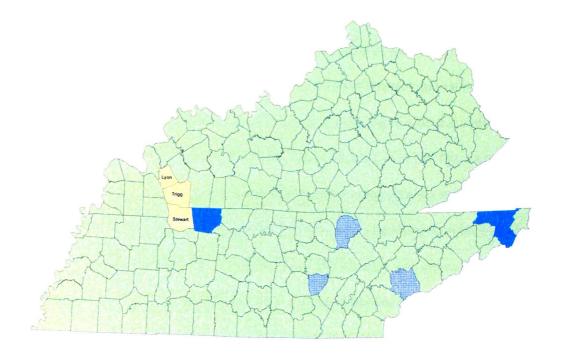


Figure 1. The Land Between the Lakes study area, counties represented by yellow. The counties represented by dark blue are confirmed cases of WNS and the counties represented by light blue with hashes are suspected of having the WNS fungus infected bats.

Austin Peay State University's (APSU) involvement in the pre-exposure TN State Plan has led to many opportunities for acoustic monitoring of bats, bat echolocation studies, as with this study to develop acoustic transect dataset and stationary site data for Land Between the Lakes (LBL). Acoustic monitoring has been a common practice for bat biologists for many years (Lance et al., 1999). Incorporating Anabat recorders (Titley Electronics, Queensland, Australia; www.titley.com.au), modular microphone units, which record ultrasonic bat frequencies, into population density surveys has greatly increased knowledge about bats. The drawback of this system however, is frequent improper identification of bat vocalizations or "calls" as these vocalizations are collected.

It is common that species are misidentified during acoustic monitoring (Lance et al., 1999; Parsons et al., 2000). Results of this misidentification of species vocalizations within an area and a lack of species richness data, confounds vital information when surveying for endangered species. The most accurate sampling of a bat population is hand collection or mist-netting (Thomas and West, 1989). Because of WNS, this accurate but intrusive way of collecting data has recently been under heavy scrutiny in the states of Kentucky and Tennessee. This is due to the fear of cross-contamination of individuals during handling or entanglement in netting, especially between tree and cave species. At this time studies report that only bats that use caves to hibernate have shown signs of WNS. Decontamination protocols must be followed at all times to ensure that spores of WNS that do come in contact with equipment are removed. The decontamination protocols suggest that all equipment be cleaned before transporting to another site (USFWS, 2011). These decontamination protocols ensure that humans minimize the spread of WNS when surveying bats at different locations. Also, the required decontamination of mist-nets and survey equipment has made surveying bat population more expensive and time consuming.

Debate among scientists evaluating the two methods of sampling for bats, acoustic or hand sampling, frequently arises in the literature. Some studies have shown that Anabat acoustic detectors have collected and identified more species diversity within a given area (O'Farrell and Gannon, 1999). Other studies maintain that mist-net surveys captured more species (Kuenzi and Morrison, 1998). Barclay (1999) however, suggested that O'Farrell et al. (1999) lacked proper understanding on how to approach the identification of bats by their acoustic signature. Therefore, whether or not acoustic

monitoring can sufficiently replace hand sampling or mist-netting should be studied. It would be beneficial to determine if acoustic transects can produce accurate data concerning bat species identification and population densities over large areas. In order to accomplish this, historical stationary monitoring data, which include both mist-netting and acoustic sampling, of LBL were used to compare data collected during this study. This historical data is a reliable representation of the true composition of the bat populations at LBL, and can therefore be used to determine if the data collected from the acoustic transects can accurately represent the bat population in a large area as well, but without having to conduct time-consuming surveys.

Therefore, the primary goal of this study was to determine if transects are a useful means to survey bat populations throughout a large area; comparing consecutive surveys completed each season. The study also investigated whether acoustic monitoring transects can be used to accurately discern bat species, bat populations, or if active hand sampling monitoring techniques still need to be done when assessing bat species density and various population richness studies.

In order to capture and assess individual bats and assess their populations, biologists have developed different techniques and strategies to help collect individuals. Mist-netting has been popular with ornithologists and bat biologists to catch birds or bats. A mist-net is a fine mesh net that is placed in heavily used flyways to capture bats while they forage or travel (Hourigan et al., 2008; Murray et al., 1999). This method is intrusive and can potentially injure the bat without employing proper removal techniques (Hoffman et. al, 2010). Also, it takes many nights to capture a significant amount of bats and requires many volunteers to help run the nets (Murray et al., 1999). The benefits of

mist-netting are that the bat can be banded with an identification band for recapture studies, the status of the individual health can be checked (for WNS using Reichard Scarring Index (Reichard, 2009; Reichard and Kunz, 2009), tears in wing, female lactating, etc.), and to determine a sex ratio of the population in an area. Another popular but less intrusive way to collect bats is harp-trapping. A harp-trap is designed to capture bats by using fishing line strung vertically with a bag below. The bats cannot detect the fishing line of the trap and fall into the bag set below the trap. This system is less intrusive than the mist-net because the individuals are not tangled in a net and are safe inside the bag until researchers retrieve them (Duffy et al., 2000). Both of these methods can be used when the appropriate setting is needed. The other technique is to record bats by using ultrasonic bat detectors.

Ultrasonic acoustic bat detectors

Over the last five decades the use of echolocation to study bats has evolved to become an important learning tool for biologists. The use of ultrasonic acoustic bat detectors, used to detect echolocation calls, has increased our knowledge on bats and has provided new techniques for species monitoring. The concept of using bat vocalizations to study bats originally was used to understand how they navigate and catch prey (Grinnel and Griffin, 1958; Galambos and Griffin 1942). Through the development of more advanced acoustic detectors, biologists have started to use this technology to identify bats species by their vocalizations (Ahlen and Baagøe, 1999; Brizke, 2003; Fenton and Bell, 1981; O'Farrell and Miller, 1999; O'Farrell et al., 1999).

Advancements in digital technology have made it more practical to use acoustic detectors in the field. In the past ultrasonic recorders used metallic tape to store data. Studies have shown that recording on metallic tape decreased the quality of calls recorded when the data was placed onto the computer for further analysis (White and Gehrt, 2001). Now all acoustic detectors use some form of compact memory to digitally record all the data, which allows users to store more information and leave the detector in the field longer. The classes of ultrasonic bat detectors depend on how the bat call is recorded and analyzed. The class of ultrasonic bat detector used in this study uses zerocrossing analysis and is called Anabat (Titley Electronics, Queensland, Australia; www.titley.com.au). The Anabat system records ultrasonic frequencies produced by bats that normally the human ear cannot hear (> 20,000 kHz). The data recorded is then analyzed with a program called AnaLookW v. 4.8q (Titley Electronics, Queensland, Australia; www.titley.com.au) which allows the user to see each echolocation vocalization. Anabat is considered a passive bat detector, which is smaller than active acoustic detectors, and can be left in the field over many nights without the user actually being present or the use of a computer attached to the device. The detector records echolocations as bats fly over the microphone. Since the Anabat system is passive it allows the user to program the detector to desired settings before placing it on trees, mounted on poles, or on the roof of the vehicles. The Anabat like other detectors on the market has drawbacks. The detector was developed to record only certain parts of bat calls without recording other important portions or attributes of the call. The drawbacks of the zero-crossing system are that it does not record the harmonics of the call and only records the loudest intensity of the call at the time it was recorded (Corben, 2002). This

means that if insects are the loudest call component during a segment of recording in the area per night, it is more likely some bats will not be recorded. The device as mentioned above does not record all the call characteristics such as structure, harmonics, or amplitude which could lead to misidentification of recorded data.

Avisoft bioacoustic UltraSoundGate 116 (Avisoft Bioacoustics, Berlin, Germany; www.avisoft.com) is another type of detector used to record ultrasonic frequencies produced by bats. While both the Anabat and Avisoft detectors are designed to perform similar functions, there are some differences between them. For example, the Avisoft detector needs to be connected to a computer to record and store the files, which can be cumbersome when in the field. Both types of detectors record the frequency and time of bat calls, but the Avisoft detector also records the amplitude. This means that Avisoft detectors will record all ultrasonic sounds present within the parameter set, while the Anabat detector will only record the loudest bat at the time of detection. The parameters can be set within a program called RECORDER v. 4.2.9 (Avisoft Bioacoustics, Berlin, Germany; www.avisoft.com), which allows the Avisoft detector to record at those particular settings or parameters (frequencies, amplitude, and time). Most importantly the Avisoft detector produces .wav files that can be used to playback each individual sound that was produced by the bat. The downside to this is that more memory and more power are required. Also, the Avisoft detector records call files sequences to a computer, so there will be a slight delay between each file while it writes to the hard drive of the computer. Avisoft detector like the Anabat detector has software used to analyze each file recorded called Avisoft-SASLab Pro v. 5.2.04 (Avisoft Bioacoustics, Berlin, Germany; www.avisoft.com). This software is used to playback .wav files and displays the bat call

sequences collected by the Avisoft detector. The Avisoft detector records all parts of the call structure including the harmonics, which provides better resolution that can be used in identification.

Identifying bats by their call or vocalization structure

Like human dialects and languages, bats have a unique social structure to their species specific echolocation vocalizations. Each bat species can be identified by their call sequence or call pattern. There are many types of calls that bats make that include social (audible to human hearing), foraging, which includes buzz calls, search calls or scan calls, and then there are maneuvering calls. Each of these calls types are placed in two main types of categories: broadband frequency modulated (FM) or narrowband constant frequency (CF). Frequency modulated calls can be used for precise prey localization during feeding but the range at which the FM calls can travel is short unlike CF calls that travel far but cannot localize prey, just detect the prey (Schnitzler and Kalko, 2001; Schnitzler et al., 2003). These two types of calls are unique for different species of bats and can be used in species identification. Frequency modulated and CF calls of bats can have distinct frequency ranges in which they are produced. These individual calls can range as high as 100 kHz and as low as 25 kHz. Some species stay in a frequency range that makes them unique among the rest which makes them easy to identify. Some species like in the genus Myotis overlap in frequencies that make it impossible to identify them to species, although they can be identified to their genus (Krusic and Neefus, 1996). The drawback to recording ultrasonic frequencies of bat calls, is that the average user cannot identify many of the calls they record. Also, one must develop a reference call library of known bat calls (Britzke, 2003; Barclay, 1999) either

by collecting individual bats and recording them or use another call library someone has already developed.

There has been much debate on the accuracy of the identification of bat calls. Bat calls can be identified using either qualitative or quantitative techniques. The qualitative technique used to identify bat calls is visual identification using a call library to compare calls collected. This technique works well if the user has a large database of calls to compare. Quantitative techniques include discriminant function analysis (DFA) and artificial neural networks (ANN). Discriminant function analysis uses a multivariate statistics program to analyze each call file by calculating the probability of a given file being produced by a particular species, based on call shape characteristics. Some of the characteristics include the top of call frequency (F_{max}) and the bottom of the frequency $(F_{min}$) that when combined make the call, and the slope (Δ) represents the curve of the call (Parsons and Jones, 2000). Artificial neural networks help the user to identify the bat calls recorded by extracting call characteristics or attributes that the user can train the computer to recognize (Parsons and Jones, 2000). Although there are different types of qualitative and quantitative techniques used to identify bats, studies have shown that discriminative functional analysis, ANN, and human visual identification all produce the same results in terms of number of correct calls identified (Jennings et al., 2008). This shows that there are many options to use when identifying bat calls, but to maintain accurate identification, one must use complete call sequences to correctly identify calls to species. It is important to realize when using the types of techniques that not all calls can be indentified and thus must be placed in some type of unidentified category when the

call sequences are incompletely recorded or the quality of the call is poorly recorded (Corben, 2002).

CHAPTER II

MATERIALS AND METHODS

Study Sites

The United States Department of Agriculture, Forest Service, Land Between the Lakes (LBL) is a national recreational area that encompasses >170,000 acres with 300 miles of undeveloped shoreline. The park resides in three counties: Stewart County in Tennessee, and Trigg County and Lyon County in Kentucky (Figure 2). Land Between the Lakes is delineated by two main rivers, Tennessee River to the West and Cumberland River to the North and East. Both of these rivers are impounded, Kentucky Lake from the Tennessee River and Lake Barkley from the Cumberland River (Figure 2). Land Between the Lakes was managed and maintained by Tennessee Valley Authority until 1998 when the U.S. Forest Service took over ownership. The national recreational park contains various habitats ranging from recreational areas, such as ATV mobile tracks and camping sites, to many distinctive habitats, such as old growth forest, streams, rivers and various grassland habitats. Land Between the Lakes also contracts out small parcels of land to farmers to plant various agricultural products.

Identification of species of bats considered for the study

The bats species that reside within LBL are the Little Brown (Myotis lucifugus),
Gray Bat (Myotis grisescens), Northern Long-eared (Myotis septentrionalis), Tri-colored
(Perimyotis subflavus), Big Brown (Eptesicus fuscus), Eastern Red Bat (Lasiurus
borealis), Hoary (Lasiurus cinereus), Silver-haired (Lasionycteris noctivagans) and
Evening (Nycticeius humeralis) (U.S. Forest Service, Personal Communication). One out

of the 10 species that reside in LBL is considered endangered, which is the Gray bat. The Gray bat is considered by U.S Fish and Wildlife an endangered species since 1976 due to habitat degradation and loss (Fish and Wildlife Service, 1997). On the Cumberland River side of LBL there is a significant Gray bat summer roost site at Tobacco Port cave. To date there has not been any record of the other endangered species that occurs in the southeast the Indiana Bat, *Myotis sodalis*.

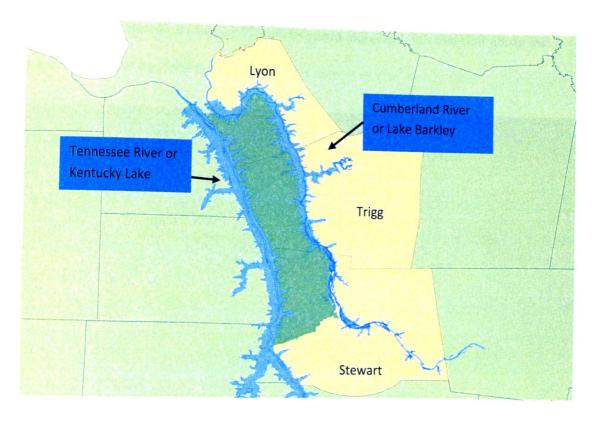


Figure 2. A map of LBL (shaded in dark green). The map shows the two rivers, Tennessee and Cumberland, which encompasses LBL and the three counties it resides within, Stewart county TN, Trigg and Lyon, counties KY.

Acoustic Collection of Bat Calls

Acoustic methods of sampling were used to record bat vocalizations throughout various habitats in LBL during the months of June and July 2009-2011. The acoustic sampling of bat vocalizations was done driving north-to south along three separate transects and at five stationary sites. The Anabat detector was strapped on the roof by a homemade protective container (Figure 3) made by Steralite©. The survey vehicle, Ford Sport Trac, was driven between 15-20 mph to record individual bats that flew above the vehicle while driving along each transect.

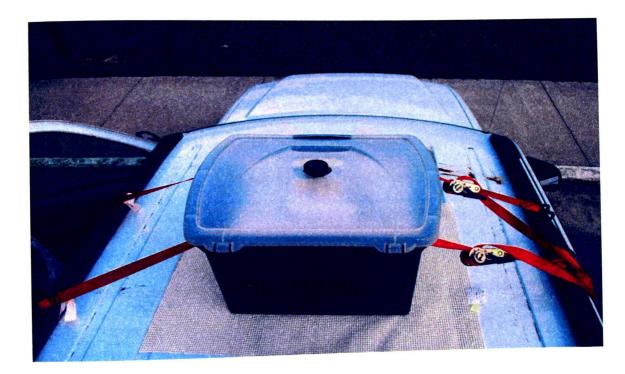


Figure 3. Photograph of the homemade protective box with the Anabat Microphone protruding from the surface.

Driven transects

The three driven transects (approximately 20-30 miles each) were as follows: the Lake Barkley side of LBL, the Tennessee River side of LBL, and the North side of LBL in Kentucky. Each transect traversed different parts of LBL, which included multiple habitats (Figure 4). The research was conducted with the assistance of other researchers from the university and supported by a contract with the U.S. Forest Service. The acoustic routes were driven over the course of three days. On the first day, transects one and two were done using two separate vehicles. On the second day, transect three was completed, while also sampling the stationary points with the assistance of other researchers.

The transect sampling began each night at sunset. Both vehicles met to prepare the vehicles for the survey at the Forest Service's administrative building. The transects were driven at an average of 15-20 mph for a distance of 30 miles. The Tennessee Whitenose guidelines (Fish and Wildlife, 2009 and 2010) were followed for the speed and distance. The microphone sensitivity of the detectors was adjusted accordingly, based on amount of insect noise being recorded versus the number of bats being recorded during the designated time period. As an example, the microphone sensitivity began at setting of 7 each night and was never lowered below 3, regardless of how much insect noise occurred, in order to assure proper continuous recording. During the first night both vehicles surveyed Eastern route (Lake Barkley side) and Western (Tennessee River side). During the second night one vehicle drove the north side of LBL, while the other group surveyed the five stationary points using the Anabat acoustic sampling devices.

Stationary sites

The five stationary sites that were surveyed include many habitat types, some are natural and others have been disturbed by human activities. The stationary sites were chosen because they encompassed the most likely diverse foraging habitats for bats. The first stationary site is Wrangler's Camp that has been heavily impacted by humans. This area is used for recreational activity specifically used as a horse camp. The second stationary site is Prior Bay that is one of many LBL embayments of Barkley Lake. This inlet is filled with water from the Cumberland River and dries completely during fall and winter. The third stationary site is Agricultural Field. These field habitats rotate between two agricultural crops, soy beans one season and corn another. The fourth stationary site is Logging Site. Before the study this site was heavily forested but the ice storm of 2009 destroyed most of the forest, which led to a salvage logging operation to thin the damaged forest. The fifth stationary site is Baird's Lake. Baird's Lake is another embayment of LBL but contains water all year around from the Cumberland River. This site also contains a gravel boat ramp to access the water. At each stationary site, the Anabat and Avisoft detectors were set out side by side to record for 10 minutes. Both were angled towards the same direction, straight up, at the same settings. These settings were between 15 kHz and 200 kHz, which will begin recording when the bat with that specific signature is detected, and will continue recording until no bats are in the area. Each site was surveyed in this order during each survey period, Wrangler's Camp, Prior Bay, Corn Field, Logging Site, and Baird's Lake. All appropriate Field Notes were recorded for each area in a notebook for later reference.

Geographic Information System mapping

A Geographic Information System (GIS) map was developed for the study area (Figure 4). The map contains transect routes, stationary locations and other useful information when analyzing the data collected. Each location was assigned attributes (Latitude, Longitude, Habitat type etc.) based on that area and placed in a Microsoft Excel file. The GIS map helped assess the habitats that were surveyed and to determine approximate driving similarity of each transects (Jaberb and Antione 2001). Also, the GIS map assisted with defining the study area and spatially locating the stationary sampling sites at varying habitat types.

Land Between the Lakes Forest Service Acoustic Bat Survey Map

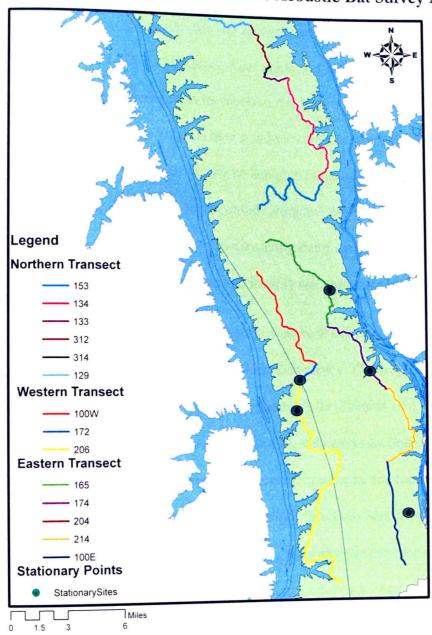


Figure 4. A GIS map of LBL including driven transects and stationary acoustic sampling sites. The different colors (road numbers) for each transect represents the road traveled or route for a specific transect. The green dots represent each stationary sampling site.

Analysis of acoustic data

The Anabat detector acoustic data was analyzed using the AnaLookW v. 4.8q software that is included with the detectors. AnaLookW v. 4.8q was used to display all call sequences recorded. Pre-made species labels in AnaLook v. 4.8q were used to assign various bat species identification tags so they could be sorted for when identified. Filters removed unwanted noise lower than 14 kHz so that high frequency bat calls were left to analyze. The frequency sweeps, the degree of the sweep and the frequency range, are used to identify each bat call sequence to species using visual identification. Known call sequence references were used to compare and identify call sequences recorded.

Avisoft-SASLab Pro v. 5.2.04 was used to view each call sequence via spectrographs and listen to each recorded file from each survey. The program also removed unwanted noise lower than 14 kHz. Sonobat v. 3.03 (Sonobat.com, 2011) was used to analyze all Avisoft .wav files recorded for each stationary site. Sonobat v. 3.03 extrapolated the .wav files that contained the frequency, amplitude, and time length to identify the files to species. Also, known call reference files from Sonobat v. 3.03 were visually compared to survey call files. After all files were identified to species for Anabat and Avisoft unit the data then were placed into a Microsoft Excel table for further evaluation. An ANOVA was used to compare means of normally distributed data. A Wilcoxon test was performed to compare means of non-normal distributed data.

Refer to the Appendix A for more information on equipment and software settings. Refer to the Appendix B for more information on how each call file sequence was identified by using both AnaLookW v. 4.8q and Sonobat v. 3.03.

CHAPTER III

RESULTS

During the course of three years a total of 4,350 Anabat call sequence files were recorded for all transects. A combined total of 622 for Anabat and Avisoft call sequence files were recorded for all stationary sites. Each of the call sequence files were identified to species with specific labels: Big Brown bat (EPFU), Red bat (LABO), Silver-haired bat (LANO), Hoary bat (LACI), Grey bat (MYGR), Little Brown bat (MYLU), Northern Long-eared bat (MYSE), Myotis species (MYsp), Evening bat (NYHU), Tri-colored bat (PESU), unidentified, and noise. Unidentified, noise and MYsp are the only labels that contain multiple meanings. Unidentified means that a bat call was recorded but not enough information (quality of call, .wav files or number of calls) was collected for identification. Noise is any unwanted sound, abiotic (car, wind, etc.) or biotic (insect, etc.), that was recorded at ultrasonic frequencies. Myotis species are bats of the Myotis genus. Myotis received their own label because this genus can be particularly difficult to indentify sometimes unless specific known characteristics are recorded on that individual file, such as MYGR's abrupt tapering off of frequency at 45 kHz. When the identifying sound characteristics of Myotis call sequences were not recorded, the file was labeled as a Myotis type call because of the frequency range in which the genus occurs, classifying it as MYsp. Each file recorded by both recording devices, for each species, does not necessarily represent one individual bat of that species, but a call sequence collected that is produced by that species. Individual bats produce numerous calls per millisecond, and the number displayed in the data tables should therefore not be confused with number of

bats present at a given site or transect. For example, one EPFU could have produced eight of the recorded call sequences for transect 3 of June 6th, 2009.

The call totals for each of the transects over the course of each year were compared to see if there was a difference among the amount of calls recorded in each transect. The data was non-normal and was therefore transformed by taking the Log of the original dataset. Also, unidentified calls and noise were excluded from the analysis. For 2009-2011 the call totals by transects were compared using a one-way ANOVA. For 2009 the call totals among transects 1-3 were not significantly different (p > F = .9050 at .05 alpha level). For 2010 the call totals among transects 1-3 were not significantly different (p > F = .1360 at .05 alpha level). For 2011 the call totals between transects 1-3 were not significantly different (p > F = .9216 at .05 alpha level). For all years combined the call totals were compared among transects 1-3. When call totals for all years were compared among each of the transects they were not significantly different (p > F = .7754 at .05 alpha level).

The call totals for each of the stationary sites over the course of each year were compared to see if there was a difference among the amount of calls recorded in each transect. Also, unidentified calls and noise were excluded from the analysis. For 2009-2011 the call totals by transects were compared using Wilcoxon test. For 2009 the call totals among the five stationary were not significantly different (p > F = .1620 at .05 alpha level). For 2010 the call totals among the stationary sites were not significantly different (p > F = .0596 at .05 alpha level). For 2011 the call totals between transects 1-3 were not significantly different (p > F = .2898 at .05 alpha level). For all years combined the call totals were compared among all stationary sites. When call totals for all years

were compared among each of the stationary sites they were significantly different (p > F = .0122 at .05 alpha level).

In 2009 the total for transect call sequence files recorded was 1,315 and for stationary files was 158. Table 1 shows the total number of all files recorded and what files were most abundant for 2009 transects. In table 2 are the totals of all files recorded for each stationary site for 2009. The 2009 survey was done a total of three times, once in June and twice in July, and all other surveys were done a total of two times due to contract agreements.

Table 1. In 2009 a total of 1,315 files were recorded for all three transects. The totals for species calls and other files are represented in this table for each sample period and for each transect. The LABO, PESU, unidentified and noise were the most abundant files recorded during 2009. For 2009 the call totals (excluding unidentified and noise) among transects 1-3 were not significantly different (p > F = .9050 at .05 alpha level).

Area	Date	EPFU	LABO	LANO	LACI	MYGR	MYLU	MYSE	MYsp	NYHU	PESU	Unidentified	Noise	Total
Transect 1	6/24/2009	0	11	0	0	2	0	0	4	4	1	18	14	54
Transect 2	6/24/2009	0	13	0	0	0	0	0	1	2	11	19	17	63
Transect 3	6/25/2009	0	27	0	0	0	0	0	0	11	18	48	18	122
Transect 1	7/1/2009	1	15	0	2	1	0	0	2	9	5	49	3	87
Transect 2	7/1/2009	1	68	0	0	0	1	0	3	4	16	47	17	157
Transect 3	7/2/2009	8	52	0	2	0	2	0	0	7	42	77	65	255
Transect 1	7/7/2009	2	27	1	1	0	1	0	3	1	23	25	36	120
Transect 2	7/7/2009) (34	0	0	1	0	0	1	7	67	68	60	238
Transect 3	7/8/2009) 4	39	0	0	0	0	0	3	11	65	71	26	219
Total		16	286	1	5	4	4	0	17	56	248	422	256	1315

Table 2. In 2009 a total of 158 files were recorded for all five stationary sites. The totals for species and other files are represented in this table for each sample period and for each individual stationary site. The LABO, PESU, and unidentified were the most abundant files recorded during 2009. For 2009 the call totals (excluding unidentified and noise) among the five stationary were not significantly different (p > F = .1620 at .05 alpha level).

Area	Date	EPFU	LABO	LANO	LACI	MYGR	MYLU	MYSE	MYsp	NYHU	PESU	Unide ntifie d	Noise	Total
Wrangler's Camp	6/25/2009	0	9	0	0	0	0	0	0	3	2	10	(24
Prior Bay	6/25/2009	0	2	0	0	0	0	0	0	1	6	7	() 16
Corn Field	6/25/2009	0	3	0	0	0	0	0	0	0	0	2	0	5
Logging site	6/25/2009	0	0	0	0	0	0	0	0	0	1	0	1	2
Baird's Lake	6/25/2009	0	0	0	0	0	0	0	0	0	0	3	0	3
Wrangler's Camp	7/2/2009	0	0	0	0	0	0	0	0	0	0	2	0	2
Prior Bay	7/2/2009	0	6	0	0	0	0	0	2	1	4	11	0	24
Corn Field	7/2/2009	C	0	0	0	0	0	0	0	0	0	1	1	2
Logging site	7/2/2009	(0	0	0	0	0	0	0	0	0	1	1	2
Baird's Lake	7/2/2009	(0	0	0	0	0	0	1	0	0	0	1	2
Wrangler's Camp	7/8/2009) (5	0	0	0	0	0	0	0	14	5	0	24
Prior Bay	7/8/2009) (7	0	1	0	0	0	1	0	18	11	0	38
Corn Field	7/8/2009) (0 2	. 0	0	0	1	0	1	0	0	2	1	7
Logging site	7/8/2009	9 (0 1	0	0	0	0	0	0	1	3	2	0	7
Baird's Lake	7/8/2009	9 (0 0	0	0	0	0	0	0	0	0	0	0	0
Total			0 35	0	1	0	1	0	5	6	48	57	5	158

In 2010 the total number of call sequence files recorded for each of the transects was 680 and 149 for all stationary sites. Table 3 shows the total number of all files recorded and what files were most abundant for each of the 2010 transects. Table 4 shows the totals of all files recorded for each stationary site for 2010. The total data collected for 2010's transects were the lowest out of the three years surveyed.

Table 3. In 2010 a total of 680 files were recorded for all three transects. The totals for species and other files are represented in this table for each sample period and for each transect. The LABO, PESU, unidentified and noise were the most abundant files recorded during 2010. For 2010 the call totals (excluding unidentified and noise) among transects 1-3 were not significantly different (p > F = .1360 at .05 alpha level).

Area	Date	EPFU	LABO	LANO	LACI	MYGR	MYLU	MYSE	MYsp	NYHU	PESU	Unidentified	Noise	Total
Transect 1	6/29/2010	0	0	0	0	0	0	0	0	0	0	7	20	27
Transect 2	6/29/2010	1	47	0	0	1	2	0	0	16	34	51	58	210
Transect 3	6/30/2010	1	15	0	0	1	1	0	0	0	24	37	21	100
Transect 1	7/13/2010	0	4	0	0	0	0	0	0	0	8	82	44	138
Transect 2	7/13/2010	2	15	0	0	1	0	0	1	0	24	32	16	91
Transect 3	7/14/2010	(27	0	0	1	0	0	2	1	43	33	7	114
Total		4	108	0	0	4	3	0	3	17	133	242	166	680

Table 4. In 2010 a total of 149 files were recorded for all five stationary sites. The totals for species and other files are represented in this table for each sample period and for each individual stationary site. The PESU and unidentified were the most abundant files recorded during 2010. For 2010 the call totals (excluding unidentified and noise) among the stationary sites were not significantly different (p > F = .0596 at .05 alpha level).

Area	Date	EPFU	LABO	LANO	LACI	MYGR	MYLU	MYSE	MYsp	NYHU	PESU	Unidentified	Noise	Total
Wrangler's Camp	6/30/2010	0	0	0	0	0	0	0	0	0	0	1		3 9
Prior Bay	6/30/2010	0	1	0	0	0	0	0	0	6	24	20	(51
Corn Field	6/30/2010	0	0	0	0	0	0	0	0	0	0	1	1	2
Logging site	6/30/2010	0	0	0	0	0	0	0	0	0	0	0	0	0
Baird's Lake	6/30/2010	0	4	0	0	0	0	0	0	0	2	0	2	8
Wrangler's Camp	7/14/2010		0	0	0	0	0	0	0	0	0	0	2	2
Prior Bay	7/14/2010) 1	. 8	0	0	0	0	0	2	5	27	8	0	51
Corn Field	7/14/2010) (0	0	0	0	0	0	0	0	4	6	0	10
Logging site	7/14/2010	0 () 1	. 0	0	0	0	0	0	0	3	3	1	8
Baird's Lake	7/14/201	0) (0	0	0	0	0	1	0	3	3	1	8
Total			1 14	0	0	0	0	0	3	11	63	42	15	149

In 2011 the total number of files recorded for each of the transects was 2,355 and for stationary it was 355. Table 5 shows the totals of all files recorded and what files were most abundant for each of the 2010 transects. Table 6 shows the totals of all files recorded for each stationary site for 2010. The total data collected for 2011's transects were the highest out of the three years surveyed.

Table 5. In 2011 a total of 2,355 files were recorded for all three transects. The totals for species and other files are represented in this table for each sample period and for each transect. The LABO, PESU, NYHU, unidentified and noise were the most abundant files recorded during 2011. For 2011 the call totals (excluding unidentified and noise) between transects 1-3 were not significantly different (p > F = .9216 at .05 alpha level).

Area	Date	EPFU	LABO	LANO	LACI	MYGR	MYLU	MYSE	MYsp	NYHU	PESU	Unidentified	Noise	Total
Transect 1	6/22/2011	2	21	0	0	0	0	0	0	4	32	22	25	106
Transect 2	6/22/2011	2	13	0	0	1	1	0	9	29	24	32	26	137
Transect 3	6/23/2011	1	8	0	0	0	0	0	1	12	17	9	23	71
Transect 1	7/12/2011	4	19	1	0	1	4	0	1	30	105	115	618	898
Transect 2	7/12/2011	. () 22	0	0	0	4	0	0	26	69	91	437	649
Transect 3	7/13/2011	1	1 12	. 0	0	0	2	0	0	38	95	76	270	494
Total		10	95	1	0	2	11	0	11	139	342	345	1399	2355

Table 6. In 2011 a total of 355 files were recorded for all three transects. The totals for species and other files are represented in this table for each sample period and for each transect. The LABO, PESU, unidentified and noise were the most abundant files recorded during 2011. For 2011 the call totals (excluding unidentified and noise) between transects 1-3 were not significantly different (p > F = .2898 at .05 alpha level).

Area	Date	EPFU	LABO	LANO	LACI	MYGR	MYLU	MYSE	MYsp	NYHU	PESU	Unidentified	Noise	Total
Wrangler's Camp	6/23/2011	0	0	0	0	0	0	0	0	0	0	0	1	1
Prior Bay	6/23/2011	1	1	0	0	0	0	0	0	0	6	1	2	11
Corn Field	6/23/2011	0	0	0	0	0	0	0	0	0	0	0	0	0
Logging site	6/23/2011	0	0	0	0	0	0	0	0	1	3	3	0	7
Baird's Lake	6/23/2011	0	0	0	0	0	0	0	1	0	0	1	3	5
Wrangler's Camp	7/13/2011	0	0	0	0	0	0	0	0	0	0	0	1	1
Prior Bay	7/13/2011	0	0	0	0	0	0	0	0	5	40	1	0	46
Corn Field	7/13/2011	. 0	0	0	0	0	0	0	0	1	10	2	0	13
Logging site	7/13/2011	0	1	0	0	0	0	0	0	4	8	5	95	113
Baird's Lake	7/13/2011	1 0	1	0	0	0	0	0	0	1	0	4	112	118
Total		1	3	0	0	0	0	0	1	12	67	17	214	315

The total number of files recorded during the three year course of this study for transects was 4,350 and for stationary sites 622. In figure 6 the totals were compiled for each transect to display what species specific call files were recorded the most for each transect during the entire study. Like the individual year data the most abundant call sequences recorded are displayed from largest to smallest: noise, unidentified, PESU, LABO, and NYHU. Figure 7 represents the same data that figure 6 displayed but for stationary sites.

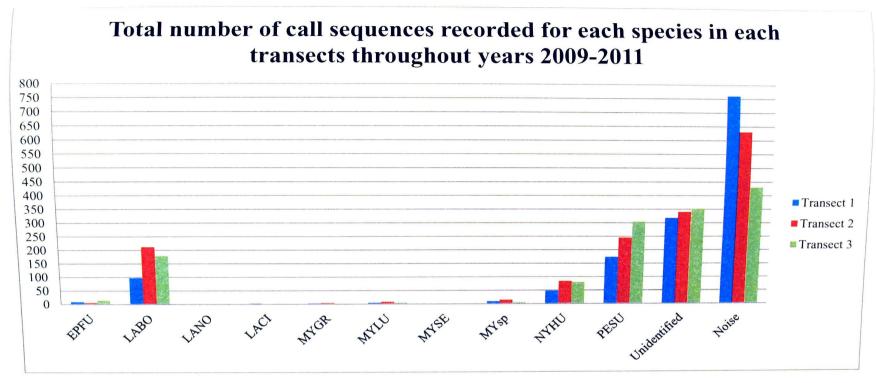
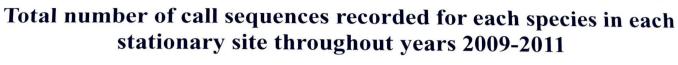


Figure 5. The data collected for each Anabat file recorded was compiled for each transect to illustrate for the entire study what files were most abundantly recorded. When call totals (excluding unidentified and noise) for all years were compared among each of the transects they were not significantly different (ANOVA, p > F = .7754 at .05 alpha level).



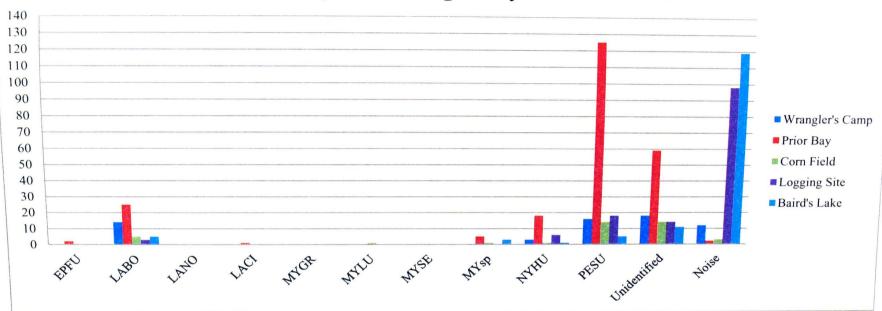


Figure 6. The data collected for each Anabat and Avisoft call sequence file recorded was compiled for each stationary site to illustrate for the entire study what files were most abundantly recorded. When call totals (excluding unidentified and noise) for all years were compared among each of the stationary sites they were significantly different (Kruskal-Wallis test, p > F = .0122 at .05 alpha level).

During the course of this study a majority of species call sequences were recorded but there was also an abundance of recorded files that could not be used for species identification. These type of files (unidentified and noise) could contain bat calls but not sequences that could be led to genus or species identification and these files are called unidentifiable. Table 7 shows the total files collected for both transects and stationary sites that indicates out of the grand total for the study what percentage of files were bat calls recorded or unidentifiable.

Table 7. The total number of files recorded for the entire study was accessed for usability for both transects and stationary sites. Unidentifiable was more that 50% for both.

Tr	ansects	Stationary Sites			
Unidentifiable Bat calls recorded		Unide ntifiable	Bat calls recorded		
2830	1520	350	272		
65.06%	34.94%	56.27%	43.73%		

The presence or absence of species was determined between the two types of recording methods, transects and stationary sites (Table 8). Only transects 1 and 2 were compared to the stationary sites since those sites were along the two adjacent to transects. Stationary sites could not be placed along transect 3 due to inaccessibility of some of the roads during the beginning of the study.

Table 8. A comparison of total years of species specific calls for transects and stationary sites.

Species	Transects	Stationary Site
EPFU	Yes	Yes
LABO	Yes	Yes
LANO	Yes	No
LACI	Yes	Yes
MYGR	Yes	No
MYLU	Yes	Yes
MYSE	No	No
Mysp	Yes	Yes
NYHU	Yes	Yes
PESU	Yes	Yes

The presence or absence of bat species using transects versus mist-netting was assessed. The mist-netting sites are historical records that were set up along all three transects. Each transect location was compared with the corresponding mist-net locations.

Table 9. Transect 1 for 2009-2011 was compared to historical mist-netting data. Mistnetting captured one more bat species that the acoustical transects, the MYSE.

Species	Transects	Mist-netting
EPFU	Yes	Yes
LABO	Yes	Yes
LACI	Yes	Yes
LANO	Yes	Yes
MYGR	Yes	Yes
MYLU	Yes	Yes
MYSE	No	Yes
NYHU	Yes	Yes
PESU	Yes	Yes

Table 10. Transect 2 for 2009-2011 was compared to historical mist-netting data. Mist-netting captured all species except EPFU. The acoustic transects did not record LACI,

Species	Transects	Mist-netting
EPFU	Yes	No
LABO	Yes	Yes
LACI	No	Yes
LANO	No	Yes
MYGR	Yes	Yes
MYLU	Yes	Yes
MYSE	No	Yes
NYHU	Yes	Yes
PESU	Yes	Yes

Table 11. Transect 3 for 2009-2011 was compared to historical mist-netting data. Mist-netting captured all species except EPFU, LACI, and LANO. The acoustic transects did not record LANO and MYSE.

Species	Transects	Mist-netting
EPFU	Yes	No
LABO	Yes	Yes
LACI	Yes	No
LANO	No	No
MYGR	Yes	Yes
MYLU	Yes	Yes
MYSE	No	Yes
NYHU	Yes	Yes
PESU	Yes	Yes

The comparison of species presence or absence was assessed between two recording devices, Anabat and Avisoft. The two recording devices recorded various bat species but neither technique recorded MYSE. Also, some species were never recorded at specific sites. Table 12 represents the comparison of presence or absence between both detectors.

Table 12. Each stationary site was surveyed by using two detectors. Anabat and Avisoft both detected various bat species. Names indicate which recording technique recorded a particular bat species

Species	Wrangler's Camp	Prior Bay	Corn Field	Logging Site	Baird's Lake
EPFU	Neither	Both	Neither	Neither	Avisoft
LABO	Anabat	Both	Anabat	Anabat	Both
LACI	Neither	Anabat	Neither	Neither	Neither
LANO	Neither	Neither	Neither	Neither	Avisoft
MYGR	Neither	Neither	Neither	Neither	Neither
MYLU	Neither	Neither	Anabat	Neither	Neither
MYSE	Neither	Neither	Neither	Neither	Neither
NYHU	Anabat	Both	Anabat	Anabat	Anabat
PESU	Anabat	Both	Both	Anabat	Both
	Anabat/Avisoft	Anabat/Avisoft	Anabat/Avisoft	Anabat/Avisoft	Anabat/Avisoft

CHAPTER IV

DISCUSSION

The driving transects and stationary sites surveyed during this study recorded a diversity of bat species throughout LBL. Other studies have successfully used these types of acoustic surveys to assess unknown populations of bats over large areas (Walsh and Harris, 1996; Russ et al., 2003). Transects can also be surveyed while walking, and this study method has shown that similar types of data can be gathered when dealing with a large-scale area and varying habitats (Ellison et al., 2005). The ability to use these types of techniques to define bat populations over large-scale areas is important when outlining and implementing management techniques.

Transects and Stationary Sites species occurrence

The data collected during the three years of this study included many of the species known to occur at LBL. During each year of study the number of each species recorded varied, and this was especially true for the species that were not as abundant. Each transect collectively sampled developed bat records for each of the following species: EPFU, LABO, MYGR, MYLU, MYsp, NYHU, and PESU, with LANO and LACI only being sampled on transects 1 and 2. Comparing the three years of data, MYSE was never recorded during any survey (Table 8). The most abundant species found for each of the transects, 2009-2011, were LABO and PESU. Comparing the three years of data for the stationary sites, the amount of bat species recorded varied as well and did not record as much variety of species as did the bat recordings by transects (Figures 5 and 6).

Important to note, the stationary sites were only surveyed for 10 minutes at specific sites. Transects went through a variety of sites with different habitats and were longer in terms of time and distance than the stationary sites. The stationary sites did not record any of the following species at all during the three years of study: LANO, MYGR, and MYSE (Table 8). The placement of the stationary sites might not have allowed for the recording of those species. Another factor could be related to the overall abundance of bat species in LBL, since the call sequences recorded for the transects were also low for certain species. The low abundance of different Myotis species may be due to the fact that their calls are similar in terms of call structure (Max/Min frequency, same call slopes, etc.) and are not easily identified by visual identification, which placed all call sequences of that genus into the category MYsp (Jung et al., 1999; Russo and Jones, 2003). This might lower the number of individual Myotis species call sequences recorded when placed in a group as a whole (Ellison et al., 2005).

Transects versus Mist-netting

The historical mist-netting data that was set up along the three transects indicated a few differences in presence or absence of bat species (Figures 9-11). Both bat sampling methods, acoustic sampling by driven transects and historical field mist-netting data collected different species. When one sampling method did not collect a species, the other one had recorded it. Transect 3 was the only sampling data set where neither method collected a particular bat species, which was LANO. Both methods did not seem to have a specific preference for a specific bat species. All acoustic transects together detected almost all bat species throughout the area of LBL similarly to the historic netting data. The acoustic transects did not record any MYSE, but mist-netting did in every area.

This suggests that acoustic monitoring in general might not be an effective method to detect MYSE. Also, analysis of the recording data implies that surveying bat populations with multiple techniques should be used in those particular survey areas prior before establishing species assemblage.

Anabat versus Avisoft detectors

The acoustic data collected for each stationary site was collected by two different types of detectors. Each detector, Anabat and Avisoft, detected bat species for each of the stationary sites. The total species recorded for each detector varied among site, with neither detecting MYGR nor MYSE. Over the five stationary sites Avisoft was the only detector to record LANO and Anabat was the only detector to record LACI and MYLU. The sites over water, Prior Bay and Baird's Lake, had Avisoft detect more species versus when the device was recording over land. Anabat detected species at every stationary site regardless if near water or all land. The amount of calls recorded for stationary sites with these two techniques was still less when compared with the amount of calls recorded for transects.

Bat calls recorded and Unidentifiable files

During the course of this study many files were recorded for both transect and stationary site sampling methods. Both of these methods followed the same pattern for the amount of unidentifiable data files collected. For transects 1-3 more than half of the files collected (65%) were unidentifiable, with noise making up 42%. Unidentified category made up the other percentage of unidentifiable files, which was 13%. For all of the stationary sites more than half of the files as well (56%) were unidentifiable. The

that were unidentifiable. Unidentified files recorded only fragments of bat calls (< 2 call sequences or half of a call), which were not enough to positively identify the species producing the calls (O'Farrell et al., 1999). This type of data collected could have been caused by a number of possibilities. The detector might have been recording passing bats that were not travelling through the entire zone of reception, creating only partial sequences and causing low call quality. This may have also been caused by the driving speed of the vehicle, which may have passed the bats before the file could finish recording. The percentage of unidentifiable files in this study affects the number of bats call sequences that were detected. Since the detector only records the loudest sounds that pass by, bats calls may have been drowned out by other louder, unwanted noises that were recorded instead.

Transects and Stationary Sites

The state of TN has volunteers which survey transects one to three times a year. Most of the acoustic data recorded in this study were sporadic and small. The data collected tended to be the same type of species each time. This could mean that either the density of those species present in LBL could be high, or that the Anabat could be particularly sensitive for calls produced by those species. The large amount of unidentifiable data resulted in a small amount of bat call sequences recorded, but the techniques used in this study make it impossible to avoid. Insect, car and terrain noise could be some of the many factors contributing to the abundance of noise collected. Each of these noises produced some type of ultrasonic frequency which the Anabat could detect, but cannot filter out due to the detectors capabilities. The only way to get enough

data is to run transects and stationary sites more frequently throughout the season. This would allow for more chances to catch more species for each area sampled, since bats use different feeding areas and some of those species might not have been in the area during the transect or stationary survey (Ellison et al, 2005). Driving the transects more frequently and leaving the detectors at stationary sites longer will get all the events in that area overtime, which will give a better understanding of activities in those specific areas, especially areas of higher use or even higher species presence.

Acoustic monitoring of bats is cost effective and not time consuming. The two types of monitoring in this study both have pros and cons that should be taken into consideration when surveying bat populations. Transect monitoring provides large amounts of data quickly over a large-area and requires only one detector to collect the data. Although you can collect data over a large-area with transects this method of survey is dependent on which areas are accessible by roads. Stationary sites cannot collect data over a large-area unless you have multiple detectors, which can be costly, and multiple sites to conduct the survey. Stationary surveys are always confined to a specific area but can be left out passively for many nights of recording. Also, if an area is less developed then multiple stationary sites and multiple nights are the best method of recording data over large-scale areas.

Both of these acoustic monitoring techniques, transect and stationary, can be used for determining the presence or absence of species in an area, however, often the data collected cannot be used to assess population numbers, simply bat abundance or habitat use activity (Walsh and Harris, 1996; Zimmerman and Glanz, 2000; Remington and Cooper, 2009). The misconception that the call sequence files collected represent the

number of individuals is a common mistake. Each file recorded should be considered when portraying the activities in each area. If continuous data is recorded in a certain area over many years then one may conclude that those areas have high or low activities for a particular species. Until a large amount of recording data is collected using acoustic methods then the data collected can only show the presence or absence of a species on that particular night. Past netting surveys, like the historical mist-netting data, should also be consulted in order to determine what species have been historically documented in that area.

White-nose syndrome and long-term management for bat species

The current status of bat populations are at an all time low with species numbers continuing to dwindle, with loses already >5.5 million (U.S. Fish and Wildlife, 2012). A hands-off approach to studying bat populations, such as acoustic monitoring, would be the best solution to conduct field surveys with the least possible disturbance to individuals and bat roost sites. Continuous use of acoustic monitoring can establish presence or absence of bat species, especially with increased regulations for decontamination of mist-nets or devices that come in physical contact with bat species. The use of acoustic monitoring should not be restricted to a set number of times during the year, but performed consistently throughout the year to insure that all species are identified. In addition, other activities such as feeding migration and mating that rely on bat vocalizations should be incorporated into the sampling so they can be documented within an area. It is also important to not restrict acoustic monitoring techniques to just one type, but to use multiple sampling techniques in areas where it is appropriate or even different recording devices.

For example, techniques in this study recorded a variety of species, but neither collected MYSE but mist-netting collected this species. Krusic et al. (1996) suggested that the MYSE were too low for the Anabat to detect. Faure et al. (1993) suggested that MYSE were less audible out of all the MYsp. This could suggest that another technique or piece of equipment should be used to record MYSE. The comparison between the Anabat and Avisoft indicated that one detector did better over water than the other but one was generally suitable across the habitats. In general the techniques used in this study caused an underrepresentation of MYsp and one may consider alternate techniques when working with *Myotis* since the species in this genera are sympatric (Weller et al., 2007). Also, the transects used in this study recorded Grey bats, which are an endangered species and Myotis. The stationary sites unfortunately did not collect this endangered species at all using the two different recorders. This would strongly suggest that multiple acoustic techniques should be performed in an area when looking for the presence or absence of a particular species.

LITERATURE CITED

- AHLEN I. AND BAAGØE H. 1999. Use of ultrasound detectors for bat studies in Europe: experiences from field identification, surveys, and monitoring. Acta Chiropterologica 2: 137-150.
- ARNOLD AIR FORCE BASE. 2009. White-nose Syndrome Cooperative Monitoring and Response Plan for Tennessee.
 - $\underline{http://www.fws.gov/cookeville/pdfs/wnstnrespplan091124.pdf}$
- ARNOLD AIR FORCE BASE. 2010. White-Nose Syndrome Monitoring and Response Report for Tennessee.
 - $\underline{http://www.caves.org/WNS/Tennessee\%20Report\%202009-2010\%5B1\%5D}.pdf$
- BARBOUR, R. W. AND W. H. DAVIS. 1969. Bats of America. University Kentucky Press, Lexington.
- BARCLAY, R. M. R. 1999. Bats Are Not Birds: A Cautionary Note on Using

 Echolocation Calls to Identify Bats: A Comment. The Journal of Mammalogy
 80:290-296.
- BLEHERT, D. S., A. C. HICKS, M. BEHR, C. U. METEYER, B. M. BERLOWSKI-ZIER, E. L. BUCKLES, J. T. H. COLEMAN, S. R. DARLING, A. GARGAS, R. NIVER, J. C. OKONIEWSKI, R. J. RUDD, AND W. B. STONE. 2008. Bat White-Nose Syndrome: An Emerging Fungal Pathogen? Science 323:227.
- BRITZKE, E. 2003. Use of ultrasonic detectors for acoustic identification and study of bat ecology in the eastern united states. Dissertation. Tennessee Technological University, Nashville, Tennessee.

- CORBEN, C. 2002. Zero-crossing analysis for bat identification: an overview. Bat Echolocation Research: tools, techniques, and analysis. Symposium.
- DUFFY, A. M., L. F. LUMSDEN, C. R. CADDLE, R. R. CHICK, AND G. R. NEWELL. 2000. The efficacy of Anabat ultrasonic detectors and harp traps for surveying microchiropterans in south-eastern Australia. Acta Chiropterologica 2: 127-144.
- ELLISON, L. E., A. L. EVERETTE AND M. A. BOGAN. 2005. Examining patterns of bat activity in Bandelier National Monument, New Mexico, by using walking point transects. The Southwestern Naturalist 50:197-208.
- FAURE, P. A., J. H. FULLARD AND J. W. DAWSON. 1993. The gleaning attacks of the Northern Long-eared bat, *Myotis septentrionalis*, are relatively inaudible to moths. Journal of Experimental Biology 178:173-189.
- FENTON, M. B. AND G.P. BELL. 1981. Recognition of species of insectivorous bats by their echolocation calls. Journal of Mammalogy 62:233-243.
- GALAMBOS, R. AND D. R. GRIFFIN. 1942. Obstacle avoidance by flying bats; the cries of bats. Journal of Experimental Zoology 89: 475-490.
- GRINNELL, A. D. AND D. R. GRIFFIN. 1958. The sensitivity of echolocation in bats.

 The Biological Bulletin 114:10-22.
- GARGAS, A., M.T. TREST, M. CHRISTENSEN, T.J. VOLK AND D.S. BLEHERT.

 2009. *Geomyces destructans* sp. nov. associated with bat white-nose syndrome.

 Mycotaxon 108:147-154.

- HOFFMANN, A., J. DECHER, F. ROVERO, J. SCHAER, C. VOIGT AND G. WIBBEIT. 2010. Field methods and techniques for monitoring mammals. Abc Taxa 19:482-529.
- HOURIGAN, C. L., C. P. CATTERALL, D. JONES AND M. RHODES. 2008.

 Comparisons of harp trap and bat detector efficiency for surveying bats in an urban landscape. Wildlife Research 35: 768-774.
- JABERB, C. AND G. ANTOINE. 2001. Modelling the distribution of bats in relation to landscape structure in a temperate mountain environment. Journal of Applied Ecology 38:1169-1181.
- JENNINGS N., S. PARSONS AND M. J. O. POCOCK. 2008. Human vs. machine: identification of bat species from their echolocation calls by humans and by artificial neural networks. Canadian Journal of Zoology 86:371-377.
- JUNG, T. S., I. D. THOMPSON, R. D. TITMAN AND A. P. APPLEJOHN. 1999.

 Habitat selection by forest bats in relation to mixed-wood stand types and structure in Central Ontario. The Journal of Wildlife Management 63:1036-1319.
- KRUSIC, R. A. AND C. D. NEEFUS. 1996. Bat habitat use in white mountain national forest. Journal of Wildlife Management 60: 625-631.
- KUENZI, A. J. AND M. L. MORRISON. 1998. Detection of bats by mist-nets and ultrasonic sensors. Wildlife Society Bulletin 26:307-311.
- LANCE, R. F., B. BOLLICH, C. L. CALLAHAN AND P. L. LEBERG. 1999. Surveying Forest-Bat Communities with Anabat Detectors.

- LEWIS, S. E. 1995. Fidelity of Bats: A Review. Journal of Mammalogy 76:481-496.
- MURRAY, K. L., E. R. BRITZKE, B. M. HADLEY AND L. W. ROBBINS. 1999.

 Surveying bat communities: a comparison between mist nets and Anabat II bat detector system. Acta Chiropterologica 1: 105-122.
- O'FARRELL, M. J., C. CORBEN, W. L. GANNON AND B. W. MILLER. 1999.

 Confronting the dogma: a reply. Journal of Mammalogy 80:297-302.
- O'FARRELL, M. J. AND W. L. GANNON. 1999. A comparison of acoustic versus capture techniques for the inventory of bats. Journal of Mammalogy 80:24-30.
- O'FARRELL, M. J., B. W. MILLER AND W. L GANNON. 1999. Qualitative identification of free-flying bats using the Anabat detector. Journal of Mammalogy 80:11-23.
- PARSONS, S., A. M. BOONMAN AND M. K. OBRIST. 2000. Advantages and
 Disadvantages of Techniques for Transforming and Analyzing Chiropteran
 Echolocation Calls. Journal of Mammalogy 81:927-938.
- PARSONS, S. AND G. JONES. 2000. Acoustic identification of twelve species of echolocating bat by discriminant function analysis and artificial neural networks.

 Journal of Experimental Biology 203:2641-2656.
- REICHARD, J. D. 2009. Wind-Damage Index Used for Characterizing Wing Condition of Bats Affected by White-nose Syndrome.

 <u>www.caves.org/WNS/Bat%20Wings.pdf.</u>

- REICHARD, J. D. AND T. H. KUNZ. 2009. White-nose syndrome inflicts lasting injuries to the wings of little brown myotis (*Myotis lucifugus*). Acta Chiropterologica 11:457-464.
- REMINGTON, S. AND D. S. COOPER. 2009. Bat survey of Griffith Park: draft report.

 Los Angeles Department of Recreation and Parks.
- RUSS, J. M., M. BRIFFA AND W. I. MONTGOMERY. Seasonal patterns in activity and habitat use by bats (*Pipistrellus* spp. And *Nyctalus leisleri*) in Northern Ireland, determined using a driven transect. Journal of Zoology (London) 259:289-299.
- RUSSO, D. AND G. JONES. 2003. Use of foraging habitats by bats in a Mediterranean area determined by acoustic surveys: conservation implications. Ecography 26:197-209.
- SCHNITZLER, H. U. AND E. K. V. KALKO. 2001. Echolocation by Insect-Eating Bats. BioScience 51:557-569.
- SCHNITZLER, H. U., C. F. MOSS AND A. DENZINGER. 2003. From spatial orientation to food acquisition in echolocating bats. Ecology and Evolution. 18:386-394.
- THOMAS, D. AND S. D. WEST. 1989. Sampling Methods for Bats. Forest Service.

 General Technical Report 9.
- U.S. FISH AND WILDLIFE SERVICE. 1997. Threatened and Endangered Species:

 Gray Bat. http://www.fws.gov/midwest/Endangered/mammals/pdf/gray-bat.pdf
- U.S. FISH AND WILDLIFE SERVICE. 2010. White-Nose Syndrome Decontamination Protocol (v.3).

- http://www.fws.gov/WhiteNoseSyndrome/pdf/WNS1pageDecontaminationProtocol_073110.pdf.
- U.S FISH AND WILDLIFE SERVICE. 2011. Supporting decontamination documentation for cavers: wns decontamination supplement 1 of 2.

 http://www.fws.gov/whitenosesyndrome/pdf/WNSDecon_Cavers_v012511.pdf
- U.S FISH AND WILDLIFE SERVICE. 2012. North American bat death toll exceeds 5.5 million from white-nose syndrome. News Release.

 http://www.fws.gov/whitenosesyndrome/pdf/WNS_Mortality_2012_NR_FINAL.pdf.

 pdf.
- UNITED STATES GEOLOGICAL SURVEY. Update on white-nose syndrome:

 Tennessee finding. USGS wildlife health bulletin. Reston (VA): The Survey;

 2010 [cited 2010 November 12].

 http://www.nwhc.usgs.gov/disease_information/white-nose_syndrome/.
- WALSH, A. L. AND S. HARRIS. 1996. Factors determining the abundance of vespertilionid bats in Britain: Geographical, land class and habitat relationships.

 Journal of Applied Ecology 33:519-529.
- WHITE E. P. AND S. D. GEHRT. 2001. Effects of recording media on echolocation data from broadband detectors. Wildlife Society Bulletin 29:974-978.
- WELLER, T. J., S. A. SCOTT, T. J. RODHOUSE, P. C. ORMSBEE AND J. M. ZINCK. 2007. Field identification of the cryptic vespertilionid bats, *Myotis lucifugus* and *M. yumanensis*. Acta Chiropterologica 9:133-147.

APPENDICES

APPENDIX A

Equipment Settings for Transect

Transect	Unit	Microphone	Detection Range	Angle of Microphone	Sensivity Ranges	Data Division	Audio Division
Transect 1	Anabat SD2	Standard Black Mic	30 meters	90°	Between 7-4	8	8
Transect 2	Anabat SD2	Green Mic	30 meters	90°	Between 7-4	8	8
Transect 3	Anabat SD2	Green Mic	30 meters	90°	Between 7-4	8	8

Equipment Settings for Stationary Sites

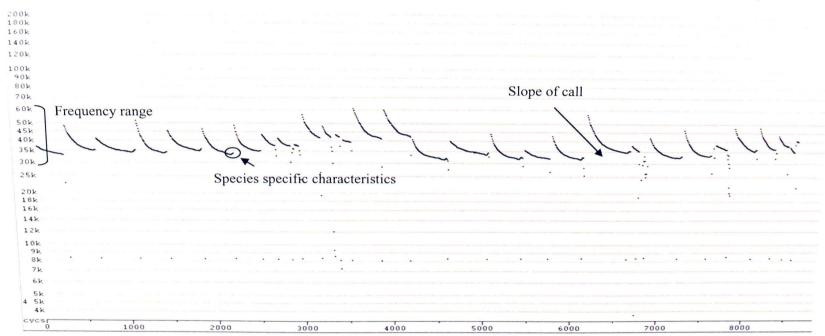
Stationary Site	Anabat/Avisoft Frequencies	Anabat/Avisoft Division Ratios	Time at each location	Angle of Detectors
Wrangler's Camp	15-200 kHz	8	10 minutes	45°
Prior Bay	15-200 kHz	8	10 minutes	45°
Corn Field	15-200 kHz	8	10 minutes	45°
Logging Site	15-200 kHz	8	10 minutes	45°
Baird's Lake	15-200 kHz	8	10 minutes	45°

Software Settings

Program	Noise Filters	Display Type	File Type	Data Extractor
AnaLookW 4.8q	Removed unwanted noise <14 kHz	Spectrograph	Anabat specific files	CFCread 4.3r
SASLab-Pro v. 5.2.04	Removed unwanted noise <14 kHz	Spectrograph	.wav file	RECORDER v. 4.2.9
Sonobat v. 3.03	Removed unwanted noise <14 kHz	Spectrograph	.wav file	Sonobat v. 3.03

APPENDIX B

Characteristics Used for Identification of Anabat Call Sequence Files with AnaLookW v. 4.8q.



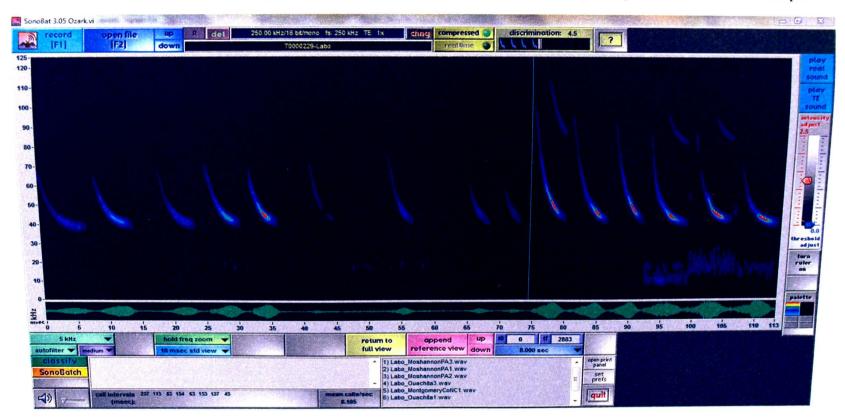
APPENDIX B CONTINUED

Characteristics Used for Identification of Avisoft Call Sequence Files with Sonobat v. 3.04.

120-110-100 -90-80-Sweep in frequency 70-Sweep in frequency 50-30-Amplitude (Coloration) 20-

APPENDIX B CONTINUED

Diagram of how Sonobat analysis survey call files against known call reference files, in this example below is a Red bat, Lasiurus borealis. The left call sequence is the unknown survey calls and the right sequence is the known comparison.



VITAE

Riley Seth McCormick graduated from Montgomery Central High School in 2006 and received his Bachelor's degree in Biology from Austin Peay State University in 2010. While pursuing his Undergraduate degree he presented at two Animal Behavior Society meetings while helping with The Center of Excellence for Field Biology's many projects. He was also awarded the Presidential Research scholarship twice to fund two separate undergraduate research projects. He started his Master's Degree in Fall of 2010 where he received a Graduate Assistantship through the Biology Department. He received a contract through the U.S. Forest Service and graduate research grant to fund his Master's research, and completed his Master's in Biology in the Spring of 2012.