

**Effectiveness of Aerial and Ground-Based Acoustic Bat
Detection Technologies**

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**Adrienne Michelle Dykstra
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ABSTRACT

Wildlife acoustic monitoring technologies have been advancing detection capabilities in many areas of species monitoring. Detection of species using remotely deployed technologies such as acoustic detectors provides population data with little effort, supplying critical information for many research analyses. Acoustic echolocation detectors have been used to detect and identify bats for several years, with constant improvements to the technologies utilized by these detectors. Because of the limitations associated with using a ground-based detection technology to monitor species flying at unknown altitudes, we designed and tested an Aerial Bat Detection Technology (ABDT) to monitor bats in flight at various altitudes (25m, 50m, 75m and 100m) above ground level. In addition, we tested the detection distance of Pettersson D500x (Pettersson Elektronik AB, Sweden) and SM2+ (Wildlife Acoustics Inc., Massachusetts, US) ultrasonic bat detectors, both of which are regularly used as part of bat monitoring programs. The ABDT was flown nightly for 3 months during the summer of 2016 and data collected by the ABDT was compared to data collected by a stationary ground-based acoustic detector run simultaneously in the same location. Of the 44 sampling nights, the ABDT recorded species missed by the ground-based detector on 20 nights. The total number of species and calls recorded by the two methods did not differ ($P= 0.676$ and $P= 0.122$, respectively) but calls/hr were different during the hours when the ABDT was located at 50m—100m ($P= 0.017$, $P= 0.001$ and $P= 0.005$, respectively). The ABDT generally recorded fewer

calls/hr at 50 – 100 m altitudes but detected species that were not recorded on the ground-based detector. Upon testing the acoustic detectors using a signal emitted at a constant frequency between 8 – 65 kHz (102.2 dB – 44.92dB, respectively, at 1m), we found that the maximum detection distances were much shorter than expected for both the Pettersson D500x and the SM2+, with the maximum detection distances being 17m and 22m, respectively. This information suggests that current ground-based acoustic bat detection methods likely provide an incomplete representation of bat communities in the eastern United States and that an aerial detector can enhance the overall monitoring approach.

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INTRODUCTION

Ultrasonic acoustic bat detectors are a widely used passive monitoring technology for surveying bat populations and are frequently employed by state agencies and environmental consulting groups. Passive monitoring of any species involves assessing the change or trend in relative abundance, composition, or distribution of populations over time without actively interfering with any individual or habitat within that population (Lacki et al. 2011). Over the past 10—12 years, there has been an increase in research on bats in the United States (U.S.) because of White-nose Syndrome (WNS), a fungal disease affecting many cave hibernating bat species (Puechmaille et al. 2011; Turner et al. 2015). The causative agent of WNS is *Pseudogymnoascus destructans*, a psychrophilic fungus first discovered in New York in 2006 (Blehert et al. 2009). Since the discovery of the fungus, it has spread to 36 states and 7 Canadian provinces, threatening over half of all North American bat species with severe population declines (Frick et al. 2010; Micalizzi et al. 2017; Whitenosesyndrome.org 2018). The risk posed by WNS has prompted an increase in research, surveys, management planning, and overall monitoring of bat species in the U.S.

Bat population monitoring is usually conducted using three main methods: Physical capture, cave surveys, and acoustic monitoring. Previously, physical capture was the most common population monitoring method employed (Berry et

al. 2004; O'Farrell and Gannon 1999) but recent advances in bioacoustics technologies and monitoring techniques have allowed for the development of improved monitoring methods that allow for the study of aerial bat communities across different spatial scales and habitats (Bergeon et al. 2014; Estrada 2015; Lintott et al. 2013). Depending on the type of data required, populations can be physically sampled using mist netting or harp trapping. These methods allow biologists to handle captured individuals and obtain a variety of morphometric measurements and samples. Mist nets are most frequently used to estimate species presence and relative abundance, but some studies show these types of surveys can be biased (Penderson et al. 2007). Over 20% of bats are able to avoid mist nets altogether, skewing capture rates and making it harder to determine if a species is present in a given area (Robbins et al. 2008). Bats are also able to learn the location of nets to avoid flying into them. This avoidance, known as "net shyness," has the greatest effect between the first and second night of sampling, reducing capture rates by 47%. Capture rates continue to decline over subsequent sampling nights (Marques et al. 2013). These reductions in capture rates have been observed even when there is no reduction in activity (Robbins et al. 2008). Many of the biases associated with mist netting also apply to harp trapping, although "net shyness" is less common with harp trapping as bats are less able to detect the wires of the harp traps with their echolocation calls (Berry et al. 2004) and generally harp traps capture greater

numbers of species and individuals when compared to mist nets (Hourigan et al. 2008).

Bats also have the ability to change call frequencies depending on the type of prey they are foraging for and the amount of clutter (physical obstructions to flight and foraging including foliage, branches, and stems; Cox et al. 2016) in an area, changing calls by several kilohertz to avoid echo returns from objects other than the targeted prey (Arlettaz et al. 2001; Dicecco et al. 2013). These changes in call frequency, especially with higher frequency calls, can help bats detect nets/traps (Berry et al. 2004). Because species that use higher frequency calls can detect nets easier than species using lower frequency calls, fewer species using high frequency calls are physically captured (Lawrence et al. 1982; Kopsinis et al. 2004). This ability to detect nets can cause a bias in species captured, which results in estimated species abundance and presence being skewed towards bats that use lower frequency calls (Dicecco et al. 2013; Berry et al. 2004; Kopsinis et al. 2010).

Because of the amount of labor, equipment, and permitting involved, and the possibility for bias, many bat surveys now use passive acoustic methods (i.e. deployment of ultrasonic acoustic bat detectors) rather than physical capture to monitor populations. Acoustic methods provide a non-invasive means of determining bat species presence, richness, and relative abundance (Tonos et al. 2014) and as a result are becoming a viable alternative to physical capture (Muray et al. 1999; O'Farrell and Gannon, 1999). There has been much research

on optimizing bat population survey techniques and the biases associated with capture alone (Berry et al. 2004; Hourigan et al. 2008; Kloepper et al. 2016; Marques et al. 2013; Murray et al. 1999; O'Farrell and Gannon, 1999; Pederson et al. 2007; Robbins et al. 2008). However, less research has been conducted examining the capabilities of various acoustic bat detectors (Adams et al. 2012; Froidevaux et al. 2014). With the advent of increased sensitivity in transducers and software capabilities that allow species determination based on calls (Alonso et al. 2015; Hugel et al. 2017), the use of acoustic monitors is increasing. In a survey of bats at nine sites in Brisbane, Australia, bat detectors recorded 3682 calls from 13 species, while harp traps captured 17 individuals from 5 species (Hourigan et al. 2008). The advantages of using acoustic monitoring over mist nets and harp traps are numerous but there are still some limitations that need to be considered.

Limitations when using acoustic detectors greatly depend on echolocation call structure. Call structure parameters differ by species but include duration, harmonic structure, arrangement of constant-frequency and frequency modulated components, and amplitude (Lawrence et al. 1982). The envelope of detection (i.e., the volume in space where detectors can pick up a bat call) may change based on these differing parameters and based on the amount of attenuation, reflection by vegetation and ground materials, and spreading loss affecting each call (Adams et al. 2012; Lawrence et al. 1982; Stahlschmidt and Bruhl 2012; Surlykke et al. 2008). Because most insectivorous bats use high frequency calls

to forage, the effects of atmospheric attenuation must be considered, since attenuation is frequency dependent and greater at higher frequencies (Jones et al. 2007; Lawrence et al. 1982). Because of this, bats emitting higher frequency calls will be harder to detect due to the increased call attenuation (Surlykke et al. 2008). Reflection of echolocation calls will also interfere with detection. This is especially true when recording bats over water, where intense reflections from the water's surface will cause a positive echolocation call return, affecting the source estimates of each recorded call (Surlykke et al. 2009). Scattering and absorption of echolocation calls will increase with increasing vegetation density (Parsons et al. 1996) and spreading loss should also be considered, as sound and sonar waves decrease in intensity as the waves move further away from the source (Lawrence et al. 1986; Schuchmann and Siemers 2010; Surlykke et al. 2008). Detection probability will also depend on the type of detector being used and microphone design and sensitivity. Acoustic detector design, microphone style, angle of the microphone to the source of the call, and movement of the call source around the microphone are all parameters that affect the chances of recording an echolocation call (Adams et al. 2012; Parsons et al. 1996). The ability of ground-based detectors to detect bats consistently has also been questioned because the detection range of most acoustic monitors is unknown (Barclay 1999). Finally, there are limitations identifying bat calls at the species level when using acoustic monitoring systems. Some species are easier for automated identification software to identify than others because of similarities of

calls and ambient noise amounts (Ford et al. 2005; Jennings et al. 2008). Despite these limitations, acoustic monitoring can still be used to estimate species richness and relative abundance (Kloepper et al. 2016).

With these limitations, sampling bat populations can be difficult, leading to confusion on the status or presence of many species (Whitby et al. 2014) and increasing the need for new monitoring techniques. Mobile acoustic echolocation monitoring has become a popular technique for surveying summer bat populations, with protocols modeled after European monitoring programs (Whitby et al. 2014). These mobile monitoring programs use roads and waterways to record echolocation calls of bats from a moving vehicle or boat but still miss several species of bats (Braun de Torrez et al. 2017; Whitby et al. 2014). However, both acoustic techniques, stationary and mobile, lack the capability to monitor a bat in flight. A previous acoustic population study of bats at various altitudes showed that bat activity levels were almost 3 times greater above a forest canopy (>30m) when compared to activity levels in the same areas at 2 and 10m above ground level (Menzel et al. 2012). Bats are very active at altitudes above ground level, but it is difficult to record high-flying bats because of the inherent signal attenuation of higher-frequency calls at higher altitudes and being incapable of placing detectors at required altitudes (Gillam et al. 2009). With data showing greater bat activity at altitudes above tree canopy levels, using an aerial-based echolocation detector may be beneficial to enhance population survey techniques, allowing detectors to be placed at varying altitudes

and record bats in flight. Because of the limitations associated with ground-based acoustic detection and evidence of greater bat activity at altitudes above the canopy, an Aerial Bat Detection Technology (ABDT) was designed to record echolocation calls of bats in flight.

The objectives of this study are focused on addressing some of the limitations of acoustic detectors and include:

- Determining the capabilities of a new novel ABDT by conducting bat population surveys at four different military installations across the southeastern United States.
- Testing the recording capabilities of two commonly used full-spectrum bat detectors by building a frequency emitter box that can emit signals similar to bat calls at varying frequencies and intensities.

By completing these objectives, we will be able to compare acoustic data gathered from detectors at various altitudes to data collected by a ground-based detector and begin to determine the detection distance for certain bat detectors.

CHAPTER I
DETERMINING THE EFFECTIVENESS OF AN AERIAL ACOUSTIC
BAT DETECTION TECHNOLOGY

Abstract

Ground-based ultrasonic acoustic bat detectors may be of limited use for monitoring an airborne species. Limitations, such as unknown detection distance and cone of detection, can affect probability of species detection, which can in turn affect determination of species presence, relative abundance, and occupancy. To overcome these limitations, an improved monitoring method is needed that can survey bats in flight and provide more accurate data for population analysis. We used an Aerial Bat Detection Technology (ABDT) to monitor bats in flight at varying altitudes (25, 50, 75, and 100m). Acoustic recordings of bat calls were analyzed for species identification using SonoBat Automated Bat Call Identification software V4.0.6. Calls collected by the ABDT were compared to calls collected from a ground-based acoustic bat detector run simultaneously throughout the collection periods. Out of the 44 sampling nights, the ABDT recorded species missed by the ground-based detector on 20 nights and ground-based detector recorded species missed by the ABDT on 3 of the 44 nights. Almost all species that were missed on the ground-based detector were recorded at >50m on the ABDT. There was no difference in the total number of calls recorded by the two methods ($P= 0.1223$, $\alpha= 0.05$) however, the ground-based detector recorded more calls per hour (calls/hr) when the ABDT was flown at the 50 – 100m levels ($P= 0.017$, $P= 0.001$ and $P= 0.005$, respectively). This suggests that using ground-based monitoring methods alone to examine population dynamics of bats may lead to an incomplete sample of species richness and relative abundance.

Introduction

Acoustic monitoring of bat populations has been extensively used for many years and provides a non-invasive, cost-effective method for collecting large amounts of data on bat species presence and relative abundance (Barclay 1999; Adams et al. 2012; Blejwas et al. 2014; Froidevaux et al. 2014). With continually improving technologies, researchers can now analyze echolocation call dynamics and have the ability to “hear” what a bat hears when echolocating, which provides the knowledge needed to improve the capabilities of acoustic detectors (Jones et al. 2007). Understanding population trends in any species is important for conservation purposes but monitoring population trends is especially important for bats due to the threats many of the species face from habitat loss and disease (Bergeson et al. 2014). When monitoring bat species that are endangered or threatened, a comprehensive examination of species presence is needed, as failing to detect or capture one species can have severe consequences.

Recent technological advances have resulted in increased bat detector sensitivity and recording rate of ground-based acoustic detectors (Adams et al. 2012; Froidevaux et al. 2014). Even with recent advances, issues still arise when conducting acoustic monitoring, such as misidentification of bat calls and unknown detection distance of most detectors. Misidentification of bat calls has led to the research and development of more sophisticated software for

automated call identification (Clement et al. 2014) in hopes of focusing conservation efforts in areas where populations are in peril. Each species of bat exhibits a wide range of calls and call structures, which are highly dependent on the ecological niche filled by that particular species and on the purpose of the call (Fenton et al. 2004; Jones et al. 2007). Calls are optimized depending on activity, such as foraging, navigation, or communication (Stahlschmidt et al. 2012). Foraging calls change depending on the environment in which the bat is foraging, such as an open field or a cluttered forest (Lacki et al. 2007; Hügél et al. 2017). However, even with increased detector sensitivity and better call identification, monitoring a flying species over a large spatial scale still limits the use of acoustic detectors for population monitoring. Monitoring a flying mammal with very directional echolocation calls (Jacobsen et al. 2012) from a stationary ground point greatly limits the area from which calls may be recorded.

With misidentification of bat calls being reduced through technological improvements and calls being species specific (Stahlschmidt et al. 2012), identification of bat populations in an area is possible using a single acoustic detector (Fenton et al. 2004; Lacki et al. 2007; Surlykke et al. 2008). With this specificity in species calls and the number of calls a single bat produces, there is a high likelihood of detecting multiple species of bats in a single night with a single acoustic detector (Adams et al. 2012). The problem with inconsistencies in sampling methodologies and the high spatial and temporal variability in a bat's activity level (Bergeson et al. 2014; Froidevaux et al. 2014) still exists and needs

to be addressed. Because the distance at which acoustic detectors can record bats is largely unknown (Hourigan et al. 2008), a detector placed at ground-level may not be recording all bats flying above its microphone, allowing for an incomplete documentation of bat species richness and biased estimates of activity levels as a measure of relative abundance. Detectors placed at altitudes above ground level (10m) and above tree canopy level (>30m) have recorded greater bat activity than those at ground level (Menzel et al. 2012). In order to address the issues of recording species with high variation in spatial activities and the unknown detection distances of acoustic detectors, we designed an Aerial Bat Detection Technology (ABDT) to record echolocation calls of bats in flight at various altitudes. The design of the ABDT was based on the Autonomous Aerial Acoustic Recording System (AAARS), which was designed to monitor threatened and endangered birds in inaccessible areas of military installations (Hockman 2018). The ABDT consists of a 300g weather balloon used to place an acoustic bat detector and data acquisition payload at various altitudes above ground level to record bats in flight. The payload contains a GPS unit for location tracking and failsafe devices to recover any lost balloons and transmit locations to a base station (Hockman 2018).

The objectives of this study were to:

1. Design and test a novel Aerial Based Detection Technology (ABDT) that could be flown at various altitudes to record bat echolocation calls; and

2. Test the capabilities of the ABDT as compared to ground-based monitoring by surveying bat populations at various locations across the southeastern United States.

Study Areas

Our study was conducted at 4 U. S. military installations across the southeastern United States: Big Oaks National Wildlife Refuge (BONWR), Indiana (Formerly Jefferson Proving Grounds, a retired Army installation); Fort Leonard Wood (FLW), Missouri; Camp Robinson (CMPR), Arkansas; and Arnold Airforce Base (AAFB), Tennessee. Three study sites were selected within each of these 4 locations for a total of 12 study sites. Each study site was chosen based on its proximity to water, where insect swarms would be greatest to attract bats for monitoring and where there would be less structural clutter for initial ABDT testing (Hügel et al. 2017).

Methods

From mid-May through mid-August 2016, depending on weather conditions and range operations, ground-based and aerial echolocation call monitoring was conducted at each installation for 5 to 6 days per month. Each installation was visited once per month and each of the 3 study sites within an installation were surveyed a minimum of 3 times per visit. This allowed for sampling at the 12 study sites at least 3 times per month, for a total of 36 sampling periods per month.

Aerial Acoustic Sampling

Aerial echolocation calls were collected by deploying the ABDT at altitudes between 25 – 100 m above ground level. Each ABDT consisted of a modified Pettersson D500X acoustic bat detector (Pettersson Elektronik AB, Sweden) attached to an electronic data acquisition payload suspended from a 300-gram, helium-filled weather balloon (Figure 2.1; Hockman 2018), all of which was tethered to the ground using 75 lb braided fishing line. The tether attached to the ABDT was used to raise and lower the entire unit during each sampling period. The ABDT's payload contained GPS tracking, a modem for communication from a ground station that controlled the helium valves and location transmission, and an automatic recovery system that was based on GPS position in case the balloon broke tether. All components inside the payload of the ABDT were the same as those used in the AAARS (Hockman 2018), with modifications made to remove a ballast dropping system and to incorporate the Pettersson acoustic bat detector for echolocation monitoring. The GPS unit within the payload monitored the ABDT altitude and horizontal location throughout the night, with information saved on a mini SD card contained within the payload. The valve attached to the balloon could be opened and closed remotely in case an emergency dump of helium was required or for venting small amounts of helium to lower the balloon. These systems were all powered by an 8-volt battery contained within the payload. Real-time altitude and horizontal location data were transmitted from the payload via a RF module to a base station laptop computer.

The computer software LabVIEW (National Instruments LabVIEW, Austin, TX) controlled the payload operation. Through LabVIEW, we could control the valves in the payload and put the balloon in emergency mode if it crashed or broke from tether.

The Pettersson D500X bat detector was modified for attachment to the payload to make it more lightweight and suitable for suspension from the balloon. All hardware was removed from the metal housing and the detector was placed into a lightweight plastic box. The microphone jack and D500x external microphone were attached to the bottom of the new housing and pointed downward towards the ground when in flight. All modified Pettersson units were tested before use to ensure consistent performance compared to unmodified units. These detectors were used to record bat calls throughout the night at varying altitudes. The detector was programmed to start recording 15 minutes before sunset and ended recording after all sampling periods were completed. The total sampling period lasted 4 hours per night and sampling altitudes were 25 m, 50 m, 75 m, and 100 m. The ADBT was deployed for 30 min at each altitude, twice per night (i.e., total of 1 hr at each altitude/night). The order in which the four altitudes were monitored was randomized each night (i.e. the 25 m altitude was not necessarily always monitored first). Calls recorded during the times when the ADBT was being raised or lowered between altitude periods were not included in analysis. Calls recorded by the ADBT were stored on compact flash (CF) cards contained within the Pettersson unit.

Ground-Based Acoustic Sampling

A ground-based Pettersson D500X bat detector was placed under the tethered ABDT, with the detector's microphone placed on a 3-m pole at a 45° angle (Armitage and Ober 2012). GPS Coordinates for placement of the ground-based detector were recorded at the beginning of the study at each sampling site so that the detector could be placed in the same location during all subsequent sampling periods. Ground-based detectors were programmed to begin recording at the same time as the ABDT, 15 min before sunset, and ended recording when all recording periods for ABDT were completed. All call recorded on the detector were also stored on CF cards.

Two different sites were sampled each night whenever possible, with one ABDT and one ground-based detector at each site. Time of sunset, site name, the number of the detector and payload used, wind conditions, start and stop times for each altitude interval and any abnormalities were recorded each night for each sampling location. The ABDT was returned to the ground once sampling at all altitude intervals was completed.

Data Analysis

All calls recorded by the ABDT and ground-based detector were batch processed through SonoBat (Szewczak 2010) to identify species and saved as Excel spreadsheets. Species were identified with >90% accuracy, according to SonoBat's call analysis software. Forty-four of the 62 sampling nights were

retained for comparison, with 18 sampling nights being removed from analysis for technical issues, weather issues, or because range operations did not allow for sampling to take place. Data was tested for normality and a two-sampled t-test was conducted in Program R statistical software (R Development Core Team 2008) to examine differences in the number of calls and species recorded by the ABDT versus the ground-based detector. Sampling units were nights of successful simultaneous recording using both the ABDT and the ground-based detector (N= 44). To conduct the t-tests on the nightly total number of species and calls recorded by the ABDT compared to the ground-based detector, the total successful sampling nights (N= 44) were divided by successful sampling nights at each location for BONWR (n= 11), FLW (n= 12), CMPR (n= 10) and AAFB (n= 11). .

The data for each night was broken down further to compare the number of calls collected by the ABDT at each altitude interval to calls collected by the ground-based detector during the corresponding hour and two-tailed t-tests were used to test for differences. We also compared the number of high and low frequency calls recorded at each altitude interval to the number of high and low frequency calls recorded by the ground-based detector. Bat calls were classified as high frequency if they were identified by SonoBat as a *Myotis* species, *Lasiurus borealis* (LABO), *Nycticeius humeralis* (NYHU), or *Perimyotis subflavus* (PESU). Calls were classified as low frequency if they were identified as

Eptesicus fuscus (EPFU), *Lasionycteris noctivagans* (LANO), *Corynorhinus rafinesquii* (CORA) and *Lasiurus cinereus* (LACI; Cox et al. 2016).

Results

There were 44 successful nights of sampling from mid-May through mid-August 2016. For the entire sampling period (n= 44), a total of 2,490 calls were recorded by the ABDT, with a mean of 57.4 calls recorded per night. A total of 3,842 calls were recorded by the ground-based detector, with a mean of 87.3 calls recorded per night (Table 1.1). The total number of calls recorded over the sampling period decreased dramatically as the ABDT increased in altitude until the 100m altitude, when the number of calls recorded increased slightly (Figure 1.1). There was no difference in the total number of calls collected by the ABDT and the ground-based detector during the entire sampling period ($P= 0.1223$).

The total number of high and low frequency calls recorded by each method did not differ ($P= 0.075$) with the mean number of high and low frequency calls being 19.6 and 24.5, respectively. Of the 2490 calls recorded by the ABDT, 1984 were identified with >90% accuracy. Of these 1984 calls, 1102 (55.6%) were from low frequency species (EPFU, LANO, CORA, and LACI) and 882 (44.5%) were from high frequency species (*Myotis*, LABO, NYHU, and PESU). The number of high and low frequency calls recorded each hour by each method did not vary greatly, only changing when a species was recorded by the ABDT but missed by the ground-based detector and vice versa. Of the calls missed by

the ADBT that were identified to the species level, 21 were classified as high frequency calls and 25 of them were classified as low frequency calls. After separating the total calls into calls/hr, there was no difference in the ability of either method to record low or high frequency calls. There were some differences in the number of calls/hr at the various altitudes. The two-tailed t-tests showed no difference in the number of hourly calls for the ADBT at 25 m vs ground-based detector but the number of calls recorded were significantly lesser for the ADBT compared to the ground-based detector at greater altitudes (Table 1.7).

The total number of species recorded by the ADBT did not differ compared to the number recorded by the ground-based detectors ($P= 0.6756$). The average number of species recorded by the ADBT at each location varied as did the average number recorded by the ground-based detector (Figures 1.2 – 1.5). The average number of species recorded by the ADBT per night at each location was greater at CMPR and BONWR but less than the average number recorded by the ground-based detector at FLW and AAFB (Figure 1.6).

Overall, the ground-based detector recorded 10 species and the ADBT recorded 8 species (Table 1.1). Of the 44 total sampling nights, there were 20 nights where the ADBT recorded the same species as the ground-based detector, plus additional species missed by the ground-based detector (Tables 1.3 – 1.6). There were 11 days where different species were detected by the ground-based detector compared to the ADBT (i.e., the ground-based detector

recorded species missed by the ABDT and the ABDT recorded some missed by the ground-based detector). There were 10 days where the ABDT and the ground-based detector recorded the same species. There were 3 days where the ground-based detector performed better than the ABDT (i.e., recorded the same species as the ABDT plus additional species missed by the ABDT).

Discussion

This study serves as the first systematic test of a novel aerial acoustic bat detector's capabilities in recording bat calls at different altitudes. It is also the first to compare the capabilities of an aerial acoustic bat detector with standard ground-based acoustic monitoring. Because the technology was new, additional evaluation is needed to develop an optimized aerial monitoring system. However, the results still provide compelling evidence that demonstrates the value of positioning acoustic detectors aloft when implementing a comprehensive bat monitoring strategy.

The ABDT provides a means to collect bat calls at altitudes unattainable when using only ground-based acoustic detectors. With a strong difference already present in an acoustic detector's ability to record bat calls based on frequency, distance and angle of the call (Adams et al. 2012), recording bat calls aurally will help provide a more complete species inventory. Most of the species that were missed on the ground-based detector were recorded at 50 m or higher, which may be out of the range of detection for a ground-based acoustic detector.

During multiple sampling days, several species of bats were recorded by the ADBT but not by the ground-based detector and some were recorded by the ground-based detector but not the ADBT with many of these species recorded at the 50 m altitude and above. Even though the ADBT recorded more calls at the 25 m altitude, most of the species missed by the ground-based detector were recorded at the 50 m and 75 m altitude. Some species such as *Myotis grisescens* (MYGR) were only recorded at 50 m and above, when the ground-based detector failed to record them, suggesting that some bats are not foraging low enough to be detected, or captured, using traditional ground-based methods. Of the 44 sampling nights, there were 3 nights where the ground-based detector recorded species that the ADBT missed, with these 3 nights having wind speeds > 10.5 kph at ground level. Because of the increased ground-level wind speed, there may have been less bat activity since bats are known to forage less during adverse weather conditions (Dina et al. 2017). It is possible that the increased wind speed could have interfered with the recording capabilities of the detector (swaying of the ADBT on tether). There were 6 days (12%) not included in the 44-night analysis where ambient noise such as insect activity prevented the ground-based detector from recording echolocation calls clear enough to be identified using automated software.

Call frequency (i.e., high or low) did not seem to affect the performance of either the ADBT or the ground-based detector. Overall, there was only a slightly greater number of low-frequency calls recorded than high-frequency calls, which

could be attributed to either detector capabilities or species composition in the sampling area. After separating the total calls into calls-per-hour, there was no difference in the ability of either method to record low or high frequency calls. The number of calls recorded per hour by the ADBT at the 25-m interval when compared to the data from the ground-based detector during the corresponding hour was very similar. There were decreases in detections per hour when the ADBT was at 50 – 100 m when compared to detections by the ground-based detector for the corresponding hours. This can possibly be attributed to greater bat activity at lower altitudes. While this data shows that a stationary ground-based detector will record more calls than a detector placed at greater altitudes, a more complete species richness may be obtained with a combination of both methods.

The ADBT, while providing valuable echolocation data, still has some limitations for bat monitoring. The device requires constant monitoring, unlike ground-based detectors that can be left unattended in the field, someone must always be present when using the ADBT. Any kind of adverse weather, such as winds, rain or mist, and atmospheric thermal inversions, will prevent the use of the ADBT or make it difficult to operate at a consistent altitude. The amount of helium used, and transportation of helium tanks must also be considered when using the ADBT. There is also the possibility of a balloon breaking or falling due various environmental hazards, atmospheric conditions such as thermal inversions or poor manufacturing, causing a loss of time and possibly data

depending on the altitude and location of the ABDT. Also, with only one microphone on the unit, it is impossible to determine the exact altitude at which a bat was recorded, only altitude estimates can be obtained. These limitations should be considered before beginning surveys.

When using an aerial method for echolocation monitoring, a targeted altitude approach may be advantageous when monitoring for a single species. The altitude at which bats are recorded depends greatly on foraging activity of each species, making a varying altitude sampling regime somewhat detrimental for a targeted species approach. However, if the goal is to determine the species richness of an area, using an aerial method such as the ABDT at varying altitudes throughout the sampling period will be more successful. With foraging area size dependent on sex and species (Lacki et al. 2011), finding a targeted sampling location and altitude would be beneficial for a single-species study.

With many acoustic sampling plans, it is common practice to place more than one detector in a sampling area to increase the probability of detecting more species. However, this approach only increases the area sampled horizontally. When studying a volant species, it is important to know the volume of airspace being sampled and increase that sample area vertically whenever possible (Adams et al. 2012; Corben and Fellers 2001; Fenton 2000). Increasing the detection distance vertically is becoming important, especially where bat mortalities are being caused by above-ground structures, such as wind turbines. Many studies are using acoustic monitoring to sample not only areas where

turbines are already present but also in areas proposed for wind turbine construction (Kunz et al. 2007). If ground-based methods alone are used, an incomplete profile of bat species richness and activity would be used in decisions regarding turbine siting. Poor siting decisions could have disastrous implications for some species which have already seen extremely high mortalities due to WNS. Based on our results, implementing an aerial sampling component in any bat monitoring program would provide a non-invasive means to gather large quantities of data and obtain a better inventory of species richness in a given area.

Literature Cited

Adams, A., Jantzen, M., Hamilton, R., and Fenton, M. (2012). Do you hear what I hear? Implications of detector selection for acoustic monitoring of bats. *Methods in Ecology and Evolution*, 3(6), 992-998.

Arduino – ArduinoBoardUno. (2017) Arduino – HomePage, Arduino.cc/en/Main/ArduinoBoardUno.

Raphaël Arlettaz, Gareth Jones, & Paul A. Racey. (2001). Effect of acoustic clutter on prey detection by bats. *Nature*, 414(6865), 742-745.

Blejwas, K., Lausen, C., and Rhea-Fournier, D. (2014). Acoustic monitoring provides first records of Hoary Bats (*Lasiurus cinereus*) and delineates the distribution of silver-haired bats (*Lasionycteris noctivagans*) in Southeast Alaska. *95*(3), 236-250.

Braun de Torrez, E., Wallrichs, M., Ober, H., and McCleery, R. (2017). Mobile acoustic transects miss rare bat species: Implications of survey method and spatio-temporal sampling for monitoring bats. *PeerJ*, 5(11), E3940.

Corben, C. and Fellers, G. (2001) Choosing the 'correct' bat detector – a reply. *Acta Chiropterologica*, 3, 245–256.

Cox, M., Willcox, E., Keyser, P., & Vander Yacht, A. (2016). Bat response to prescribed fire and overstory thinning in hardwood forest on the Cumberland Plateau, Tennessee. *Forest Ecology and Management*, 359(C), 221-231. Dicecco, J., Gaudette, J., and Simmons, J. (2013). Multi-component separation and analysis of bat echolocation calls. *The Journal of the Acoustical Society of America*, 133(1), 538-46.

Dina, K., Dechmann, N., Wilkelski, D., Ellis-Soto, K., Safi, M., and O'Mara, T. 2017. Determinants of spring migration departure decision in a bat. *Biol. Lett.* Published 20 September 2017.

Estrada Villegas, S., Meyer, C., McGill, B., and Kalko, E., (2015). "Assessing the structure of a Neotropical bat community using acoustic monitoring techniques." *The Journal of the Acoustical Society of America* 138(3): 1905-1905.

Fenton, M.B. (2000) Choosing the 'correct' bat detector. *Acta Chiropterologica*, 2, 215–224.

Frick, W., Pollock, J., Hicks, A., Langwig, K., Reynolds, D., Turner, G., Kunz, T. (2010). An emerging disease causes regional population collapse of a common North American bat species. *Science (New York, N.Y.)*, 329(5992), 679-82.

Froidevaux, J. S. P., Zellweger, F., Bollmann, K., and Obrist, M. (2014). Optimizing passive acoustic sampling of bats in forests." *Ecology and Evolution* 4(24): 4690-4700.

Gillam, E., McCracken, H., Westbrook, G., Lee, F., Jensen, J., & Balsley, K. (2009). Bats aloft: Variability in echolocation call structure at high altitudes. *Behavioral Ecology and Sociobiology*, 64(1), 69-79.

Hockman, E. (2018) Acoustic monitoring of wildlife in inaccessible areas and automatic detection of bird songs from continuous recordings. Dissertation. The University of Tennessee, Knoxville.

Hügel, T., Meir, V., Muñoz-Meneses, A., Clarin, B., Siemers, B., and Goerlitz, M. (2017). Does similarity in call structure or foraging ecology explain interspecific information transfer in wild *Myotis* bats? *Behavioral Ecology and Sociobiology*, 71(11), 1-16.

Jones, G. W., and Holderied, M. (2007). Bat echolocation calls: Adaptation and convergent evolution. *Proceedings of the Royal Society B: Biological Sciences*, 274(1612), 905-912.

Kopsinis, Y., Aboutanios, E., Waters, D., and Mclaughlin, S. (2010). Time-frequency and advanced frequency estimation techniques for the investigation of bat echolocation calls. *The Journal of the Acoustical Society of America*, 127(2), 1124-34.

Kunz, T., Arnett, A., Erickson, W., Hoar, A., Johnson, G., Larkin, R., Strickland, M., Thresher, R. and Tuttle, M. (2007) Ecological impacts of wind energy developments on bats: questions, research needs, and hypotheses. *Frontiers in Ecology and the Environment*, 5, 315–324

Lacki M, Hayes J, and Kurta A. (2007). *Bats in Forests*. The Johns Hopkins University Press, Baltimore, MD, USA, Pp. 84-85.

Lawrence, B., and Simmons, J. (1982). Measurements of atmospheric attenuation at ultrasonic frequencies and the significance for echolocation by bats. *The Journal of the Acoustical Society of America*, 71(3), 585-90.

Marques, J., Ramos, P., Maria, J., Marques, T., Santos, C., Santana, J., Beja, P., and Palmeirim, J. (2013). "Optimizing sampling design to deal with mist-net avoidance in Amazonian birds and bats." *PloS one* 8(9): e74505.

Menzel, J., Menzel, M., Kilgo, J., Ford, W., Edwards, J., and McCracken., G. (2005) Effect of habitat and foraging altitude on bat activity in the costal plain of South Carolina. *The Journal of Wildlife Management*. 69: 235-245

Parsons, S. (1996). "A comparison of the performance of a brand of broad-band and several brands of narrow-band bat detectors in two different habitat types." *Bioacoustics* 7(1): 33-43.

O'Farrell, M., and Gannon, W. (1999). A comparison of acoustic vs capture techniques for the inventory of bats. *Brief Article*). *Journal of Mammalogy*, 80(1), 24-30.

Puechmaille, S., Frick, W., Kunz, T., Racey, P., Voigt, C., Wibbelt, G., and Teeling, E. (2011). White-nose syndrome: Is this emerging disease a threat to European bats? *Trends in Ecology & Evolution*, 26(11), 570-576.

R Development Core Team. (2008). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0

Schuchmann, M., Siemers, B., and Somers, M. (2010). Variability in Echolocation Call Intensity in a Community of Horseshoe Bats: A Role for Resource Partitioning or Communication? (*Echolocation Call Intensities*). *PLoS ONE*, 5(9), E12842.

Stahlschmidt, P. and Brühl C. (2012). "Bats as bioindicators – the need of a standardized method for acoustic bat activity surveys." *Methods in Ecology and Evolution* 3(3): 503-508.

Surlykke, A., Kalko, E., and Giurfa, M. (2008). Echolocating Bats Cry Out Loud to Detect Their Prey (High Intensity Bat Calls). *PLoS ONE*, 3(4), E2036.

Szewczak, J.M. (2010) SonoBat v.4.0.6 www.sonobat.com

Turner, J., Warnecks, L., Wilcox, A., Baloun, D., Bollinger, T., Misra, V., and Willis, C. (2015). "Conspecific disturbance contributes to altered hibernation patterns in bats with white-nose syndrome." *Physiology & Behavior* 140: 71-78.

U.S. Fish and Wildlife Service. (2016) "Environmental Conservation Online System." Species Search Results, U.S. Fish and Wildlife Service.

Whitby, M., Carter, T., Britzke, E., and Bergeson, S. (2014). Evaluation of mobile acoustic techniques for bat population monitoring. *Acta Chiropterologica*, 16(1), 223-230.

White-Nose Syndrome. *About White-Nose Syndrome*. 1 June (2018).
www.whitenosesyndrome.org/resources/map

Wood, J. (2010) Marine Mammal Species Conservation: A Review of Developments in the Uses of Acoustics, *Journal of International Wildlife Law & Policy*, 13:4, 311-325

Appendix

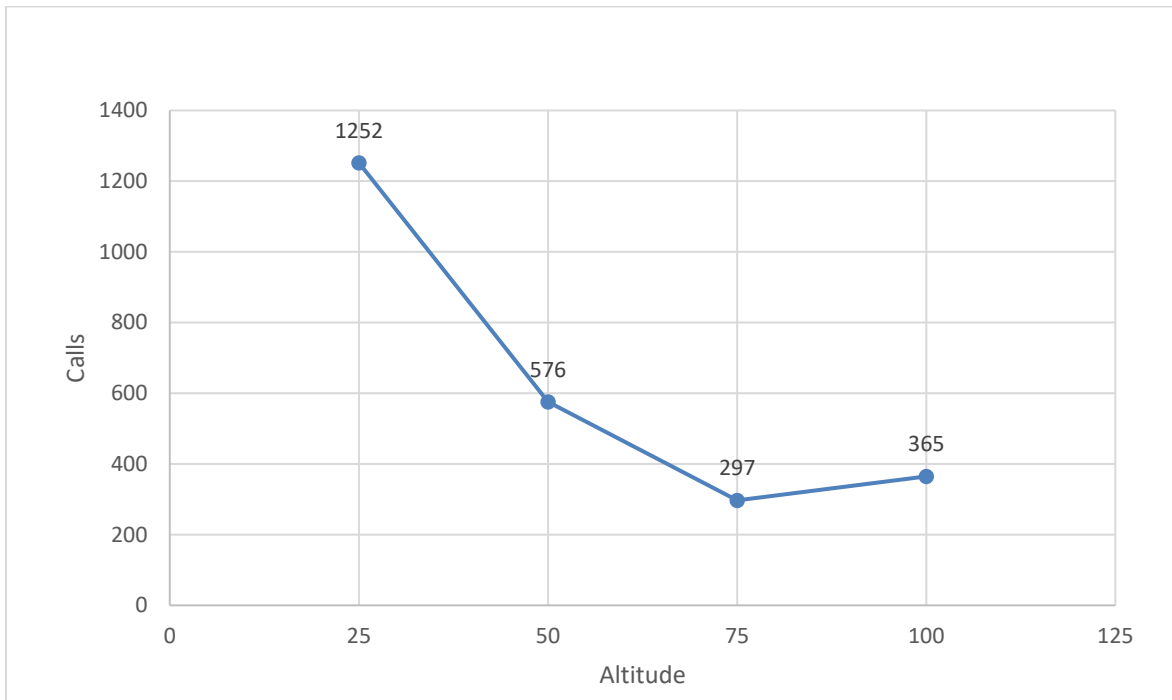


Figure 1.1 Total bat echolocation calls recorded by an Aerial Bat Detection Technology flown at 4 altitudes above ground level (25m, 50m, 75m and 100m) at Big Oaks National Wildlife Refuge, IN, Fort Leonard Wood, MO, Camp Robinson, AR, and Arnold Airforce Base, TN, summer 2016.

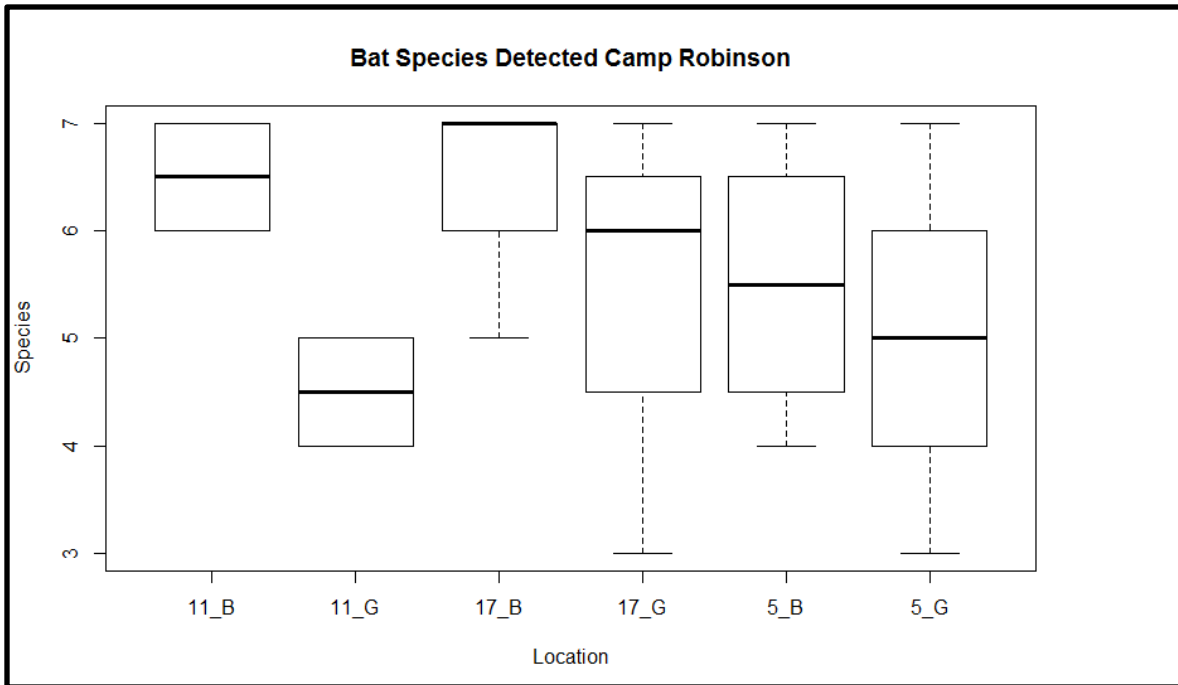


Figure 1.2 Number of bat species detected at Camp Robinson, AR by a ground-based acoustic detector (G) and the Aerial Bat Detection Technology (B) at each of three locations (sites 11, 17, and 5), summer 2016.

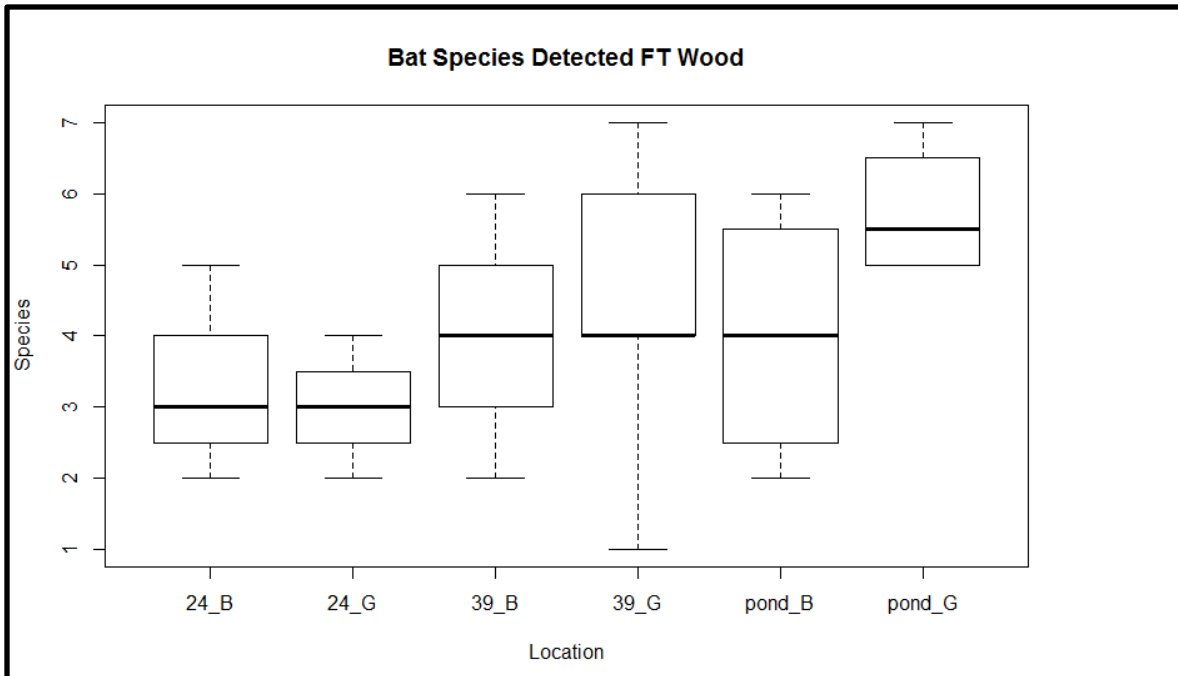


Figure 1.3 Number of bat species detected at Fort Leonard Wood, MO by a ground-based acoustic detector (G) and the Aerial Bat Detection Technology (B) at each of the three locations (sites 24, 39, and McCaan Pond), summer 2016.

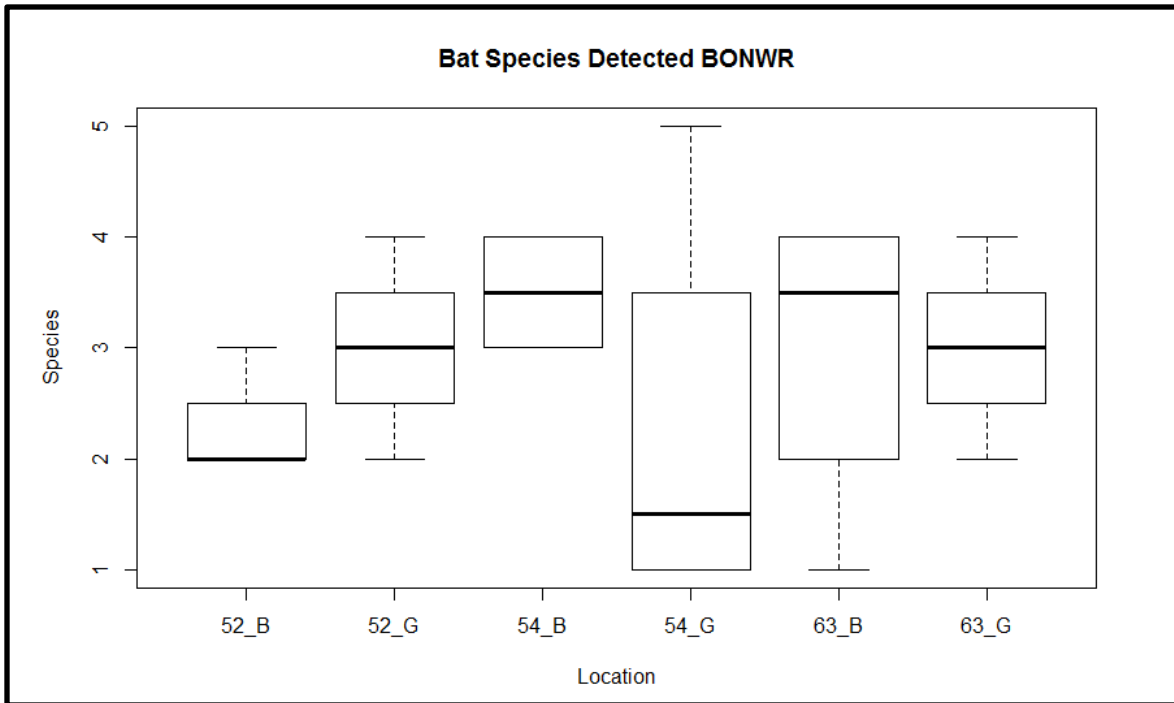


Figure 1.4 Number of bat species detected at Big Oaks National Wildlife Refuge, IN, by a ground-based acoustic detector (G) and the Aerial Bat Detection Technology (B) at each of three locations (sites 52, 54, and 63), summer of 2016.

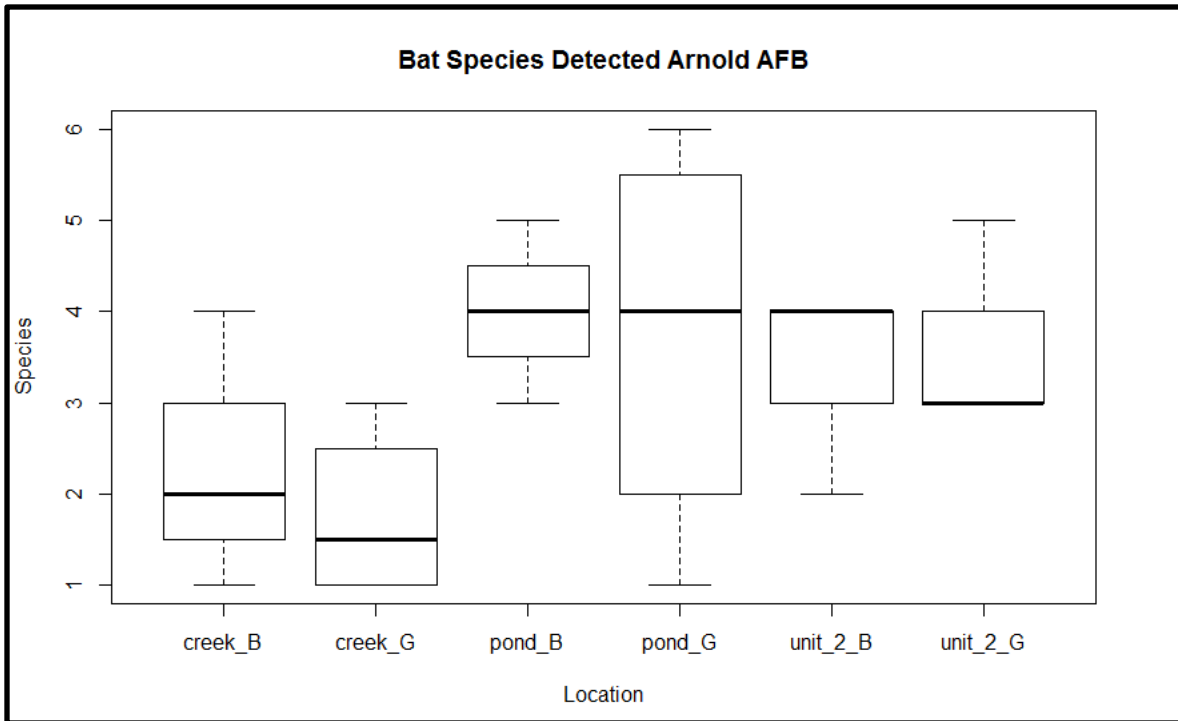


Figure 1.5 Number of bat species detected at Arnold Airforce Base, TN, by a ground-based acoustic detector (G) and the Aerial Bat Detection Technology B) at each of three locations (sites Huckleberry Creek, Sinking Pond, and Unit 2), summer 2016.

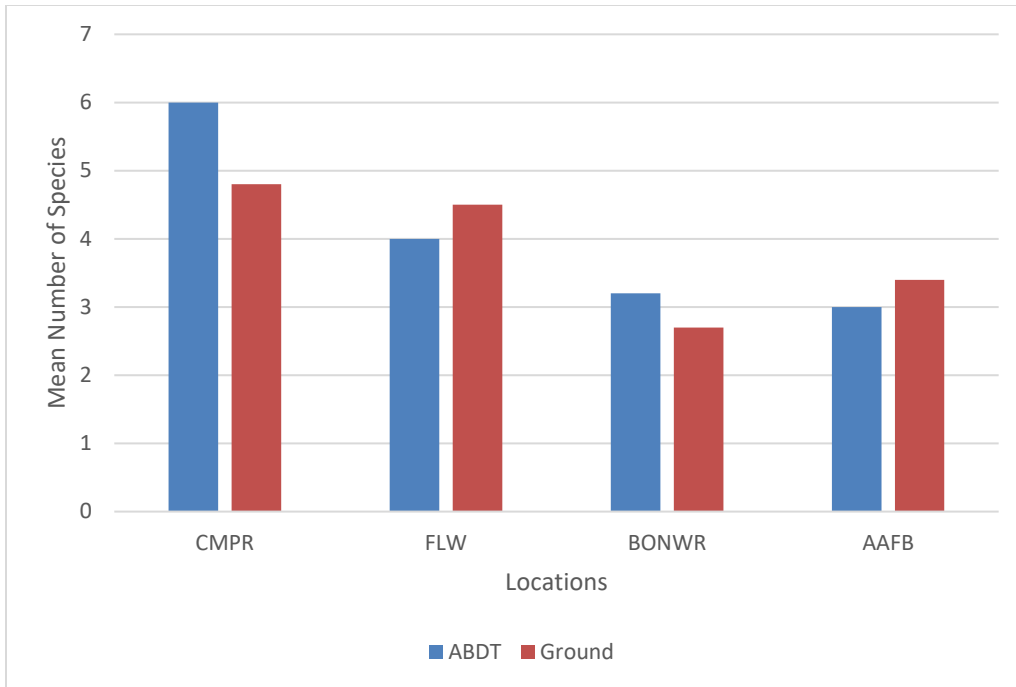


Figure 1.6 Mean number of bat species recorded per night by an Aerial Bat Detection Technology (ABDT) and a ground-based acoustic detector (Ground) at Big Oaks National Wildlife Refuge, IN, Fort Leonard Wood, MO, Camp Robinson, AR, and Arnold Airforce Base, TN, summer 2016.

Table 1.1 Total number of calls recorded at Big Oaks National Wildlife Refuge, IN, Fort Leonard Wood, MO, Camp Robinson, AR, and Arnold Airforce Base, TN by an Aerial Bat Detection Technology (ABDT) and ground-based acoustic detector (Ground), summer 2016.

LOCATION	TOTAL	MEAN
BONWR		
ABDT	398	36.181
GROUND	536	48.727
FLW		
ABDT	599	49.917
GROUND	1382	91.7
CMPR		
ABDT	1245	65.88
GROUND	1461	73.21
AAFB		
ABDT	248	28.4
GROUND	463	46.3

Table 1.2 Bat species recorded at Big Oaks National Wildlife Refuge, IN, Fort Leonard Wood, MO, Camp Robinson, AR, and Arnold Airforce Base, TN by an Aerial Bat Detection Technology (ABDT) and ground-based acoustic detector (Ground), summer 2016.

Species	
Ground	ABDT
<i>Eptesicus fuscus</i>	<i>Eptesicus fuscus</i>
<i>Lasiurus borealis</i>	<i>Lasiurus borealis</i>
<i>Lasiurus cinereus</i>	<i>Lasiurus cinereus</i>
<i>Lasionycteris noctivagans</i>	<i>Lasionycteris noctivagans</i>
<i>Myotis grisescens</i>	<i>Myotis grisescens</i>
<i>Myotis leibii</i>	None
<i>Myotis sodalis</i>	None
<i>Nycticeius humeralis</i>	<i>Nycticeius humeralis</i>
<i>Perimyotis subflavus</i>	<i>Perimyotis subflavus</i>
<i>Corynorhinus rafinesquii</i>	<i>Corynorhinus rafinesquii</i>

Table 1.3 Species recorded by an Aerial Bat Detection Technology that were not recorded by a ground-based acoustic detector at Camp Robinson, AR, summer 2016.

DATE	SAMPLE AREA	SPECIES ¹	ALTITUDE (M)
13-JUN-16	TA11	MYGR	50
13-JUN-16	TA11	LANO	100
14-JUN-16	TA17	MYGR	100 and 75
13-JUL-16	TA17	LACI	75
13-JUL-16	TA17	NYHU	75
16-JUL-16	TA11	LACI	25,50, and 75
16-JUL-16	TA11	LANO	75
11-AUG-16	TA5	LANO	100
12-AUG-16	TA17	LANO	50
12-AUG-16	TA17	CORA	50 and 75
12-AUG-16	TA5	EPFU	25, 50, and 75

¹MYGR= *Myotis grisescens*, EPFU= *Eptesicus fuscus*, LACI= *Lasiurus cinereus*, NYHU= *Nycticeius humeralis*, LANO= *Lasionycteris noctivagans*, CORA= *Corynorhinus rafinesquii*

Table 1.4 Species recorded by an Aerial Bat Detection Technology that were not recorded by a ground-based acoustic detector at Fort Wood, summer 2016.

DATE	SAMPLE AREA	SPECIES¹	ALTITUDE (M)
8-JUN-16	Range 24	MYGR	75
8-JUN-16	Range 24	PESU	50
8-JUN-16	Range 24	NYHU	75
2-AUG-16	Range 39	CORA	50
2-AUG-16	Range 39	LANO	50
3-AUG-16	Range 24	NYHU	75
6-AUG-16	McCaan Pond	LANO	100

¹MYGR= *Myotis grisescens*, NYHU= *Nycticeius humeralis*, LANO= *Lasionycteris noctivagans*, CORA= *Corynorhinus rafinesquii*, PESU= *Perimyotis subflavus*

Table 1.5 Species recorded an Aerial Bat Detection Technology that were not recorded by a ground-based acoustic detector at Big Oaks National Wildlife Refuge (BONWR), IN, summer 2016.

DATE	SAMPLE AREA	SPECIES¹	ALTITUDE (M)
25-JUN-16	TA 63	PESU	100 and 25
25-JUN-16	TA 63	PESU	75
26-JUN-16	TA 63	LANO	75
27-JUN-16	TA 52	LANO	75
27-JUN-16	TA 54	LANO	75
27-JUN-16	TA 54	LACI	75 and 25
30-JUL-16	TA 52	LABO	50
31-JUL-16	TA 63	LABO	25
31-JUL-16	TA 63	LABO	100

¹LANO= *Lasionycteris noctivagans*, PESU= *Perimyotis subflavus*, LABO= *Lasiurus borealis*, LACI= *Lasiurus cinereus*

Table 1.6 Species recorded by an Aerial Bat Detection Technology that were not recorded by a ground-based acoustic detector at Arnold Airforce Base (AAFB), TN, summer 2016.

DATE	SAMPLE AREA	SPECIES¹	ALTITUDE (M)
16-JUN-16	Sinking Pond	PESU	100
21-JUL-16	Unit 2	LABO	50 and 25
21-JUL-16	Unit 2	LACI	75
22-JUL-16	Unit 2	LANO	50 and 25
22-JUL-16	Unit 2	CORA	100
17-AUG-16	Sinking Pond	LABO	50
17-AUG-16	Sinking Pond	NYHU	50
17-AUG-16	Sinking Pond	PESU	50
19-AUG-16	Huckleberry Creek	LABO	50

¹NYHU= *Nycticeius humeralis*, LANO= *Lasionycteris noctivagans*, CORA= *Corynorhinus rafinesquii*, PESU= *Perimyotis subflavus*, LACI= *Lasiurus cinereus*, LABO= *Lasiurus borealis*

Table 1.7 Comparison of the number of bat calls recorded per hour by an Aerial Bat Detection Technology (ABDT) and ground-based acoustic detector (Ground) at Big Oaks National Wildlife Refuge, IN, Fort Leonard Wood, MO, Camp Robinson, AR, and Arnold Airforce Base, TN, summer 2016.

Mean Number of Calls			
Altitude	ABDT	Ground	<i>P</i>
25m	27.8	25.8	0.697
50m	12.6	22.0	0.017
75m	6.6	22.3	0.001
100m	8.1	18.0	0.005

CHAPTER II
EVALUATING ACOUSTIC BAT DETECTOR PERFORMANCE

Abstract

Acoustic detection of bats has become an integral part of many monitoring programs. Detection capabilities have been greatly improved through technological advances, making this an effective way to gather large amounts of data on population dynamics quickly and easily. However, acoustic bat detectors are constrained by certain unknown limitations, such as signal detection distance. Using an acoustic detector with an unknown limit of detection can lead to population surveys that miss key species, especially in areas where threatened and endangered bats may be present. In order to determine the detection distance of Pettersson D500x (Pettersson Elektronik AB, Sweden) and the SM2+ (Wildlife Acoustics Inc., Massachusetts, US) ultrasonic bat detectors, an ultrasonic signal emitter was designed and built to transmit signals at a constant frequency and decibel level to test the signal recording limits for these two detectors. It was found that a signal emitted at 20kHz (approximately 89.92 dB) was not recorded by the Pettersson D500x or the SM2+ at a distance further than 17m and 22m respectively. This preliminary test for two acoustic detectors shows some of the capabilities when used to record very low frequency calls.

Introduction

Acoustic detection is a common monitoring technique used to survey many different species, allowing large quantities of data to be quickly gathered with minimal manpower (Adams et al. 2012; Wood 2010). By sampling environments acoustically, researchers can gather presence and absence data for use in small or large-scale biodiversity monitoring and conservation planning with very little effort (Froidevaux et al. 2014; Stathopoulos et al. 2017). Acoustic sampling is an effective and common method for monitoring bat populations, the second largest order of mammals, of which many species in the United States have been listed as threatened or endangered (Adams et al. 2012; Stathopoulos et al. 2017; US Fish and Wildlife Service 2016). Echolocation calls, which bats use for navigation, orientation and foraging, can be recorded and used to determine the presence or absence of different species and determine relative abundance, making the acoustic monitoring of bat populations a powerful conservation tool (Kloepper et al. 2016; Froidevaux et al. 2014; Adams et al. 2012; Stathopoulos et al. 2017; Surlykke et al. 2008). Acoustic detection has become an integral part of many bat monitoring programs around the world, with new monitoring protocols using echolocation detection being implemented in many states in the US (Stathopoulos et al. 2017; Froidevaux et al. 2014; Whitby et al. 2014; Surlykke et al. 2008)

With bats being an essential part of many ecosystems and providing many ecosystem services (Alonso et al. 2015; Kunz et al. 2011), monitoring

populations is an important part of any land management plan (Blejwas et al. 2014). Surveying efforts across the Southeastern United States has increased over the past decade due to mortalities caused by WNS and anthropogenic activity that reduces habitat (Stathopoulos et al. 2017; Hoyt et al. 2016; Lorch et al. 2011).

Because bats are an important part of many ecosystems and in light of WNS, there has been an increase in monitoring efforts. Better monitoring techniques and equipment have been designed to achieve greater monitoring success, along with a heavier reliance on acoustic detection methods (Adams et al. 2012; Hourigan et al. 2008; Alonso et al. 2006). Acoustic bat detectors are deployed in the field to record bat echolocation calls that are then used to determine species presence, richness, and relative abundance, but the hardware used can affect the quality of data collected and call variation makes species determination difficult (Adams et al. 2012). Detecting certain bat calls is already a challenge due to environmental conditions and the nature of sonar waves, along with the effects of atmospheric attenuation on echolocation calls (Whitby et al. 2014; Kopsinis et al. 2010; Surlykke et al. 2009; Lawrence et al. 1982). Due to greater attenuation, bat calls on the higher end of their frequency range with lower intensity have a smaller chance of detection by acoustic monitors than calls of lower frequency with high intensity, which can lead to an underestimation of species presence for bats using high-frequency calls (Murray et al. 2007; Lawrence et al. 1982). A bat foraging in a cluttered area, such as a forest, will

emit high frequency, lower intensity calls, which are harder to record (Lacki et al. 2011; Lawrence et al. 1982), limiting the ability to detect bats in forested areas when using acoustic methods alone. A bat call is also highly directional and will become even more directional with higher frequencies (Lacki et al. 2011; Surlykke et al. 2009). The more directional the call, the harder it is to record because the call must be emitted by the bat when aimed almost directly at the microphone in order to be recorded (Surlykke et al. 2009). These limitations in call detection capabilities are the main factors in determining detection distance (the distance at which an echolocation call can be recorded by an acoustic detector and still be identified) of acoustic detectors. Because acoustic monitoring programs are a large part of almost every bat population survey, detection distance of bat detectors given environmental influences and call strength need to be determined.

The objective of this study was to determine vertical and horizontal detection distance of two commonly used full spectrum acoustic bat detectors, the Pettersson D500x (the Pettersson) and the SM2+ made by Wildlife Acoustics. To determine the detection distance, an ultrasonic signal emitter was built that could emit a signal at frequencies similar to bat calls and used to test the detection distance. By determining a detection distance for these detectors, wildlife managers can better implement the use of acoustic detectors for bat monitoring plans.

Methods

We designed a bat call emitter to test the Pettersson D500x acoustic bat detector (Pettersson Elektronik) and the SM2+ (Wildlife Acoustics). The emitter did not mimic an echolocation call but transmitted a signal of similar length at various frequencies. The emitter consisted of a 15.24 cm wide by 27.94 cm long metal box that was 10.16 cm deep. It contained one Arduino Uno, an open-source electronic platform board able to read code inputs and turn it into activation outputs (Arduino 2017), one signal amplifier, MAX9744 20W Class D amplifier (Adafruit Industries, New York, U.S.), and one Peerless XT25SC90 1inch dual ring radiator speaker (Madisound, Wisconsin, U.S) mounted to the surface of the housing. These components are powered by 8 AA batteries. Signals, or “calls,” can be emitted by the box at frequencies ranging from 0 – 100 kHz, but for our testing, we chose to program the box to emit calls at 20, 25, 45, and 65 kHz, which are common frequencies used by bats in the eastern U.S. (Adams et al. 2012). Laboratory tests were conducted to determine the decibel level achieved by the emitter using a Martel C-322 Sound Pressure Level Meter/Data Logger (Reed Instruments, North Carolina, U.S.). From these lab tests, we determined that the bat call box was capable of emitting signals at sound pressure levels of approximately 89.92 dB at 20kHz when measured 1m from the emitter (Table 2.1).

To calculate the dBSPL for each signal, an initial dBSPL (L_1 ; Table 2.1) was determined for frequencies from 6 kHz-65 kHz using the Martel C-322

Sound Pressure Level Meter to measure the signal strength at 1m distance (r_1) from the source. As the emitter was moved away from the detector the new distance (r_2) was placed in the equation (Figure 2.2), and the absolute value of 20 times the log of the initial distance divided by the new distance was used to determine the new dBSPL (L_2) (Sengpiel, 2012).

The emitter and detectors were placed in a field location with minimal ambient noise to test detection distance. The emitter was secured to a tripod and faced the detector, which was also secured onto a tripod, and each was sighted using distance scopes to ensure signal would reach the microphone on the detector directly, without any angles or interference. The emitter was placed 1 meter (m) from the detector and programmed to emit a constant signal for 2 min at 20 kHz (approximately 89.92 dB). The time of each trial was recorded and after the 2 min period was completed, the emitter was moved 1 m away from the detector and the process repeated. This process was repeated until the emitter was 25m from the detector. The same process was repeated to determine vertical detection distance by taking the emitter up a fire lookout tower until it was 25m from the detector, at 1 m intervals. Sample period times and weather conditions, including temperature, wind speed and relative humidity were recorded for each sample. Once the trials were completed, the data collected was processed through SonoBat Automated Bat Call Identification Software (SonoBat Inc., Arcata, California), which showed which calls were recorded, the times they were recorded, and the frequency of the recorded call. The times

recorded in the field were compared to the timestamp obtained through the SonoBat output to verify which signals were recorded and at what frequency.

Results

The signal emitter was capable of reaching almost 90 dB at 1 m with a 20kHz signal, but because of the nature of these calls, decibel levels would decrease as distance from the source increased at a rate of -6 dB per distance doubling (the distance between the signal emitter and detector is doubled; Sengpiel 2012). Estimates of decibel sound pressure level (dBSPL) were determined for each of the four signals used to test the detectors as distance from the detector increased (Figure 2.1).

Detection distance was maximized at lower frequencies, with the lowest frequency tested being 20kHz with an approximate decibel level of 89.92 dB. Detection distance for signals emitted at 20kHz were 17m for the Pettersson and 23m for the SM2+, with the ability of each detector to record signals decreasing greatly as frequency increased (Figure 2.3). The highest frequency tested was 65kHz, which had a very low detection distance with the maximum being 1m for the Pettersson and 3m for the SM2+.

Maximum vertical detection distance was the same as maximum horizontal detection distance. The maximum vertical detection distance for a signal emitted at 20kHz for the Pettersson was 17m and 23m for the SM2+. For

signals emitted at 25kHz, the maximum detection distance was 17m for the Pettersson and 20m for the SM2+. For signals emitted at 45kHz, the maximum detection distance was 2m for the Pettersson and 4m for the SM2+. For signals emitted at 65kHz, the maximum detection distance was 1m for the Pettersson and 3m for the SM2+.

Discussion

These results did not determine absolute detection distance of the two acoustic bat detectors due to the inability of the emitter to reach calls of similar sound pressure levels as bat calls. Each detector shows very limited capabilities to detect low frequency calls at short distances, with the greatest influence to successfully being able to record a call being frequency and distance from the microphone. Understanding the capabilities of tools used in bat detection is vital when designing and implementing monitoring programs. Different acoustic monitors, microphone types, and microphone placement all add to the ability of a detector to record a bat call (Adams et al. 2012; Parsons et al. 1996). Along with hardware capabilities, these results show that the detection of bat calls is very dependent on frequency and sound pressure level. The sound pressure level estimates were calculated in a laboratory environment with constant atmospheric conditions but because these levels are related to temperature and humidity, they can change depending on the conditions at the time of detection. High frequency, low intensity calls are harder to record due to the narrow beam of

waves emitted, the low sound pressure level, and due to some of the effects of attenuation on higher frequencies (Lawrence et al. 1982). Because the sound pressure level of most bat calls is mostly unknown and varies depending on the species, sex, and type of call (Adams et al. 2012; Jakobsen et al. 2013; Alonso et al. 2015), call intensity of bat calls remains to be a missing piece of information needed to improve detection capabilities.

The results from our trials were very similar for both acoustic detectors. The microphones used with each detector are built using the same transducer, however the microphone design affects the detection capabilities. The Pettersson microphone contains a transducer surrounded by a cone to help record clearer echolocation calls and weed out ambient noise. The SM2+ microphone contains the same transducer covered in a windscreen to prevent air flow from interfering with recordings.

With bats calls ranging in frequency from 11kHz (The Spotted Bat, *Euderma maculatum*) to 212 kHz (Percival's Trident Bat, *Cloeotis percivali*), detectors need to be capable of recording this same range in order to avoid sampling error (Jones et al. 2007). Because these results suggest that frequency is a significant factor in detection success, bats using higher frequency calls may be under-represented in acoustic surveys. Because low frequency calls with short wavelengths are not able to return the echolocation call for bats to target their prey (Jones et al. 2007), foraging bats usually resort to higher frequency calls, which are greatly affected by atmospheric attenuation which are harder for

detectors to record due to the nature of high frequency signals (Dicecco et al. 2013; Parsons et al. 1999 Lawrence et al. 1982). However, because our tests were conducted in a very short time span with very little change to atmospheric conditions, and because sound pressure levels are related to temperature and humidity, the full effects of attenuation cannot be determined by this experiment alone. The altitude at which a call is emitted and the frequency of the call affect attenuation rates, with calls placed closer to ground levels being more attenuated and lower frequency calls carrying more distance than higher frequency calls (Marten et al. 1977). Further studies under a varying range of conditions will give more information on attenuation rates. Until a complete understanding of bat call intensities, directionality, and attenuation can be determined, multiple strategies should be implemented to fully sample bat populations in any ecosystem.

Literature Cited

Adams, A., Jantzen, M., Hamilton, R., and Fenton, M. (2012). Do you hear what I hear? Implications of detector selection for acoustic monitoring of bats. *Methods in Ecology and Evolution*, 3(6), 992-998.

Arduino – ArduinoBoardUno. (2017) Arduino – HomePage, Arduino.cc/en/Main/ArduinoBoardUno.

Blejwas, K., Lausen, C., and Rhea-Fournier, D. (2014). Acoustic monitoring provides first records of Hoary Bats (*Lasiurus cinereus*) and delineates the distribution of silver-haired bats (*Lasionycteris noctivagans*) in Southeast Alaska. *95*(3), 236-250.

Dicecco, J., Gaudette, J., and Simmons, J. (2013). Multi-component separation and analysis of bat echolocation calls. *The Journal of the Acoustical Society of America*, 133(1), 538-46.

Froidevaux, J., Zellweger, F., Bollmann, K., and Obrist, M. (2014). "Optimizing passive acoustic sampling of bats in forests." *Ecology and Evolution* 4(24): 4690-4700.

Hoyt, J., Langwig, K., Sun, K., Lu, G., Parise, K., Jiang, T., Frick, W., Foster J., Feng, J., and Kilpatrick, A. (2016). Host persistence or extinction from emerging infectious disease: Insights from white-nose syndrome in endemic and invading regions. *Proceedings. Biological Sciences*, 283(1826), 20152861.

Jakobsen, L., Brinkløy, S., and Surlykke, A. 2013. Intensity and directionality of bat echolocation signals. *Frontiers in Physiology*, 4, 89.

Jones, G. W., and Holderied, M. (2007). Bat echolocation calls: Adaptation and convergent evolution. *Proceedings of the Royal Society B: Biological Sciences*, 274(1612), 905-912.

Kopsinis, Y., Aboutanios, E., Waters, D., and Mclaughlin, S. (2010). Time-frequency and advanced frequency estimation techniques for the investigation of bat echolocation calls. *The Journal of the Acoustical Society of America*, 127(2), 1124-34.

Kunz, T., Arnett, A., Erickson, W., Hoar, A., Johnson, G., Larkin, R., Strickland, M., Thresher, R. and Tuttle, M. (2007) Ecological impacts of wind energy developments on bats: questions, research needs, and hypotheses. *Frontiers in Ecology and the Environment*, 5, 315–324

Lacki, M., Hayes, J., and Kurta, A. 2007. Bats in Forests. The Johns Hopkins University Press, Baltimore, MD, USA, Pp. 84-85.

Lawrence, B., and Simmons, J. (1982). Measurements of atmospheric attenuation at ultrasonic frequencies and the significance for echolocation by bats. *The Journal of the Acoustical Society of America*, 71(3), 585-90.

Marten, K., & Marler, P. (1977). Sound transmission and its significance for animal vocalization. *Behavioral Ecology and Sociobiology*, 2(3), 271-290.

Murray, K.L., Fraser, E., Davy, C., Fleming, T.H. and Fenton, M.B. (2007) Characterization of the echolocation calls of bats from Exuma, Bahamas. *Acta Chiropterologica*, 11, 415–424.

Murray, K. L., E. R. Britzke, B. M. Hadley, and L. W. Robbins. (1999). Surveying bat communities: a comparison between mist nets and the Anabat II bat detector system. *Acta Chiropterologica* 1:105–112.

Puechmaille, Frick, Kunz, Racey, Voigt, Wibbelt, and Teeling. (2011). White-nose syndrome: Is this emerging disease a threat to European bats? *Trends in Ecology & Evolution*, 26(11), 570-576.

Sengpiel, Eberhard. (2012). Dampening of Sound: Decibel vs Distance. Sound Level Distance Damping Decibel Calculator - Sengpielaudio Sengpiel Berlin, 2012, www.sengpielaudio.com/calculator-distance.htm.

Surlykke, A., Kalko, E., & Giurfa, M. (2008). Echolocating Bats Cry Out Loud to Detect Their Prey (High Intensity Bat Calls). *PLoS ONE*, 3(4), E2036.

Whitby, M., Carter, T., Britzke, E., and Bergeson, S. (2014). Evaluation of mobile acoustic techniques for bat population monitoring. *Acta Chiropterologica*, 16(1), 223-230.

Wood, J., (2010) Marine Mammal Species Conservation: A Review of Developments in the Uses of Acoustics, *Journal of International Wildlife Law & Policy*, 13:4, 311-325

Appendix

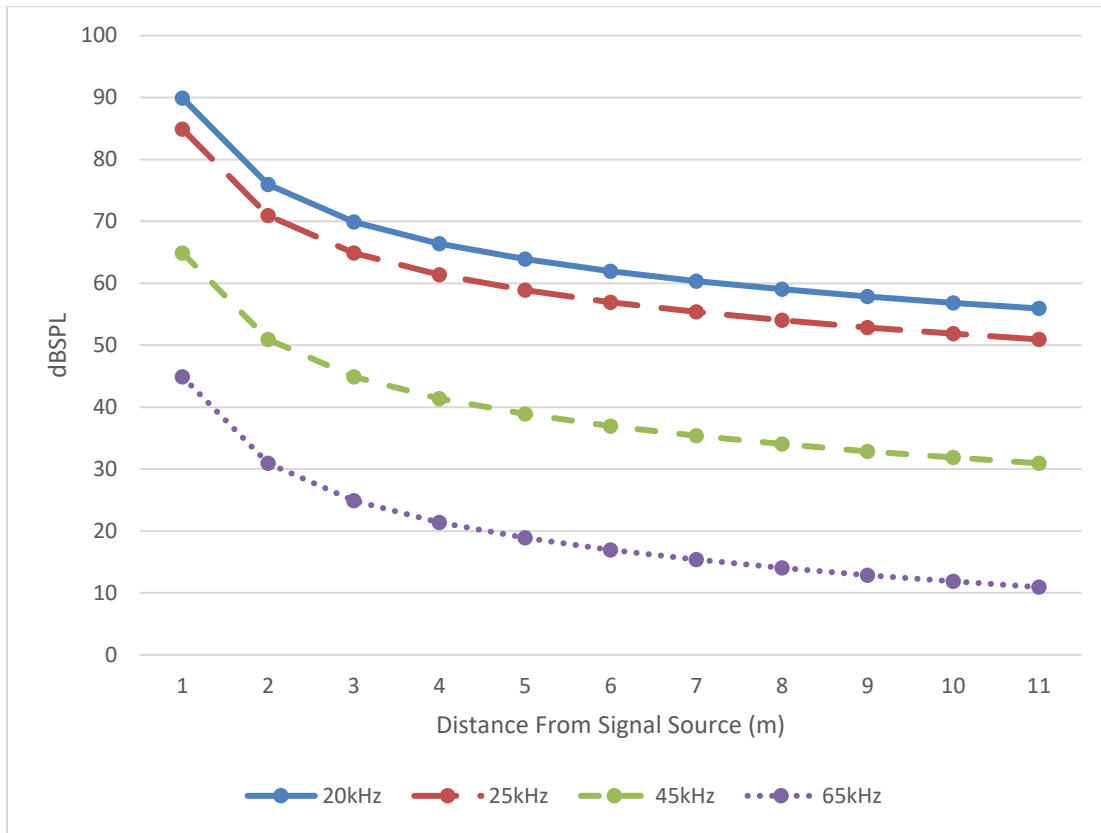


Figure 2.1 Estimated decibel sound pressure levels (dBSPL) of signals emitted by the Bat Call Emitter at 20, 25, 45, and 65kHz as distance from the signal source increases.

Sound level L and Distance r

$$L_2 = L_1 - \left| 20 \cdot \log \left(\frac{r_1}{r_2} \right) \right| \quad L_2 = L_1 - \left| 10 \cdot \log \left(\frac{r_1}{r_2} \right)^2 \right|$$
$$r_2 = r_1 \cdot 10^{\left(\frac{|L_1 - L_2|}{20} \right)} \quad r_1 = \frac{r_2}{10^{\left(\frac{|L_1 - L_2|}{20} \right)}}$$

Figure 2.2 Equation used to determine decibel sound pressure levels (dBSPL) of signals emitted by the bat call emitter where L_1 is the initial dBSPL measurement at 1m (r_1), r_2 is the new distance from the source signal, and L_2 is the dBSPL of the signal at the new distance (Sengpiel, 2012).

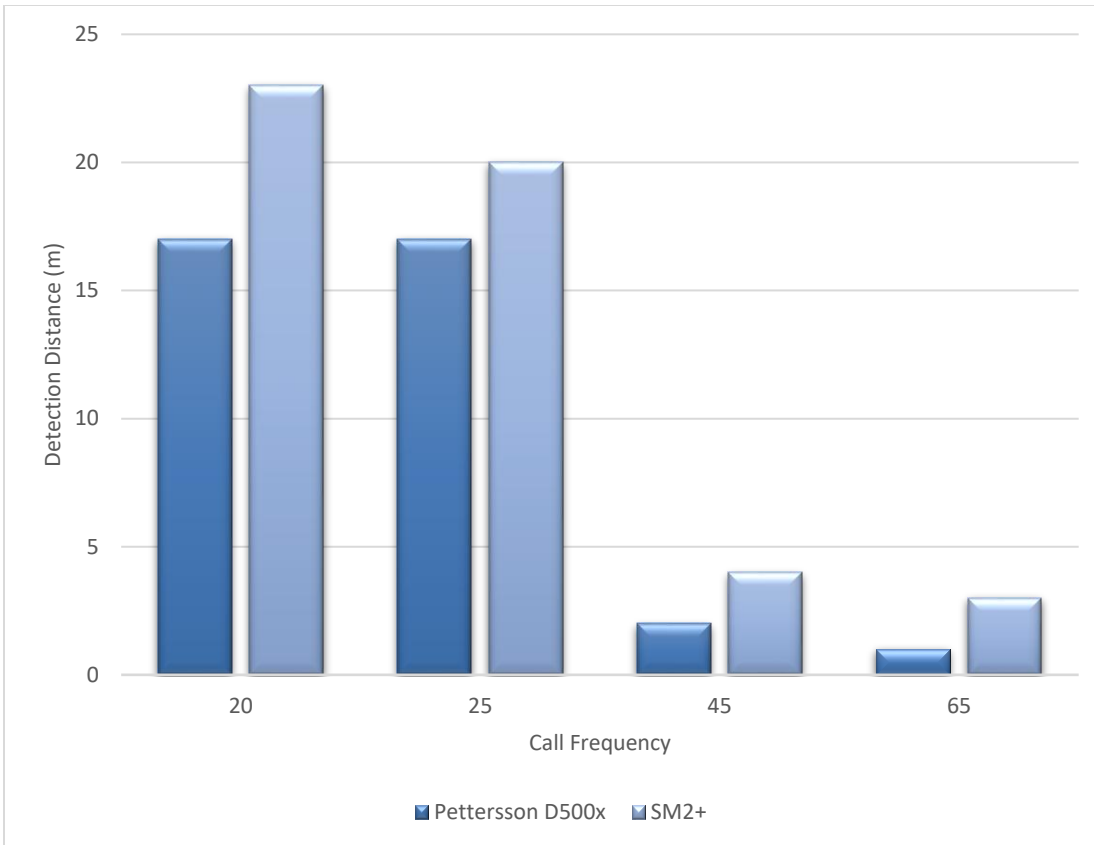


Figure 2.3 Detection distance (m) for a Pettersson D500x (Pettersson Elektronik AB, Sweden) and SM2+ (Wildlife Acoustics Inc., Massachusetts, US) acoustic bat detectors for signals emitted by the bat call emitter at 20kHz (89.92dB at 1m), 25kHz (84.92dB at 1m), 45kHz (64.92dB at 1m), and 65kHz (44.92dB) tested during July 2017 at Chuck Swan Wildlife Management Area, TN.

Table 2.1 Estimated sound pressure levels (dBSPL) of signals emitted by a bat call emitter at 1 m with corresponding frequencies.

Frequency (Hz)	Sound Pressure Level (dBSPL)
6000	102.2
9000	102
12000	99.3
15000	95.2
18000	90.5
20000	89.92
25000	84.92
30000	79.92
35000	74.92
40000	69.92
45000	64.92
50000	59.92
55000	54.92
60000	49.92
65000	44.92

CONCLUSION

This research was used to test a new method of acoustic bat detection using aerial means as well as serving as preliminary testing of two commonly used full-spectrum acoustic bat detectors to begin to better understand their detection capabilities. The ABDT provided a means to record bats in flight and at various altitudes and showed some usefulness in obtaining a more complete estimate of species richness. The tethered ABDT when run in conjunction with a ground-based detector could survey a larger column of space vertically than a ground-based detector alone. Even though the ground-based detectors were able to record more calls/hr than the ABDT while it was flown between 50 – 100m, most species missed by ground-based detectors were recorded on the ABDT when it was located at 50 – 100m, suggesting that the ground-based detectors were unable to record calls at these altitudes.

Because the detection distance of acoustic bat detectors remains largely unknown, a bat call emitter was built that had the capabilities of emitting signals at constant frequencies between 8 – 65 kHz and 102.2 - 44.92 dB at 1m, respectively. The Pettersson D50x and SM2+ acoustic bat detectors were tested in a field setting using signal emitted at 20, 25, 45, and 65 kHz, with a signal intensity starting at approximately 89.92 dB and decreasing as frequency and distance increased. The detection distance for both detectors was <20m but may

be greater with a stronger signal. These baseline measurements could help determine the area that is being sampled around acoustic detectors.

VITA

Adrienne Dykstra is from Monterey, Tennessee. She attended Daniel I Academy for her K-12 education and after her graduation, she attended Tennessee Technological University for 3 years before joining the Navy. During her 5-year military service, she worked as an intelligence collector and analyst at Fort Meade, Maryland and attended the University of Maryland College Park to continue her work in Wildlife Ecology. After her honorable discharge from the Navy, she attended the University of Nevada, Reno where she graduated with a Bachelor's degree in Wildlife Ecology and Conservation. She accepted the graduate position at the University of Tennessee Knoxville under Dr. Emma Willcox and Dr. David Buehler to obtain her Master's degree in Forestry, Wildlife and Fisheries Science.