DISSERTATION

INVESTIGATING THE EFFECTS OF NOISE POLLUTION FROM ENERGY DEVELOPMENT ON THE BAT COMMUNITY IN THE PICEANCE BASIN

Submitted by

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ABSTRACT

INVESTIGATING THE EFFECTS OF NOISE POLLUTION FROM ENERGY DEVELOPMENT ON THE BAT COMMUNITY IN THE PICEANCE BASIN

Throughout the United States, and globally, there has been recent interest in large-scale monitoring of bats, driven largely by the many threats that bats currently face such as climate change, white nose syndrome, habitat loss, and wind energy development. Additionally, many human activities generate sensory disturbances including anthropogenic light and noise pollution that have been shown to affect habitat use and foraging efficiency in bats and other wildlife. My research took place in the Piceance Basin of northwestern Colorado, where there has been considerable development of natural gas resources in recent years. During the drilling phase for natural gas, drill rigs run continuously for weeks to months at each well development site. In addition to the physical disturbance and increased human presence at the well pad, drill rigs are brightly lit, and also emit high amplitude anthropogenic noise. The light and noise from active drill rigs can travel many miles from the source, far beyond areas where wildlife habitat has been physically disturbed. The goal of my research was to isolate noise from the other associated forms of disturbance, and investigate what, if any, effects drilling noise is having on the bat community.

My dissertation is comprised of three stand-alone chapters, as follows. In Chapter 1, I test two different strategies for deploying bat detectors to determine which strategy yields higher detection and species identification rates. In Chapters 2 and 3, I present the results of noise playback experiments. Chapter 2 compares bat activity levels at control sites, and at treatment

sites where noise was added experimentally. In Chapter 3, I monitor bat activity patterns both during and after a noise playback experiment to determine how rapidly activity levels recover post-exposure. A brief summary of each chapter follows.

In Chapter 1, I focus on the methods for recording bat echolocation calls, and identifying free-flying bats in the field. Unlike capture techniques, current acoustical methods for bat monitoring do not provide information about unique individuals, age, sex, or reproductive status. What acoustical monitoring can provide is information about bat activity levels, habitat use, and species identification in some cases, without interfering with bat movement, foraging, or other activities. The commercially available technology for recording bat echolocation calls has rapidly advanced, and there are many ultrasound detector–recorder systems (hereafter, 'bat detectors') available with a wide range of recoding options. Due to rapid attenuation of ultrasound signals, one of the challenges to acoustical monitoring is the relatively limited recording range of bat detectors. To increase this range, I took advantage of a bat detector that had the ability to record on two channels (in stereo). By attaching microphone extension cables, I was able to increase the distance between the left and right channel microphones, thereby increasing the acoustical sampling space. When this data collection effort took place, the SM2BAT+ detector from Wildlife Acoustics, Inc. was the only commercially available bat detector that had the two-channel recording capability. I deployed two identical bat detectors at each study site, and compared the recordings made using the stereo option to recordings made from a single channel.

In general, the stereo setup outperformed the single-channel systems. With the stereo microphones separated by approximately 10 m, the bat detectors that recorded in stereo produced 2.7 times more recordings overall. The increased number of recordings resulted in a higher number of calls that could be identified to species. The benefit of the stereo setup was not equal for all species. With the stereo microphones only about 10 m apart, there was some overlap between the calls that were identified on the left and right channels. The highest rate of overlap (19.5%) was in big brown bats (*Eptesicus fuscus*). Rates of overlap for species in the *Myotis* genus were all less than 5%, and none of the recordings of pallid bats (*Antrozous pallidus*) were identified on both channels for the same bat-pass-event. The stereo option is a promising way to increase the number of bat recordings, which may be a particularly useful when surveying for rare species.

In Chapter 2, I used a noise playback experiment to isolate noise from other forms of anthropogenic disturbance, and monitored the bat activity level response. I recorded the sounds of an active drill rig, and played these recordings at treatment sites. I measured sound pressure levels at the drill site, and estimated the sound pressure levels at the noise playback sites. Using outdoor speakers, I was unable to project the drill rig noise at the same amplitude of an actual rig, but I was able to significantly elevate the sound levels at treatment sites. The noise levels at treatment sites roughly corresponded to noise levels that can be experienced approximately 100 m from a drill rig. This distance from a drill rig is typically beyond the well pad, in habitat that is not physically disturbed. There is widespread recognition that noise, light, and other sensory disturbances can affect the behavior and physiology of wildlife. The goal of the experiment was to determine if noise alone impacted the activity levels of bats, after being separated from the other forms of disturbance at a drilling site. I projected noise at treatment sites that were not already developed, and paired these treatment sites with control sites with no added noise. I conducted this experiment in 2013 and 2014, and present the results from 20 sites each year (10 control-treatment pairs annually).

Both years, there was an overall decrease in bat activity at treatment sites, when compared to control sites. In 2013, 8 of the 10 treatment sites had lower estimated bat activity levels. In 2014, all 10 treatment sites had lower estimated activity levels, although for some control-treatment pairs there was overlap in the credible intervals. Multiple species showed signs of reduced activity at treatment sites. For both years, *M. ciliolabrum* and *L. cinarius* had reduced activity levels at treatment sites. The response of other species was more idiosyncratic, with reduced activity in one field season, and inconclusive or no response during the other year. The species that did respond to the noise treatment have very different life histories, making it difficult to generalize about how any given species may respond to noise.

Chapter 3 focuses on bat activity level trends over time during a two period cross-over experiment. The classic two-period crossover experiment consists of two treatments (i.e., treatments 'A' and 'B'), where each site is exposed to both treatments, and the order of the treatments is randomly assigned. For this study, 'A' refers to no added noise, and 'B' refers to a noise treatment consisting of the projected recording of drilling noise. A total of 12 sites were randomly assigned to the A:B sequence, and 13 sites were assigned to the B:A sequence. I acoustically monitored bat activity throughout the experiment, with particular interest in understanding the activity level dynamics post-exposure to the noise treatment. Most studies that investigate the impacts of noise on wildlife have focused on the response to noise during a noise treatment period, or noise event. Only a handful of previous studies have addressed the post-exposure period after noise ends. In these studies, noise treatments or events were relatively short in duration (a few minutes), and the corresponding recovery period was monitored over a similarly brief timeframe. My study differs in both the duration of the noise treatment (continuous noise over six days/nights), and in the duration of the post-exposure monitoring

period (also six days/nights). I focused on the response of four bat species, *Myotis ciliolabrum*, *Myotis evotis, Lasiurus cinareus*, and *Taderida brasiliensis*.

Of the four focal species, *M. evotis* showed no substantial response to the noise treatment. The responses of *M. ciliolabrum* and *T. brasiliensis* were somewhat challenging to interpret. The overall activity levels of these species were lower during the treatment period in the A:B sequence, but they also had declining activity levels throughout the pre-exposure period. The activity patterns of *L. cinareus* provided the most convincing evidence of noise avoidance. Furthermore, *L. cinareus* activity levels did not show signs of recovery after the noise treatment ended in the B:A sequence. This suggests that after the six-day post-exposure period, there were still lingering effects of noise on *L. cinareus* activity levels. This study provides evidence that the effects of noise can linger for multiple days post-exposure. Future wildlife studies that assess periods of post-exposure could contribute meaningfully to this area of research, and aid in the development of conservation and mitigation efforts.

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DEDICATION

To my parents

Nancy Stoddard Warner and Robert Waite Warner

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CHAPTER 1

ARE TWO MICS BETTER THAN ONE? COMPARING BAT DETECTOR STRATEGIES

Introduction

There are over 1100 recognized species of bats globally (Kunz and Parsons 2009), and approximately 45 species in the continental United States (O'Shea and Bogan 2003). Recent interest in large-scale monitoring efforts of bats in the US have been driven largely by the many threats that they face, including white nose syndrome, wind energy development, climate change, and habitat loss (Jones et al. 2009, Rodhouse et al. 2012, Loeb et al. 2015). While there are many creative methods for capturing and observing bats (see Hayes et al. 2009), monitoring small, highly mobile, nocturnal, cryptic species, remains difficult. Adding to the monitoring challenge, basic life history information is not well known for many bat species (O'Shea and Bogan 2003).

The majority of bats in the continental US are insectivorous, and use echolocation to navigate and locate insect prey. Since most US bat species constantly "leak information about themselves" through their echolocation calls (Walters et al. 2013), acoustical monitoring is a useful tool, especially at the landscape scale (Fischer et al. 2009, Szewczak et al. 2011a). Every technique has its limitations, but monitoring bats using acoustical methods may be preferable in cases when capture is not feasible, or to avoid disturbing the behavior of free-flying bats (Preatoni et al. 2005, Parsons and Szewczak 2009).

The fact that some bat species navigate using ultrasound signals has been known for about a century (Hahn 1908, Hartridge 1920), but the ability to identify bat species from field recordings has a much shorter history. Fenton and Bell (1981) were the first to use acoustical monitoring to identify a suite of North American bats from field recordings. Since then, the technology to record and analyze field recordings of bats has rapidly evolved, and there are now multiple commercially available bat detector options, as well as multiple software options for call analysis (see Parsons and Szewczak 2009, Britzke et al. 2013, and Loeb et al. 2015). Britzke et al. (2013) and Parsons and Szewczak (2009) discuss the benefits and limitations of several categories of bat detectors (zero crossing, time expansion, heterodyning, and direct recording), and Loeb et al. (2015) provides a list of commercially available detectors.

Acoustical monitoring for bats cannot currently identify unique individuals, but it can provide information about habitat use, activity levels, presence/absence, and in some cases, species identification (Hayes et al. 2009). Using acoustical data (often in conjunction with other data types) has been suggested as an efficient approach for assessing habitat use, and for monitoring changes in species status over large spatial scales (e.g., MacKenzie 2005, Weller 2008, Loeb et al. 2015). For example, bat monitoring programs in the Pacific Northwest and across North America are using acoustical monitoring to estimate occupancy and track changes over time (Weller 2008, Rodhouse et al. 2012, Loeb et al. 2015). Occupancy analysis requires fairly simple detection-nondetection information from repeated surveys, and can account for imperfect detection (MacKenzie et al. 2002, MacKenzie 2005). However, implementing occupancy models can still be challenging if detection histories are too sparse to generate meaningful detection probability and occupancy estimates (Weller 2008); for example, based on very limited detection histories for some species, Weller (2008) was only able to model the occupancy for 8 of the 13 bat species that were identified through both capture and acoustical methods. There are many ways to increase the number of detections; however, most of these strategies involve an increase in survey effort (number of sites or number of nights). Since equipment is often limited, I wanted to know if different deployment strategies of the same equipment could increase the number of bat encounters.

Among the commercially available detectors, Wildlife Acoustics, Inc. is the only company to currently offer a two-channel (‘stereo’) recording option (wildlifeacoustics.com). The Wildlife Acoustics Song Meter SM2BAT+ offers the option and flexibility to record either at a higher sampling rate (384 kHz) on a single channel (‘mono’), or split the effort and sample at a rate of 192 kHz on left and right ‘stereo’ channels, simultaneously. A newer model, the SM3BAT, also has this feature and allows stereo recordings at sampling rates up to 256 kHz (wildlifeacoustics.com). Higher sampling rates are especially important for ultrasound recording, because the maximum frequency that can be represented digitally is limited to ½ of the sampling rate, known as the Niquist frequency. When sampling at 192 kHz, the uppermost frequency that can be digitally recorded without aliasing the signal is 96 kHz. Several species of North American bats emit calls that include frequency components exceeding the 96 kHz limit, and sampling at 256 kHz or 384 kHz allows the entire range of those calls to be recorded (Parsons and Szewczak 2009).

In a synthetic call playback study, Adams et al. (2012) reported that at 10 meters SM2BAT detectors had less than a 0.50 probability of detecting a 55 kHz signal. In the same study, signals at 25 kHz could be detected with a 0.50 probability at up to ~20 m with the SM2BAT, using a sampling rate of 192 kHz. Ultrasound signals attenuate rapidly, and limited detection distance is one of the challenges to using ultrasound detector/recorder units in bat research and for bat monitoring (Parsons and Szewczak 2009). The detection range for all commercially available bat detectors is frequency-dependent and also affected by the strength of the signal, atmospheric conditions, directionality of the signal, and site-specific conditions (Adams et al. 2012, Santos-Moreno et al. 2013). The higher the frequency, the more rapidly a signal attenuates as it travels through the air, which disproportionately impacts the ability to detect and identify bats that have high frequency call components (Parsons and Szewczak 2009).

With the goal of maximizing the capabilities of existing technology, I set up two SM2BAT+ bat detectors per study site to test the 'mono' and 'stereo' microphone deployment strategies. I compared 1) the number of recordings generated, 2) the efficiency in terms of identified calls, storage space, and processing time, 3) the number of nights each species was identified, and 4) the probability of detection as estimated through simple occupancy simulations. I predicted that the stereo deployment strategy would yield more triggered calls, improve detection probability, and identify more species due to the increase in detection area. I did not expect to see measurable differences in the rate of identification between the two strategies, but did anticipate that the stereo strategy would require more storage space and processing time.

Methods

Study area and sites – The study occurred in Rio Blanco County, Colorado, on Bureau of Land Management lands managed by the White River Field Office from June through early August during the summers of 2013 and 2014. In this semi-arid landscape that is typically dry except during rain events, drainages with even ephemeral water sources are hotspots for wildlife. The dominant vegetation included mixed native grasses and forbes, non-native grasses, common rabbitbrush (*Chrysothamnus nauseosus*), and big sage (*Artemisia tridentata*). The nearby landscape included a mosaic of grasses and shrubs, mixed with pinyon pine (*Pinus edulis*) and juniper (*Juniperus* sp.). Due to the combination of more open grass and shrub land habitat, pinion-juniper woodlands, and abundant cliff and rock outcropping for roosting, this area supports at least 16 species of bats (Pers. comm. Heather Sauls, BLM White River Field Office, Nov. 2014). Elevation throughout the study area ranged from 1750 m to 2150 m above sea level.

A total of 13 sites were included in this study, 5 from 2013 and 8 from 2014. This resulted in a total of 102 site-nights of surveys with a range of 6 to 13 nights per site (Table 1.1). Study sites were located along dry drainages throughout the study area, to increase the chances of recording commuting and foraging bats. Drainages were identified in ArcGIS10.0, and ground-truthed prior to site set-up to ensure that they were 1) located on BLM lands, 2) in areas where no drilling or construction was taking place, and 3) had at least one vehicle access point along the drainage for equipment drop off. After a drainage was identified, the site location along the drainage was determined by generating a random point in ArcGIS.

Bat detector setup

All recordings for this study were made using Song Meter SM2BAT+ Ultrasonic Recorder units from Wildlife Acoustics, Inc., with SMX-US omnidirectional microphones (wildlifeacoustics.com). SM2BAT+ recorders have the capability to record on a single channel (‘mono’) at a sampling rate of 384 kHz, or split the sampling effort and record in stereo, with each channel (left and right) sampling at 192 kHz. I deployed two SM2BAT+ recorders at each study site; one unit recorded in mono, and the other recorded in stereo. Other than the sampling rate, the settings were identical. For the digital settings, I used an 18-dB trigger level, 16-kHz digital high pass filter, a 2.0-second window minimum, and a 5.0-second window maximum. Using the analog filter switches, I followed the Wildlife Acoustics recommended settings (Wildlife Acoustics 2011), +36-dB gain, the analog high-pass filter set to 1 kHz, and microphone bias off. The digital high pass filter defines the frequency band of interest, but does not actually filter out all lower frequencies (see Wildlife Acoustics 2011 for details). I used the triggered

.wav mode, which records sounds in .wav format directly to SD cards in the SM2BAT+ unit. Files were transferred to external hard drives for later analysis. All units were scheduled to power on at 2000 hrs (before sunset) and run until 0600 hrs (after sunrise).

For the bat detectors recording in stereo, I used external microphone extension cables to space the left and right microphones approximately 10 m apart. The left and right channels monitor the background sound level and trigger on ultrasound signals independently, however, when either channel triggers, a stereo recording from both channels is created. The mono microphone was centered between the two stereo microphones, making it possible for both bat detectors to share an equipment box that housed the recorders and their external batteries. Microphones were mounted on custom-made tripods, at a height of approximately 2.15 m (7 ft) above ground level. At this height, the microphones were above the tallest vegetation. Using an Ultrasonic Calibrator (Wildlife Acoustics) with a 40-kHz pulse, I tested the SMX-US microphones before deployment (Parsons and Szewczak 2009). I used external 6V sealed lead acid 36-amp hour batteries to power all SM2BAT+ recorders. These batteries consistently powered the detectors for at least 6 days before needing to be recharged.

No animals were handled in this study, and all research described followed the rules and guidelines of the Colorado State University institutional animal care and use committee (IACUC Protocol # 12-3574A).

Audio processing

SonoBat 3.2.1 West Suite is a commercially available software package that includes several tools for sorting and processing ultrasonic .wav files (sonobat.com). Following the examples of others (e.g., Kalcounis et al. 2013, Kennedy et al. 2014, Parkins and Clark 2015), I passed all triggered recordings through the SonoBat Batch Scrubber 5.4 before automated species identification. The SonoBat Scrubber removed low quality calls, and files that did not contain bat call sequences. I used the "high grade" setting, and included calls in the 5-20 kHz frequency range. Prior to running the stereo files through the scrubber, the files were split into separate left and right channel recordings using the 'seewave' package and custom R scripts (Sueur et al. 2008, R Core Team, 2014). The mono .wav recordings were batch scrubbed in their raw form.

After removing low quality calls with SonoBat Scubber, the remaining recordings were processed by the SonoBat 3.2.1 West Batch Classifier. The SonoBat Classifier identifies bat call sequences using two methods. As described in Clement et al. (2014), "SonoBat separately classified sequences by evaluating mean parameter values of acceptable calls within a sequence and by evaluating classification agreement among individual calls within a sequence. SonoBat estimated posterior probabilities for identified call sequences using a discriminate function model trained on the library of known calls." When both methods agree on the classification, there is a "consensus" decision.

To avoid misclassification, I followed the lead of other researchers, and only accepted the classification decisions for call sequences when SonoBat reached a "consensus" decision (e.g., Kalcounis et al. 2013, Bunkley et al. 2015). Additionally, I required inclusion of at least 4 pulses from a sequence in all identification decisions. A ≥3 pulse criterion has been used in studies where calls were manually identified (e.g., Johnson et al. 2002; Hourigan et al. 2009). The addition of the 4-pulse criterion is more conservative than using the "consensus" decision alone, and is intended to further reduce misclassification during the automated classification process. For species with fewer than 50 recordings identified to species using the automated methods, each call was manually vetted to visually identify diagnostic call features. For species with more than 50 identified recordings, a minimum of 20 call sequences were verified by manually vetting. A species is only included in the results if manual vetting confirmed species presence in the area.

Statistical analyses

Percent overlap – For all recordings that passed through the SonoBat Scrubber (recordings of bats that were not identified to species), I calculated the three different types of overlap: 1) percent overlap between the L and R stereo microphones without consideration of the mono microphone, 2) overlap between the mono microphone and both stereo microphones, and 3) overlap between the mono mic and only one (L or R) stereo microphone. To determine overlap, I compared the timestamps from the recordings at each site. At the time of deployment, I synchronized the clocks on the mono and stereo SM2BAT+ recorders, to the second. To allow for some time drift over the survey period, I included a window of ±5 seconds of time difference between the mono and stereo recorders when assessing overlap.

After processing the recordings and identifying species using the SonoBat Classifier, I tabulated the number of nights that each species was identified, and the number of sites where species were identified by each deployment strategy. I also calculated the three different types of overlap (describe above) for calls that were identified to species. Additionally, I used single-tailed Wilcoxon signed rank tests in R using package 'exactRankTests' (Hothorn et al. 2015) to test whether the average per site number of recordings identified to species from stereo systems was greater than from the paired mono systems.

Occupancy simulation – Due to interest in using occupancy methods in large-scale bat monitoring programs (e.g., Loeb et al. 2015), I wanted to investigate how occupancy and detection rate estimates might be affected by the mono and stereo deployment strategies. The occupancy status for a given site refers to whether a species is present or absent. If a species is detected at a site, we have evidence that the site is occupied; however, most methods for surveying wildlife do not result in perfect detection, so failure to detect a species does not necessarily mean the site is unoccupied. Occupancy models incorporate repeated detection-nondetection information from surveys, and jointly estimate the probability of occupancy and detection rate (MacKenzie 2002).

The simple single-season, single-species site occupancy model can be described as:

$$Z_i \sim \text{Bernoulli}(\psi) \quad \text{for } i = 1, 2, \ldots, \text{M}$$

$$Y_{ij}|Z_i \sim \text{Bernoulli}(Z_i * p) \quad \text{for j} = 1, 2, \ldots, J_i$$

Z_i is the true occupancy state for site i, where ψ is the probability of a site being occupied (the probability that $Z_i = 1$), and M is the number of sites. Y_{ij} describes the binary detection (1) or non-detection (0) observation at site i during survey j, where J_i is the number of surveys at site i. In the simplest case, p is constant over all sites and all surveys, and is the probability of detecting a species that is present (MacKenzie 2002, Fiske and Chandler 2011).

To understand how the two deployment strategies (mono vs stereo) might affect occupancy and detection rate estimates, I used a simulation approach. This approach incorporated species-specific detection histories from the field, and provided a comparison of the efficacy of the two microphone deployment strategies. Since sites were sampled in two different years, and a different set of sites were included each year, the field data did not meet the criteria to be directly included in either a single-season or multi-season occupancy model (see MacKenzie et al. 2002, MacKenzie 2006). This model also requires the assumption of closure at the species-level, which is more likely to have been met, because all sites were sampled during the non-migratory summer season, and over a relatively short time periods.

For each of the 13 field sites, I generated nightly, species-specific detection histories from the recordings of bat calls described in the previous section. To create the detection histories, I collapsed the count data to the binary detection-nondetection format needed for the occupancy models. Separate detection histories were compiled for the mono and stereo systems. Using these field-collected detection histories as the data pools, I randomly sampled the detection histories from each site, and for each deployment strategy, to generate the detection histories for a simple occupancy simulation. By randomly drawing from the field-observed encounters, I was able to incorporate realistic detection rates for each species.

I generated 10,000 new detection histories from the mono recordings and from the stereo recordings for 4 common bat species in the study area, *Myotis evotis*, *Myotis lucifugus*, *Eptesicus fuscus*, and *Lasiurus cinereus*. Each of the 10,000 generated detection histories included 13 sites with 5 nights/surveys. I used the simple single-species, single-season occupancy model (MacKenzie et al. 2002) with no covariates, implemented using the 'unmarked' package in R (Fiske and Chandler 2011, 2012). For each of the simulated detection histories, I estimated occupancy and detection rates. By sampling from the observed data, and including these simulated detection histories into a simple occupancy model, I was able to explore the effects of the deployment strategies more formally.

Results

Over all sites, the mono and stereo setups (L-and R-channel combined) generated 7,831 and 25,820 recordings, respectively. After passing through the scrubber, 7,662 and 20,802 recordings remained for the mono and stereo recordings. Considering overlap within the stereo recordings, 7946 (38.19%) were R+L pairs with the same timestamp. Of the mono recording, the majority (5395 of 7662) overlapped with both the R- and L-channel stereo recordings. An additional 1101 of mono recordings overlapped with only one (R or L) stereo channel recording, but not with both stereo mics. For 1166 of the mono recordings, neither R- nor L-channel mics recorded a bat call sequence within the 5-second time window.

Of the 7662 and 20,802 recordings that passed through the scrubbing process for the mono and stereo setups, 1682 (21.95%) and 2403 (11.55%) were identified to species, respectively. The SonoBat Classifier identified 12 different bat species in the study area: *Antrozous pallidus* (Anpa)*, Eptesicus fuscus* (Epfu)*, Lasiurus blossevillii*, *Lasiurus cinereus* (Laci)*, Lasionycteris noctivagans* (Lano)*, Myotis californicus*, *Myotis ciliolabrum* (Myci)*, Myotis evotis* (Myev)*, Myotis lucifugus* (Mylu)*, Myotis volans*, *Parastrellus Hesperus, and Tadarida brasiliensis* (Tabr). Five species (*A. pallidus*, *L. blossevillii*, *M. californicus*, *M. volans*, and *P. hesperus*) were identified fewer than 50 times. Calls from *L. blossevillii*, *M. californicus*, *M. volans*, and *P. hesperus* were inconclusive, lacking visually diagnostic call features needed to confirm their presence through manual vetting. Presence of the remaining species was verified by the manual vetting process, and only the eight species that passed the vetting process, whose abbreviations are listed above are included in the remainder of the results.

For the stereo calls that were identified to species, only 186 (7.74%) contained matching timestamps and matching species identifications between stereo channels. For the remaining 92.26% of identified stereo recordings, the species identified on one channel was not identifiable on the corresponding channel. The highest overlap rates between the left and right stereo mics occurred for *E. fuscus*, *L. cinereus*, and *T. brasilensis* (Table 1.2).

The majority of the mono recordings (62.13%) were only identified to species by the mono microphone, without corresponding identification from either stereo channel within the ±5 second time window. Both stereo mics and the mono mic identified the same species-event 8.2%

of the time (138 cases). The mono mic and only one stereo channel (L or R) identified the same species in 499 cases (29.67%).

The stereo system produced 2.7 times more recordings than the mono system, resulting in similar increases in processing time and data storage. Five species were identified at all 13 sites; of the three species that were not identified at every site, two species (*A. pallidus* and *L. noctivagans)* were each recorded at one additional site by the stereo system; and *M. lucifugus* was identified at four additional sites by the stereo systems than by the mono systems. *A. pallidus* was recorded an equal number of nights by both mono and stereo systems; however, all other species were recorded on more nights by the stereo systems. More calls were identified to species on the stereo systems for every species except *M. ciliolabrum* (Table 1.3), and for all species including *M. ciliolabrum*, the average number of calls by site was higher on stereo systems (Table 1.4).

Based on 10,000 runs of each simulation, both occupancy and detection rate estimates increased when the simulated detection histories were generated from stereo observations versus mono observations. The magnitude of these increases differed by species. The estimates for some species benefited more than others, but stereo estimates were consistently higher for all species (Figure 1.1 and Figure 1.2).

Discussion

The stereo-microphone deployment strategy outperformed the mono-microphone strategy in nearly every category assessed. The stereo systems increased the overall number of triggered recordings, and the number of call sequences identified to species. For three species, the stereo deployment strategy increased the number of sites where the species were identified. The remaining species were recorded at all sites by both systems. For seven of the eight species, the stereo systems identified calls on more nights than the mono systems. For the eighth species (*A. pallidus*), both the mono and the stereo systems identified *A. pallidus* on only three nights; however, the stereo system identified more *A. pallidus* calls than the mono system (5 and 3, respectively).

For all 4 species included in the simulation study, the stereo deployment strategy led to increases in detection rate, although the magnitude of the increase was species-specific and only a modest increase in some cases. For rare or elusive species, even a few additional recordings could be important for species identification. Law et al. (2015) found that additional bat detectors (up to three) in their study area increased the precision of activity level estimates, particularly for less common species. Bat detectors that have the stereo microphone option offer an alternative to adding more detectors. An additional benefit is that if one microphone gets damaged or fails, the second microphone serves as a back-up. Since the left and right stereo channels trigger independently, the second mic is still fully functional even if the other mic is damaged.

While the stereo systems produced more identifiable call sequences than the mono systems, there are drawbacks to using the stereo deployment strategy. Deploying two microphones per detector requires an extra microphone, microphone extension cable, and equipment to elevate and mount the extra microphone at each site. With the addition of extra equipment at each site, there is an increased risk of equipment damage or loss from weather, wildlife, livestock, etc. Since the stereo systems trigger more often, they also require more audio storage, e.g., extra SD cards and file storage space. More events triggering on the stereo systems may result in the need for more frequent battery replacement, especially with lower capacity batteries, or for long-term deployments. For remote sites, this also adds to the logistics, since additional site visits may be necessary, although each additional bat detector deployed requires a completely new setup of all equipment (recorder, mic, batter, cables) and the logistics to set up an additional site.

To manage the automated processing of the stereo files, I split the stereo recordings into separate R- and L-channel files prior to using the SonoBat Classifier. Splitting files adds an extra step to the process, but helped with file organization. Since there were 2.7 times more triggered recordings from the stereo systems, processing time increased proportionally. Additionally, the SonoBat Classifier did not identify the stereo recordings as efficiently as the recordings made on the mono systems, possibly due to the different sampling rates used in these setups. The triggered stereo recordings were identified to species only 11.55% of the time, while the triggered mono recordings were identified 21.95% of the time. Since the analysis software is constantly improving, future versions of SonoBat, or other software, may have increased efficiency and identification rates.

Stereo recording did not provide an equal benefit for all species. The species with the most identified calls was *M. ciliolabrum* (968 identified calls from stereo recordings and 1097 from mono recordings). This species was identified by mono and stereo systems at every site, and was the only species that was identified more times by the mono systems than by the stereo systems. On a site-by-site basis, however, there was only one site where more mono recordings of *M. ciliolabrum* were identified than stereo recordings. The site where the mono system outperformed the stereo system for *M. ciliolabrum* was a site with high *M. ciliolabrum* activity overall. *M. ciliolabrum* uses a frequency modulated call that can initiate at frequencies as high as 106 kHz (Szewczak et al. 2011b). The mono systems sample at the 384 kHz rate, making it possible to digitally represent higher frequencies than the stereo system that sampled at the lower rate of 192 kHz. Since the recordings from the mono systems were able to capture the entire frequency range of *M. ciliolabrum* calls, this may have influenced the classification rates.

M. ciliolabrum was ubiquitous, but pallid bats (*A. pallidus*) were only identified in five stereo recordings, and never identified on both stereo channels from the same recording event. Looking only at the L-channel from the stereo, there were only three *A. pallidus* recordings from two sites. The R-channel recorded only two recordings from two sites. Only three call sequences from two sites were identified from mono recordings of *A. pallidus*. When surveying for cryptic species like *A. pallidus* that either have very low occupancy rates, low detection rates, low identification rates, or a combination, there is a clear benefit to adding a second microphone.

According to Wildlife Acoustics (http://www.wildlifeacoustics.com/support/product-faqs-2/), their "guestimate is that most bat species can be detected well over 30 m," depending on a suite of factors including frequency and directionality of bat echolocation calls, along with temperature, humidity, and wind. There seems to be a large difference between the range of detection of bat activity (triggering distance), and the distance triggered call sequences can be identified to species. Based on the low overlap rates in identification made by the equipment and software used in this study, a range of 5 – 10 m is more realistic for species identification from field recordings. This identification range is similar to the results reported by Adams et al. (2012) from the lab.

The bat detectors with the capability to record in stereo have been marketed for their ability to record ultrasound on one channel, and audible spectrum on the second channel (wildlifeacoustics.com). This deployment strategy has been used to monitor birds on one channel, and bats on the other channel (e.g., Heist 2014). Kalda et al. (2015) capitalized on the stereo capabilities of these systems to survey bats at the forest edge on one channel, and bats in the forest interior on the other channel. Kalda et al. (2015) did not mention the possibility of call overlap in their study, but because they separated their microphones using 10 m and 50 m extension cables stretched in opposite directions, it is unlikely that high quality calls from an individual bat were recorded on both channels simultaneously.

Acoustical monitoring for bats can be a useful tool, especially for large-scale studies. However, the process of species-identification of bats acoustically presents several challenges. Software for bat species identification is based on search-phase echolocation calls from extensive call reference libraries (Szewczak et al. 2011a). Collecting these reference calls typically requires a bat to be captured, identified in hand, (sometimes) marked, and recorded upon release (Parsons and Szewczak 2009, Szewczak et al. 2011a). SonoBat and other analysis programs are based on thousands of these reference recordings, but the available reference recordings represent only a small fraction of all possible call types for each species (Parsons and Szewczak 2009). Search-phase echolocation calls can vary both between and within habitat types (Parsons et al. 1997), and call reference libraries do not reflect the entire range of variation. Additionally, bats emit many ‘social calls’ that are not well understood (Altringham and Fenton 2009), and are not always included in software reference libraries.

Interest in understanding the effects of white nose syndrome, energy development, and climate change on bat communities has been driving interest in landscape-level studies of bats in North America (Loeb et al. 2015). Acoustical monitoring is one of the few available methods to survey bats over large spatial scales, and is sometimes the only practical method to survey areas with limited access. However, species identification using acoustical methods has its limitations, and it is not possible to identify every recording to species. One way to increase the number of identified calls at a survey site is to take advantage of the stereo recording capabilities of some of the currently available bat detectors. When deciding whether or not to use the stereo capabilities of the Wildlife Acoustics detectors (or future detectors with this capability), it is important to consider the costs, benefits, and project goals.

As of 2016, Wildlife Acoustics is the only company with a commercially available bat detector-recorder that can record in stereo. Wildlife Acoustics, Inc. released the first commercially available two-channel ultrasound detector/recorders in 2009, and the SM2BAT+ systems used in this study were released in 2012. In 2014, Wildlife Acoustics released the SM3BAT detector, increasing the stereo sampling rate to 256 kHz (www.wildlifeacoustics.com/products/song-meter-sm3bat). Using the stereo capability of these recorders increases the sampling area and detection rate, which may be particularly beneficial when surveying for rare species. Recording and analyzing ultrasound signals is an amazing way to peek into the activities of bats, and new technological advances are continually being made that increase the ability to indirectly observe these cryptic species.

Table 1.1. Deployment dates, site names, and number of survey nights for each site included in this study

Site	Year	# nights	deployment dates
PICE204	2013	13	6/13/2013 - 6/26/2013
PICE210	2013	7	7/17/2013 - 7/24/2013
PICE214	2013	7	7/19/2013 - 7/26/2013
PICE216	2013	7	7/25/2013 - 8/1/2013
PICE222	2013	7	8/2/2013 - 8/9/2013
PICE302	2014	7	6/23/2014 - 6/30/2014
PICE309	2014	7	6/21/2014 - 6/28/2014
PICE310	2014	8	6/6/2014 - 6/14/2014
PICE312	2014	7	7/6/2014 - 7/13/2014
PICE315	2014	6	6/10/2014 - 6/16/2014
PICE317	2014	13	6/22/2014 - 7/5/2014
PICE321	2014	6	7/15/2014 - 7/21/2014
PICE322	2014	7	7/7/2014 - 7/14/2014

Table 1.2. Calls of each species that were identified from stereo recordings. Total ID is the number of recordings identified to species from one or both stereo channels. Duplicate ID is the subset of the Total ID recordings that were identified to species on both stereo channels. The duplicate information is also expressed in terms of the Percent Overlap between the left and right channels.

Species	Total ID	Duplicate ID	Percent Overlap
Anpa	5	0	0
Epfu	261	51	19.54
Laci	192	27	14.06
Lano	61	2	3.28
Myci	968	23	2.38
Myev	219	9	4.11
Mylu	135	5	3.70
Tabr	541	68	12.57

Table 1.3. Species-specific recordings for both the mono and stereo systems, summarized by site, and by night. Summary of number of sites where each species was identified, the number of nights each species was recorded, and the total number of identified calls for each species. Table results are based on consensus decisions from the SonoBat Classifier, and the addition of the 4-pulse criterion.

	Sites		Nights		Total calls	
Species	Mono	Stereo	Mono	Stereo	Mono	Stereo
Anpa	2	3	3	3	3	5
Epfu	13	13	49	54	128	261
Laci	13	13	43	58	98	192
Lano	9	10	21	35	40	61
Myci	13	13	79	88	1097	968
Myev	13	13	43	67	82	219
Mylu	8	12	21	40	41	135
Tabr	13	13	64	83	184	541

Table 1.4. Average number of calls per site for paired mono and stereo systems. Results from a single-tailed exact Wilcoxon signed rank test for differences in (H_o: average number of calls is equal from paired mono and stereo systems, and H_a: Stereo systems produce higher average calls per site).

	Mono		Stereo		Exact Wilcoxon signed rank test	
Species	$\bar{x}$	SE	$\bar{x}$	SE	V	*p-value*
Epfu	9.85	1.99	20.08	5.58	76.5	0.0007
Laci	7.54	2.58	14.77	3.52	56.5	0.0166
Lano	3.08	1.38	4.69	1.34	46	0.0342
Myci	84.38	52.36	74.46	22.54	70	0.0453
Myev	6.31	1.59	16.85	3.76	91	0.0001
Mylu	3.15	1.20	10.38	3.98	76	0.0010
Tabr	14.15	3.56	41.62	9.18	91	0.0001

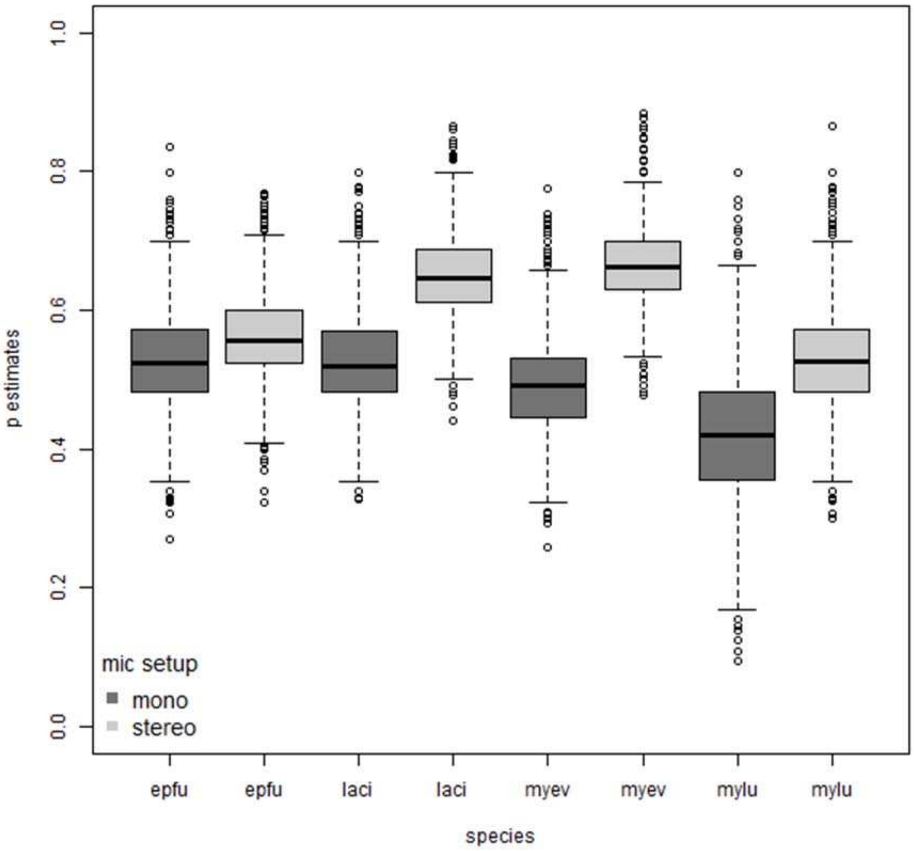

Figure 1.1. Distribution of detection probability (*p*) estimates, by species, from each of the 10000 occupancy simulations. For the four species listed on the x-axis, 10 000 new detection histories were generated by sampling from the field-collected detection histories. Simulated detection histories included 13 sites, and 5 survey nights.

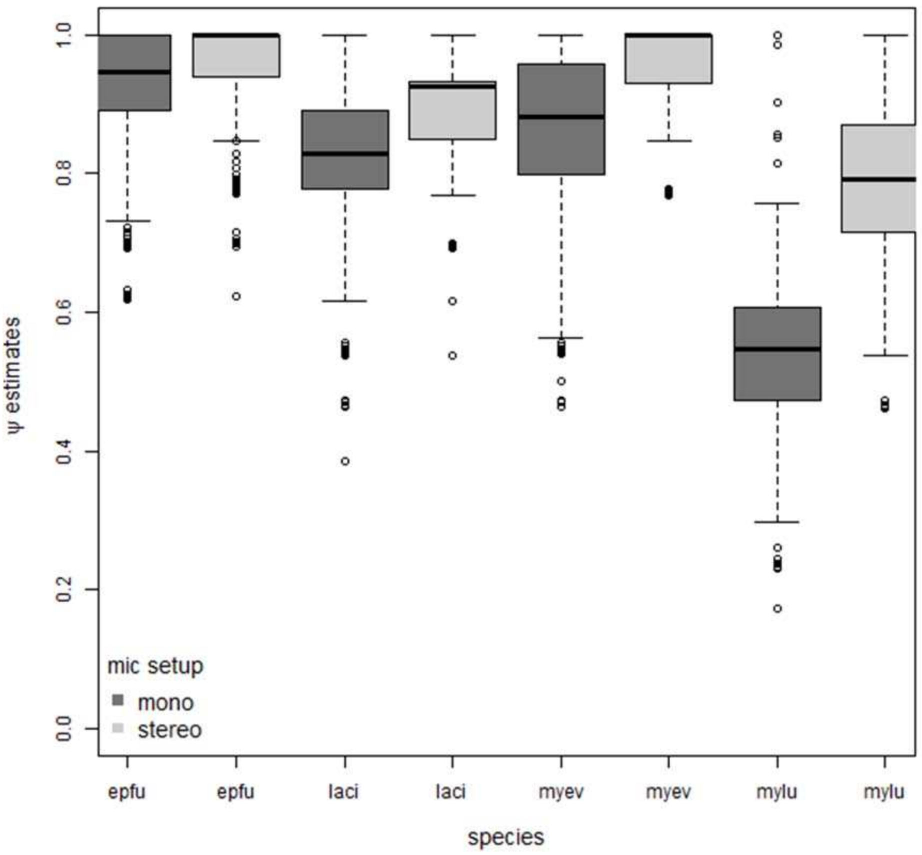

Figure 1.2. Distribution of occupancy rate (*ψ*) estimates, by species, from each of the 10 000 occupancy simulations. For the four species listed on the x-axis, 10 000 new detection histories were generated by sampling from the field-collected detection histories. Simulated detection histories included 13 sites, and 5 survey nights.

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CHAPTER 2

REDUCED ACTIVITY LEVELS OF BATS DURING A FIELD-BASED NOISE EXPERIMENT

Introduction

Anthropogenic noise from aircraft, railways, highways, and energy development, is pervasive throughout the continental United States (Barber et al. 2011). A growing body of literature indicates that anthropogenic noise pollution (hereafter, 'noise') can have negative effects on a wide range of terrestrial and aquatic taxa (see reviews by Barber et al. 2010, Laiolo 2010, Shannon et al. 2015). Noise pollution propagates far beyond areas of physical disturbance, causing "acoustical fragmentation" (Barber et al. 2011), and habitat degradation (Ware et al. 2015), in what may appear to be otherwise suitable habitat. The impacts of noise on wildlife differ by species, and can manifest in many ways including: changes in vocalizations (e.g., great tits, *Parus major*, Slabbekoorn and den Boer-Visser 2006), in lekking behavior (e.g., greater sage-grouse, *Centrocercus urophasianus*, Blickley et al. 2012), in occupancy rates (Francis et al. 2009), in predator-prey relationships (Francis et al. 2009), and in ecological services (Francis et al. 2012).

Most studies investigating noise effects on wildlife have been single-species, but a handful of songbird studies suggest that there are a variety of community-level effects (Francis et al. 2009, 2012, McClure et al. 2013, Ware et al. 2015). Using an experimental approach to create a "Phantom Road", McClure et al. (2013) showed that noise alone affects the distribution of migratory song birds, with reduced songbird abundance at noise treatment sites. Ware et al. (2105) called the same Phantom Road "an invisible source of habitat degradation" after finding lowered body condition of songbirds at sites exposed to the traffic noise playback. Using a ‘natural experiment’ where noisy gas compressor sites serve as treatments and quiet well pad sites serve as controls, Francis et al. (2009) reported decreased species richness at treatment sites, and altered predator-prey interactions. Using the same study system, Francis et al. (2012) demonstrated that changes in the vertebrate community at sites with higher noise levels altered the ecological services of pollination and seed dispersal.

Predators, especially those that use auditory cues to search for prey, may be particularly sensitive to changes in the acoustical environment (Siemers and Schaub 2011). Due to their reliance on auditory cues, bats have been a taxon of interest in laboratory noise exposure experiments. Schaub et al. (2008) and Siemers and Schaub (2011) showed that noise can negatively affect foraging success of the European bat *Myotis myotis*. *M. myotis* uses a strategy known as gleaning to locate prey by listening for the sounds of prey movement. More recently, a lab study by Bunkley and Barber (2015) found that *Antrozous pallidus,* a North American gleaning bat, also displays reduced foraging efficiency in noisy conditions. The authors conducted a laboratory experiment using playback noise of both traffic and gas compressor stations, at varying noise exposure levels ranging from 35-76 dBA. For all noise exposure levels, there was a twofold to threefold increase in time to locate prey (Bunkley and Barber 2015). Similarly, Luo et al. 2015 in another laboratory playback study found reduced foraging efficiency under noisy conditions for the Eurasian bat *Myotis daubentonii*, which uses echolocation, rather than passive listening, to find prey. Noise reduced foraging efficiency in *M. daubentonii*, even when the noise frequencies in the playback experiment did not overlap the frequencies used in echolocation (Luo et al. 2015). Despite these lab studies, there is limited information about the response of free-flying bats to noise in the field.

Bunkley et al. (2015) used the same ‘natural experiment’ approach as Francis et al. (2009, 2012) to begin investigating the impact of noise from natural gas compressor stations and well pads on bats in the field. Bats using ‘low frequency’ echolocation calls (<35 kHz) showed a reduction in activity levels at noisy compressor sites, while bats using higher frequency echolocation calls (>35 kHz) did not show reduced activity levels. One species, *Tadarida brasiliensis* had reduced activity levels at compressor sites, and also altered the duration and bandwidth of its echolocation search calls at the sites with higher noise levels (Bunkley et al. 2015). The results of this study indicate that compressor noise from energy development may be impacting the habitat use of at least some bat species.

The compressor stations used by Francis et al. (2009, 2012) and by Bunkley et al. (2015) for their ‘natural experiments’ in New Mexico are not used as frequently in other places where natural gas exploration is occurring. The Piceance Basin region in Northwestern Colorado is another area with oil and natural gas reserves, but the infrastructure differs considerably from the infrastructure described in the studies mentioned above. Large centralized gas plants take the place of most well-site compressors. Construction of well sites, roads and pipelines, heavy truck traffic, and drill rig noise are all common noise sources in the Piceance Basin.

During the drilling phase at well pads, drill rigs run continuously for several weeks to several months, depending on the site. Since construction noise and truck traffic taper off at night, noise from drilling has the greatest potential to affect bats and other nocturnal and crepuscular species in the Piceance Basin. In this study, I isolated drilling noise from the physical disturbance, human presence, truck traffic, and light pollution at a typical well pad drill site by using a noise playback experiment. I acoustically monitored bat the activity levels of free-flying bats at replicated “control” and “treatment” locations, where treatments included playback of drill rig noise. Based on previous lab and field studies, I expected to detect: 1) reduced bat activity levels at noise treatment sites, driven by 2) reduced activity of species using 'low-frequency' echolocation calls, and 3) no reduction in activity levels of species using 'high frequency' echolocation calls.

Methods

Study area – Study sites were located on Bureau of Land Management lands in Rio Blanco County, Colorado, U.S.A. (39.98°N 108.20°W). The dominant vegetation included mixed native grasses and forbes, non-native grasses, common rabbitbrush (*Chrysothamnus nauseosus*), and big sage (*Artemisia tridentata*). The nearby landscape included a mosaic of grasses and shrubs, mixed with pinyon pine (*Pinus edulis*) and juniper (*Juniperus* sp.). Due to the combination of more open grass and shrub land habitat, pinion-juniper woodlands, and abundant cliff and rock outcropping for roosting, this area supports at least 16 species of bats (Pers. comm. Heather Sauls, BLM White River Field Office, Nov. 2014). Elevation throughout the study area ranged from 1750 m to 2150 m. Primary land uses included cattle ranching, hunting, natural gas extraction, and recreation. The landscape is semi-arid with a 30-year average annual rainfall of 44.3 cm from 1981 - 2010 (WRCC 2016), and topographically diverse with many drainages and ephemeral streams, but flowing water is uncommon in summer months.

Site selection –To improve the chances of encountering commuting and foraging bats, all sites were located along drainages in the study area. I identified potential drainages in ArcGIS, and ground-truthed all drainages to make sure they met the following criteria, 1) located on BLM land, 2) drilling for natural gas was not currently taking place along the drainage, 3) over 1 km in length, and 4) at least one vehicle access point along the drainage for equipment drop off. Due to the high concentration of small 4x4 roads throughout the area, access to sites was not a significant limitation. The location of the paired sites was determined by drawing a random center point along the drainage in ArcGIS, and establishing paired sites 500 m in either direction (paired sites 1 km apart). I considered establishing more than one pair of sites along a drainage if the drainage was over 5 km in length, however, only one drainage in the study area met the length criterion (at approximately 9 km in length). In 2013, 11 drainages were selected, resulting in 24 sites (12 control-treatment pairs). In 2014, 28 sites (14 pairs) were established along 13 drainages. Due to equipment failures, 10 pairs (20 sites) from each year are included in the analysis. Data collection efforts for this project occurred only during summer months of 2013 and 2014 when bats were non-migratory.

Noise treatment –Using a Type I calibrated Larson Davis Sound Pressure Level (SPL) Meter, I took SPL readings and recorded audio of the noise 113 m from an active drill rig. Access to active drill sites is limited, and at this distance, I was at the edge of the physically disturbed construction site. The 15-minute equivalent continuous sound pressure level (L_{eq}) in A-weighted decibels (dBA) for the drill noise recording was 70.6 dBA (reference pressure of 20 µPa is used for all sounds pressure metrics). At each treatment site, I broadcast the drill rig noise recording so the levels reaching the central microphone (10 m from the speaker) roughly matched the L_{eq} levels approximately 100 m from a drill rig. To simulate actual drill rig activity, the noise playback was designed to run continuously for the duration of the treatment. Community brand R.5 outdoor speakers, in conjunction with Kemo electronics 6-16 volt 40 watt amps projected the drill rig playback, and were powered using two Power-Sonic 12-volt 103-Amp-hour rechargeable batteries connected in parallel, which could power the speakers continuously for approximately 48 hours. Due to equipment failures, some sites experienced gaps in the noise treatment and results are only reported for those sites where the noise treatment ran continuously from 2000 to 0600 hrs for at least four consecutive nights (Appendix 2.1).

Acoustical monitoring – At all sites, I continuously recorded the audible acoustical spectrum using Zoom H2 digital audio recorders (Zoom North America, Ronkonkoma, NY) in MP3, 64-kbps mode. As described by Mennitt and Fristrup (2012), sound pressure level metrics can be estimated using Zoom H2 units in the field, and can be accurate to within 1 dBA for summary metrics including A-weighted L_{90}, L_{50}, and L_{10}. L_x levels refer to percentile-exceeded sound levels, for example, the L_{90} for a site represents the sound level that is exceeded 90 percent of the time (NPS 2013).

In addition to recording the audible spectrum, at each site I deployed two SM2BAT+ ultrasound detector-recorders (‘bat detectors’) with SMX-US omnidirectional microphones (Wildlife Acoustics, Maynard, MA). SM2BAT+ units recorded in ‘triggered .wav mode.’ In this recording mode, the bat detector monitors the rolling background ambient sound levels, and when an ultrasound signal exceeds a user-defined threshold level, the detector initiates a recording and creates a file in .wav format. The central bat detector used the highest available sampling rate of 384 kHz. The second bat detector recorded in stereo, using two microphone extension cables to space the left and right microphones approximately 10 m apart, with each channel recorded at a sampling rate of 192 kHz. This was the maximum available sampling rate for ultrasound stereo recording on the SM2BAT+ units. Other than the sampling rate, the settings were identical. For the digital settings, I used an 18-dB trigger level, 16-kHz digital high pass filter, a 2.0-second window minimum, and a 5.0-second window maximum. Using the analog filter switches, I followed Wildlife Acoustics’ recommended settings (Wildlife Acoustics 2011), +36 dB gain, the analog high-pass filter set to 1 kHz, and microphone bias off. When using triggered .wav mode, the digital high pass filter defines the frequency band of interest, but does not actually filter out all lower frequencies (see Wildlife Acoustics 2011 for more details). Recordings saved directly to SD cards in the SM2BAT+ unit and were transferred to external hard drives for later analysis. All bat detectors were scheduled to power on at 2000 hrs (before sunset) and run until 0600 hrs (after sunrise). For the units recording in stereo, the left and right channels monitor the background and trigger independently, however, when either channel triggers, a stereo recording from both channels is created. I used external 6V sealed lead acid 36-amp hour batteries to power all SM2BAT+ recorders. These batteries consistently ran for at least 6 days before needing to be recharged and replaced. Ultrasound microphones were mounted at the top of large tripods approximately 2.15 m above ground. At this height, the ultrasound mics were above the vegetation. At each site, ultrasound recordings from both detectors were combined for analysis. See Appendix 2.2 for full details on recording equipment settings.

No animals were handled in this study, and all research described followed protocols approved by Colorado State University Institutional Animal Care and Use Committee (Protocol #12-3574A).

Audio processing – MP3 files from the Zoom H2 units were processed using Program AUDIO2NVSPL and the Acoustical Monitoring Toolbox (AMT, Damon Joyce, National Park Service Natural Sounds and Night Skies Division). I used this software to extract summary metrics of the acoustical conditions at each site, and report the nighttime L_{90}, L_{50}, and L_{10} in dBA for the duration of the experiment.

Ultrasound recordings (.wav files) were processed using SonoBat 3.2.1 West Suite (sonobat.com). Following the examples of others (e.g., Kalcounis et al. 2013, Kennedy et al. 2014, Parkins and Clark 2015), I passed all triggered .wav recordings through the SonoBat Batch

Scrubber 5.4 before automated classification. The SonoBat Batch Scrubber 5.4 removed low quality calls, and files that did not contain bat call sequences. I used the "high grade" setting, and included calls in the 5-20 kHz frequency range. Prior to running the stereo files through the scrubber, I split the stereo files into separate left and right channel recordings using the 'seewave' package and custom scripts written in R (R Core Team, 2013). The single-channel 384-kHz .wav recordings were batch scrubbed in their raw form.

Call sequences that were not removed by the scrubbing process were passed to the SonoBat 3.2.1 West Batch Classifier and identified using two methods as described in Clement et al. (2014): 1) "by evaluating mean parameter values of acceptable calls within a sequence" and 2) "by evaluating classification agreement among individual calls within a sequence". I followed the lead of other researchers (e.g., Kalcounis et al. 2013, Bunkley et al. 2015), and only accepted the classification decisions for call sequences when the SonoBat Classifier reached a "consensus" decision, i.e., both methods agreed on the classification. Additionally, I required at least 4 pulses from a sequence be included in all identification decisions. A ≥3 pulse criterion has been used in studies where calls were manually identified (e.g., Johnson et al. 2002; Hourigan et al. 2009). The addition of the 4-pulse criterion is more conservative than using the "consensus" decision alone, and is intended to further reduce misclassification during the automated classification process.

For species with fewer than 50 recordings identified using the SonoBat Classifier, each call was manually vetted to visually confirm diagnostic call features. For species with more than 50 identified recordings, a minimum of 20 call sequences were verified by manually vetting. For species that were confirmed to be present in the study area, the results and models include all automated identifications, not only the subset of calls that were visually vetted.

I used the SonoBat Suite to summarize ultrasound recordings in two ways. First, the number of recordings that passed the SonoBat Batch Scrubber (described above), were tallied by site and by night. These counts of ‘all bats’ provide a coarse look at bat activity prior to species identification. Throughout this chapter, the term ‘all bats’ refers to recordings that contain bat calls, but that have not been identified to species. Only a small subset of the ‘all bats’ recordings could be identified to species using the SonoBat Classifier (Figure 2.1). I summarized the number of species-specific nightly recordings at each site (i.e., species-specific encounter histories, for use in the multi-species model described below).

Statistical analyses –

Site-level covariates – Three site-level covariates (noise treatment, elevation, and percent shrub cover) were included in all models described below. Elevation was measured using a Garmin GPSMAP 64s unit, and standardized for inclusion in the model. Percent shrub cover was estimated visually and included in the model as a proportion ranging from 0 to 1. The treatment covariate for each site was included in the models as either 0 (control site) or 1 (noise site).

Survey-level covariates – Moisture and temperature both affect sound propagation, which could influence the recordings of bats, and the subsequent identification/detection using automated methods. Thus, the nightly survey-level covariates of ‘minimum temperature’ and ‘minutes of rain’ were included in the 2013 and 2014 ‘all bats’ models described below. Site-specific temperature was not recorded; however, temperature data were available from a NOAA weather station at Pinto Creek, near the southern edge of the study area (elevation 2115 m, latitude 40.0097, longitude -108.4597). For the 2013 sites, the distance to this weather station ranged from 1.9 km to 43.3 km. In 2014, the distance to sites ranged from 0.4 km to 51.6 km. I obtained the overnight minimum temperatures (in Celsius) from the Pinto Creek NOAA station, and applied these nightly minimum temperatures as a proxy for the temperatures at the study sites. To calculate the number of minutes of rain each night, I used the NPS Acoustical Monitoring Toolbox software, and identified the duration (in minutes) of nightly rain events during survey nights. Rain events were identified by visual inspection of hourly spectrograms, and confirmed by listening to the associated audio files from control sites. The nightly rain duration from each control site was also applied to the corresponding treatment site. Due to the noise playback at treatment sites, it was not possible to extract rain events directly from treatment sites.

Modeling bat activity – I used a Bayesian modeling approach (e.g., Kéry and Schaub 2012, Hobbs and Hooten 2015) and modeled bat activity levels as a function of covariates using a hierarchical binomial-mixture model (Royle 2004), in this case, a binomial-Poisson model. The Royle (2004) model was the first to estimate both abundance (N) and detection (p) in a model of point count data from bird surveys, where multiple sites were surveyed on multiple occasions within a single season. One assumption of this class of models is that there are no double counts of individuals (Kéry and Schaub 2012). However, with acoustical monitoring techniques, it is not possible to determine if multiple recordings of the same species are from one bat making multiple passes or multiple individuals making separate passes. Since the count data from acoustical bat surveys cannot differentiate between individuals, the model estimates should be interpreted as 'bat activity levels,' rather than abundance. The Royle (2004) model assumes closure during the survey season. To address this assumption, all bat surveys were limited to no more than seven consecutive nights, and surveys occurred during the summer season when migrations were not occurring. An additional model assumption is that the detection probability is constant for all individuals (Kéry and Schaub 2012). Since individual bats are not marked, and unique individuals are not identifiable using current acoustical monitoring techniques, it was not possible to test this assumption.

The 'all-bats' model used a simple single-species model, but in this case, nightly recordings that contain unidentified bat calls are aggregated and included in a single count for each night, at each site. As in the Royle (2004) abundance model, the true activity level (N) cannot be directly observed, and can be described as: $N_i \sim \text{Poisson}(\lambda_i)$, where N_i is the true activity level at each site (i), and λ_i is the mean. To evaluate the effects of noise treatment, elevation, and shrub cover on activity levels, I used a log link:

$\log(\lambda_i) = \beta_0+\beta_1*\text{noise}_i+\beta_2*\text{elev}_i+\beta_3*\text{cover}_i$.

Count data are incorporated into the binomial observation model such that: $y_{ij} \sim \text{Binomial}(N_i, p_{ij})$, where y_{ij} is the observed count of triggered bat recordings at site i during survey j, N_i is the bat activity level at site i, and p_{ij} is the detection probability for site i, on survey j. Temperature and rain, survey-level covariates thought to affect detection, were included in the detection model as $\text{logit}(p_{ij}) = \alpha_0+\alpha_1*\text{temp}_{ij}+\alpha_2*\text{rain}_{ij}$.

Vague normal priors were used for α and β.

I also estimated species-specific bat activity level by developing a simplified version of a multi-species binomial-Poisson mixture model, as described by Yamaura et al. (2012). The structure of the 'multi-species' model is similar to the 'all bats' model, with the addition of a species-specific index, s:

$$N_{is} \sim \text{Poisson}(\lambda_{is})$$

$$y_{ijs} \mid N_{is} \sim \text{Binomial}(N_{is}, p_s)$$

$$\log(\lambda_{is}) = \beta_{0s}+\beta_{1s}*\text{noise}_i+\beta_{2s}*\text{elev}_i+\beta_{3s}*\text{cover}_i$$

Recordings of bats that were identified to species by the SonoBat Classifier, and met the consensus and 4-pulse criteria, were incorporated into the observation model where y_{ijs} is the number of identified calls of species s, at site i, on night j. N_{is} is the estimated activity level of species s at site i. As in the 'all bats' model, the log-transformed expected activity levels are described by a linear combination of the species-specific intercept β_{0s}, and the site-specific covariates and their coefficients $\boldsymbol{\beta}_s$.

Convergence of hierarchical mixture models in Bayesian analyses can be difficult to achieve (Kéry 2010), especially for more models with more complexity. To facilitate convergence, the multi-species model does not include covariates on detection, but still allows detection estimates to vary by species: $p_s \sim \text{Beta}(0,1)$. Vague normal priors were used for all β_s.

The 'all bats' and 'multi-species' mixture models were implemented using the software program WinBUGS (Spiegelhalter et al. 2003) and the R2WINBUGS package in program R (R Core Team 2013). Data from the 2013 and 2014 field seasons were analyzed separately, using the same model structure. To fit each of the above models, I ran 5 parallel MCMC algorithms for 150,000 iterations, with a thinning rate of 5, and the first 100,000 iterations were discarded as burn-in. Chain mixing and convergence were assessed using the Gelman-Rubin statistic ($\hat{R}$) provided by WinBUGS ($\hat{R} < 1.05$ for all parameters) (Brooks and Gelman 1998). For the WinBUGS code for these models, see Appendix 2.3

Results

Acoustic environment – The nighttime L_{50} over the duration of the control or treatment period was estimated from MP3 recordings at the central microphone, located 10 m from the speaker at treatment sites. In 2013, estimates of the nighttime L_{50} at control sites ranged from 24.2 to 34.4 dBA. Similar nighttime L_{50} estimates in 2014 ranged from 23.7 to 31.8 dBA over the duration of the control period. The projection of drill rig noise elevated the L_{50} estimates at treatment sites, where nighttime L_{50} levels ranging from 63.2 to 79.3 dBA over the duration of the treatment period in 2013. Estimated L_{50} levels for the nighttime treatment period ranged from 55.0 to 76.0 dBA in 2014 (Figure 2.2).

Ultrasound recordings – Prior to species identification, the SonoBat Scrubber processed the ultrasound .wav files, and removed recordings that were unlikely to contain bats calls. The remaining recordings, containing unidentified bat calls, were incorporated into the 'all bats' models. In 2013, 18,173 recordings of unidentified bat calls were included in the 'all bats' model, and 9,361 calls were included in the 2014 model.

From the pool of 'all bat' recordings, only a subset could be identified to species. The SonoBat Classifier identified 13 bat species at the study sites: *Antrozous pallidus*, *Eptesicus fuscus*, *Lasionycteris noctivagans*, *Lasiurus blossevillii*, *Lasiurus cinereus*, *Myotis californicus*, *Myotis ciliolabrum*, *Myotis evotis*, *Myotis lucifugus, Myotis thysanodes*, *Myotis volans*, *Parastrellus* (formerly *Pipistrellus*) *hesperus*, and *Tadarida brasiliensis*. Six species *E. fuscus*, *L. cinereus*, *M. ciliolabrum*, *M. evotis*, *M. lucifugus,* and *T. brasiliensis* met the quality criteria (see Methods) for inclusion in the multi-species models. Over 100 calls were identified by the SonoBat Classifier for each of these species in 2013 and 2014 (Table 2.1). Manual vetting confirmed the presence of *A. pallidus* and *L. noctivagans* in the study area, but due to the low number of recordings of these species and failure to meet the inclusion criteria, they were not included in the multi-species models. Due to the lack of visually diagnostic features in the available recordings, I was not able to verify the presence of *L. blossevillii*, *M. californicus*, *M. thysanodes*, *M. volans*, or *P. hesperus*.

'All bats' model – In 2013, 8 out of 10 control sites had higher estimated bat activity levels when compared with the paired noise treatment sites (Figure 2.3). In 2014, all 10 treatment sites had lower estimated bat activity, although 95% credible intervals overlapped control estimates for some pairs (Figure 2.3).

In both 2013 and 2014, noise treatment and elevation had negative effects on overall bat activity levels in the 'all bats' model (Figure 2.4). In 2013, shrub cover had a negative effect on bat activity levels, but this effect was not significant in 2014. Rain had a negative effect on detection in 2013, but in the 2014 the 95% credible interval for the noise parameter overlapped 0 (Figure 2.5). The effect of minimum nightly temperature on overall bat detection was inconsistent between the two years, with a negative effect on detection in 2013, and positive effect on detection in 2014. The posterior mean for detection rate in the 'all bats' model was 0.18 (credible interval 0.10 to 0.23) in 2013, and 0.14 in 2014 (credible interval 0.07 to 0.24) (Figure 2.6).

Multi-species model – In 2013 and 2014, the SonoBat Classifier identified over 100 calls for each of the six bat species in the multi-species model (*E. fuscus*, *L. cinereus*, *M. ciliolabrum*, *M. evotis*, *M. lucifugus,* and *T. brasiliensis*) (Table 2.1).

Noise had a consistent negative effect on activity levels of *M. ciliolabrum* and *L. cinarius* in 2013 and 2014, but the response to noise for other four species was inconsistent or neutral between years (Figure 2.7a). Elevation did not have a consistent effect on activity level for any of the six species in the model (Figure 2.7b), and only *M. ciliolabrum* responded consistently to shrub cover both years (Figure 2.7c). Detection rates varied by species and by year, with most 95% credible intervals overlapping between years (Figure 2.7d). Mean species-specific detection estimates in 2013 ranged from 0.06 for *L. cinereus* to 0.23 for *M. evotis*. In 2014, detection estimates ranged from 0.06 for *E. fuscus* to 0.22 for *M. evotis*.

Discussion

In this designed field experiment, the playback of drill rig noise had a negative effect on overall bat activity levels in both 2013 and 2014. 'Noise' was the only site-level covariate that consistently had a negative effect on overall activity level in both study years (Figure 2.4).

Although multiple species of bats responded to the noise playback, the affected species did not fit neatly into the expected categories. Based on the 'natural experiment' by Bunkley et al. (2015), I anticipated that any reduction in bat activity would be attributable to bats in the 'low frequency' assemblage. However, no obvious assemblage-level patterns were detected in this study. Of the six species of bats included in the multi-species model, only *M. ciliolabrum* and *L. cinarius* showed consistent negative response to the noise treatment in both years. Interestingly, these two species have very different life histories and echolocation characteristics. *L. cinarius* is the largest bat species occurring in Colorado, and *M. ciliolabrum* is one of the smallest (Armstrong 2011). *L. cinarius* is a migratory, tree-roosting species in the assemblage of 'low-frequency' bats, and uses characteristic echolocation frequencies below 35 kHz. *M. ciliolabrum* is non-migratory, uses crevices and caves as roosts and hibernacula, and is part of the 'high frequency' assemblage of species, using echolocation calls predominantly > 35 kHz (e.g., as characterized by Humboldt State University Bat Lab 2011, and as grouped by Bunkley et al. 2015).

For two additional species, *T. brasiliensis* and *M. evotis*, the noise treatment had a negative effect on average activity level, but the 95% credible intervals for the noise treatment parameters overlapped with zero in at least one of the years (Figure 2.7a). In 2014, reduced activity levels of *T. brasiliensis* at noise treatment sites seemed fairly clear (Figure 2.7a), but in 2013, there was at best a weak response to the noise treatment. The results from my experiment, combined with the results from the study by Bunkley et al. (2015), suggest that *T. brasiliensis* may have some level of avoidance of noisy environments.

Understanding why some bat species respond to anthropogenic noise is an area of active research. For bats that rely on passive listening to locate prey, it has been suggested that noise may be masking the sounds of prey movement (Schaub et al 2008, Siemers and Schaub 2011, Bunkley and Barber 2015). Noise may also be functioning as a distraction or an impediment to signal processing (Barber et al 2003, Chan et al. 2010), or the novelty of noise sources may lead to avoidance behavior (Luo et al. 2015). For free-flying bats in a natural setting, the mechanisms that influence behavior likely vary between species. Growing evidence suggests that noise can affect habitat use and foraging efficiency of species that rely on gleaning methods to locate prey (Schaub et al 2008, Siemers and Schaub 2011, Bunkley and Barber 2015), as well as bats that hunt using echolocation for prey localization (Luo et al. 2015). *A. pallidus* was the only obligate gleaner present in the noise experiment study area, but due to very few *A. pallidus* encounters, it is not possible to say what effect the noise treatment had on this species in the field. All of the species that responded negatively to noise were either aerial foragers, using echolocation to locate prey (*M. ciliolabrum*, *L. cinarius*, and *T. brasiliensis*), or in the case of *M. evotis*, are known for using both aerial foraging and gleaning techniques (Faure and Barclay 1994).

Although there are many noise sources associated with natural gas development, this study focused only on noise produced by drilling for natural gas. Active drill rigs produce continuous noise 24-hour a day for the weeks or months it can take to develop a well site. Unlike traffic noise that fluctuates throughout the day, and often tapers off at night, drilling noise is a fairly constant broadband signal (Figure 2.8 and Figure **2.9**) that affects the nighttime hours when bats are most active. The broadband signal with little variation also made this noise source a good candidate for a playback experiment, and the noise treatment reduced activity levels for some bat species. If this reduction in activity is resulting in lower quality of habitat for certain species, then at the landscape level the outcome is a form of habitat fragmentation (Barber et al. 2011).

Understanding the spatial extent of the noise impact on bats would is helpful for potential management or noise mitigation efforts. To understand the spatial impacts, we need a better understanding of the noise levels that elicit a response from each bat species. The maximum and minimum nighttime L_{50} levels (55.0 to 79.3 dBA) at treatment sites in this experiment correspond to the noise levels experienced approximately 40 m – 650 m from an active drill rig. These levels of noise exposure encompass the exposure levels from several laboratory studies (e.g., Bunkley and Barber 2015, Luo et al. 2015). However, audio distortions (Figure 2.10), may have affected the acoustic metrics that were estimated from some MP3 files. It is not possible to determine exactly how many audio files were affected in this way, because these distortions were only visible in the spectrograms from control sites (and audible in the corresponding MP3 recordings), in low-wind conditions. At treatment sites, or in the presence of high winds, this buzzing is not audible or visually identifiable (Figure 2.8). Future field studies that explicitly test a wide range of noise levels, and that quantify those levels more accurately, are needed to fully understand the spatial extent of noise disturbance for bats and other wildlife.

There are still many things to learn about the response of different bat species to noise pollution and other anthropogenic stressors. For some species, year-to-year response to the noise experiment seemed idiosyncratic. Similarly, the response by bats to light pollution has been variable. Stone et al. (2012) found that LED lighting decreased activity levels of one bat species (*Rhinolophus hipposideros*), but did not affect another species (*Pipistrellus pipistrellus*). Luck et al. (2013) found no clear assemblage-level response to light pollution, and posits that the responses are more nuanced (see also Scanlon and Petit 2008, and Stone et al. 2015). Individual heterogeneity may also be a factor. For example, only three of four individual bats responded to noise treatments in a lab setting (Luo et al. 2015). Noise pollution and light pollution often occur together, and the effects of these sensory disturbances are often difficult to disentangle (Longcore and Rich 2004, Kempenaers et al. 2010). Further investigation is needed to understand how the interaction between these two sources affects bats.

My study supports the findings that some bat species alter activity levels in noise-polluted environments. Further, reduced activity level is a more subtle response than complete abandonment of an area. Whether noise alone could exclude some species from otherwise usable habitat over a longer time period is unclear. In many cases, sensory disturbances are part of permanent land conversion. However, in this study system, noise disturbance from the drilling process is relatively short-term. The conservation community is beginning to recognize that sensory disturbances can impact many species (e.g., reviews by Barber et al. 2010, Laiolo 2010, and Shannon et al. 2015), but we still know very little about how long these effects persist, which is a focus of a companion study (see Chapter 3).

Table 2.1. Observed counts of recordings of each bat species (y_{ij} for each species for all sites combined) included in the 'multi-species' model per year. These are recordings that were identified to species using the SonoBat Classifier, and met all additional inclusion criteria.

species	2013	2014
E. fuscus	441	378
L. cinereus	183	303
T. brasiliensis	1021	793
M. ciliolabrum	1445	1416
M. evotis	336	420
M. lucifugus	177	275

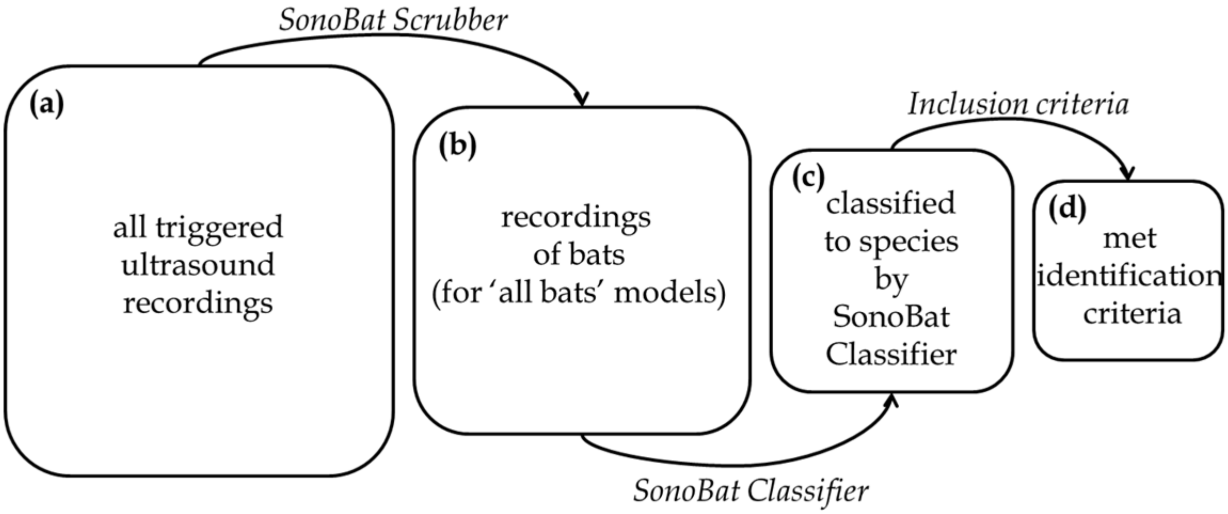

Figure 2.1. Diagram of ultrasound file sorting and species identification process. The SonoBat Scrubber scanned all triggered ultrasound .wav recordings (a), and removed recordings that did not contain bat calls. Files that passed through the SonoBat Scrubber contained ultrasound signals consistent with bat calls (b). These unidentified recordings of bats were included in the 'all bats' model. The SonoBat Classifier identified a subset of the 'all bat' recordings to species (c), but only calls that met the inclusion criteria (d) were included in the 'multi-species' models.

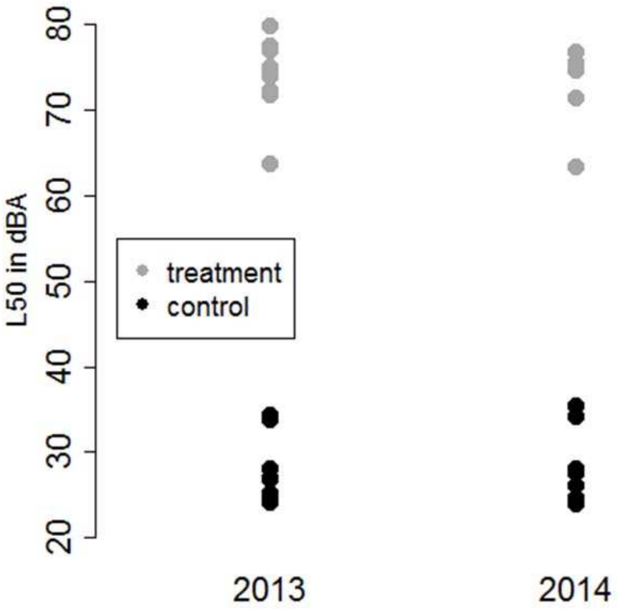

Figure 2.2. Control and treatment nighttime L_{50} levels for the duration of the experiment, as estimated from MP3 recordings made at the center microphone of each site (10 m from the noise source).

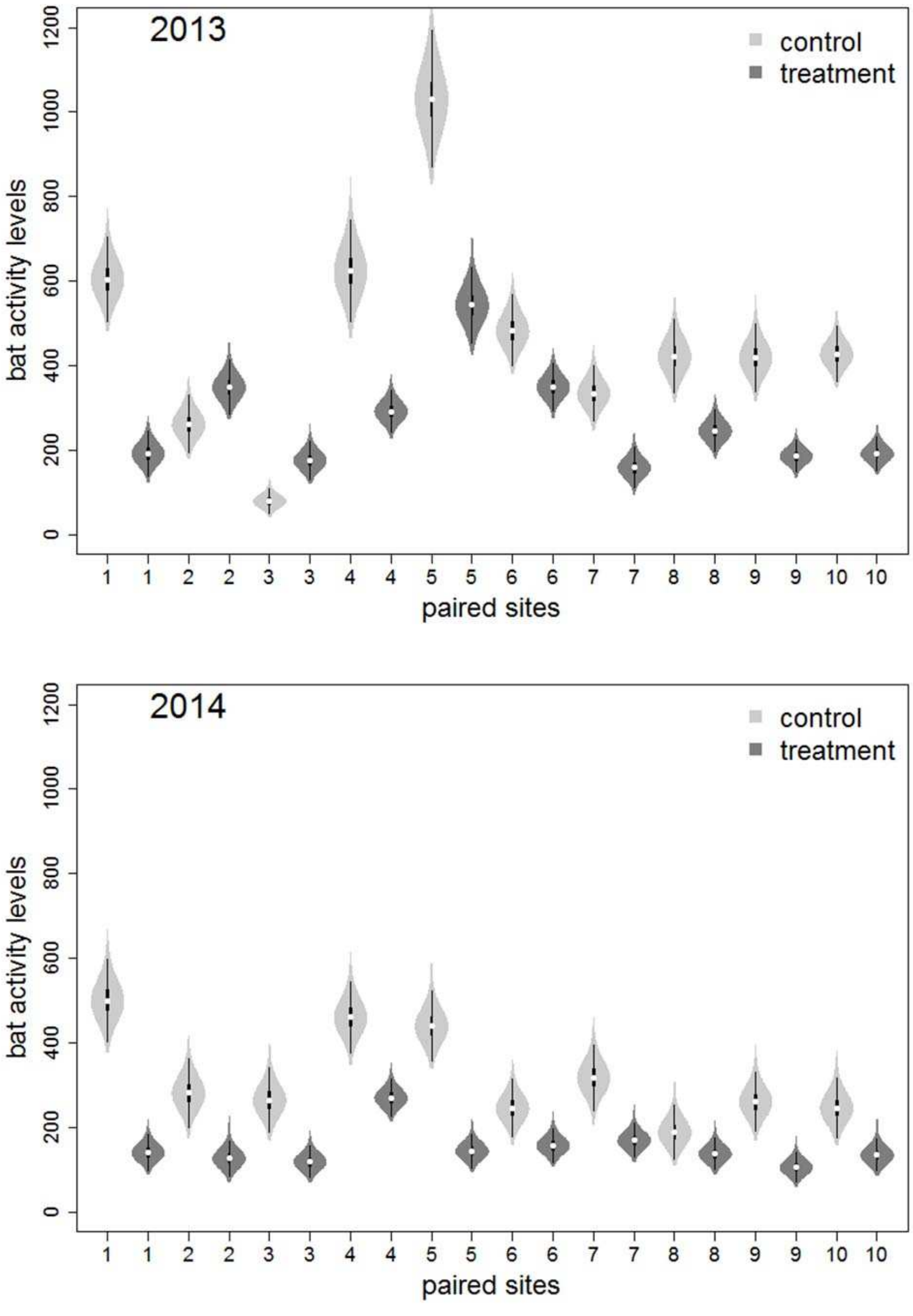

Figure 2.3. 'Violin plots' of the posterior distribution of overall bat activity estimates (N_i) for all paired control and treatment sites in the 2013 and 2014 'all bats' models. Violin plots display a boxplot within a density plot.

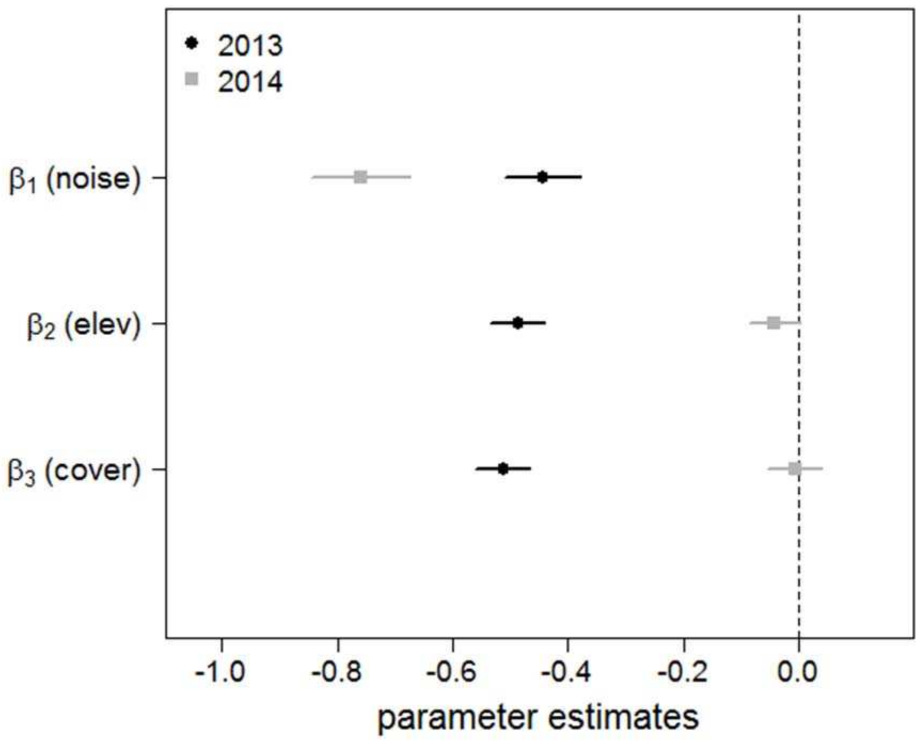

Figure 2.4. Mean parameter estimates (with 95% credible intervals) for site-level covariates in the 2013 and 2014 ‘all bats’ models of overall bat activity.

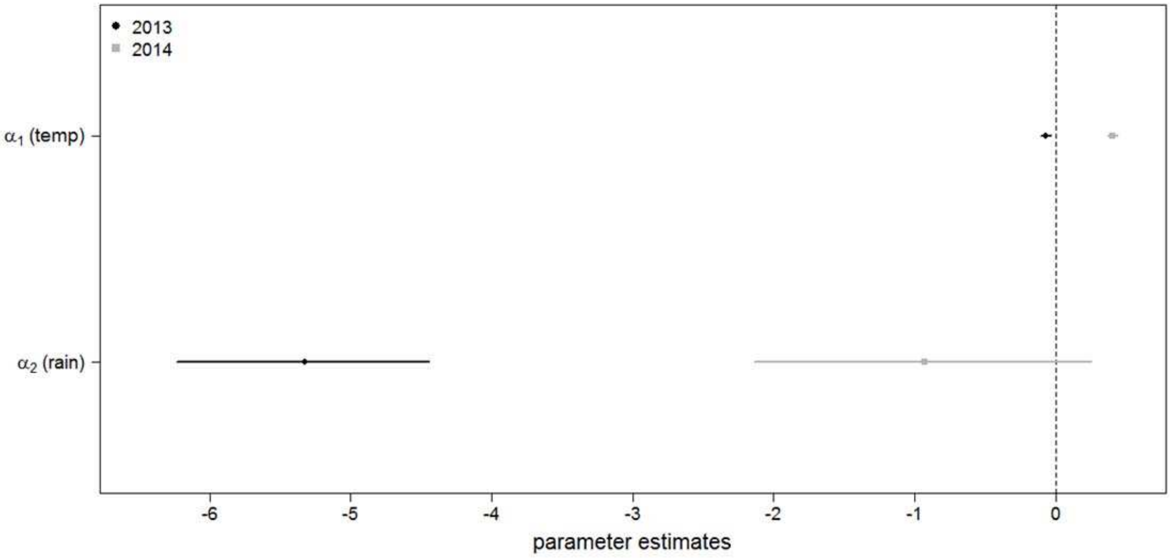

Figure 2.5. Mean parameter estimates (with 95% credible intervals) for survey-level covariates in the 2013 and 2014 'all bats' models of overall bat activity.

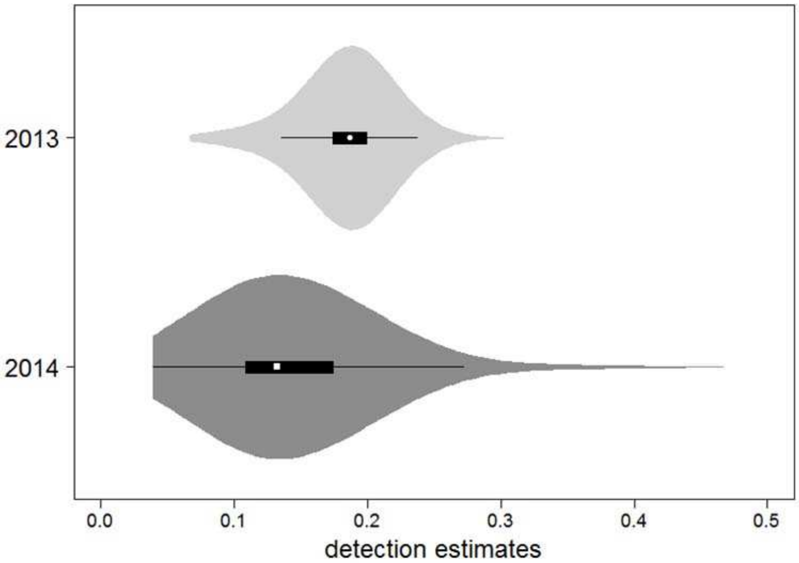

Figure 2.6. Violin plots of the posterior distributions of detection rates over all sites and all nights in the 2013 and 2014 'all bats' binomial-mixture models.

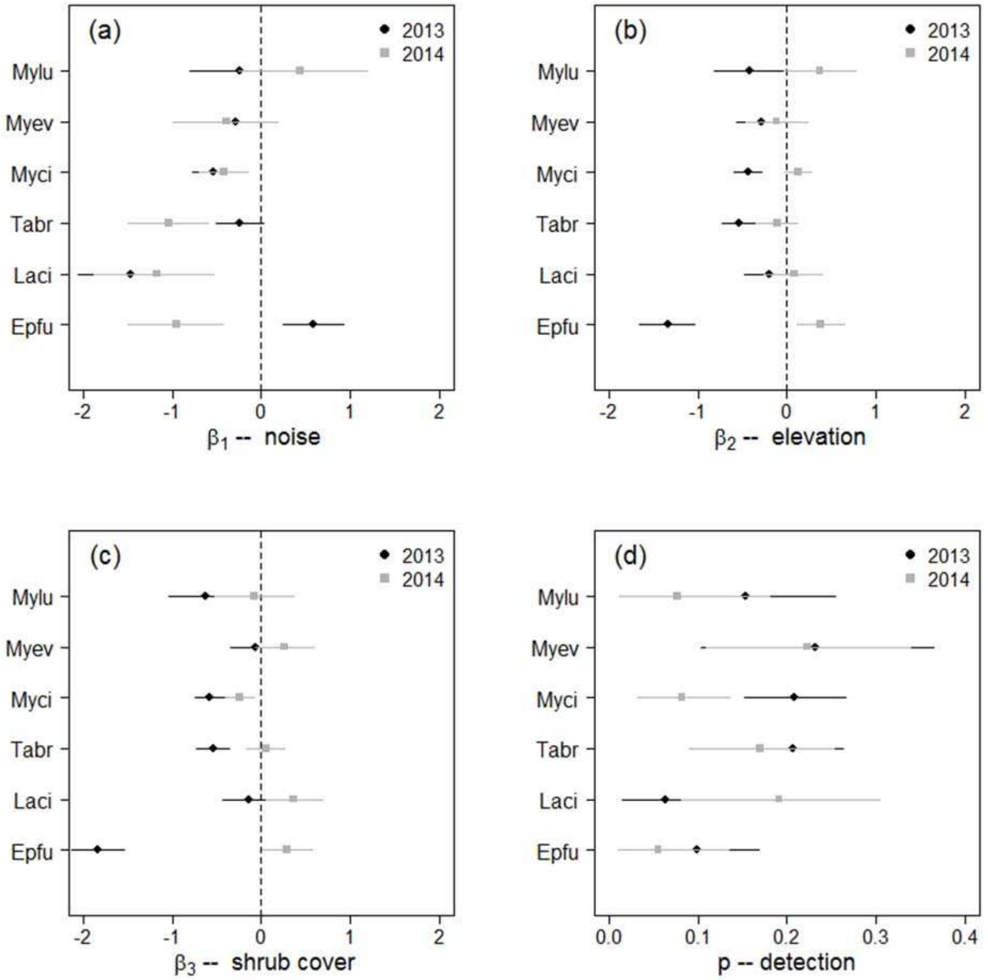

Figure 2.7. Estimates ± 95% credible intervals for the parameters [(a) noise treatment, (b) elevation, (c) shrub cover, and (d) detection rate] in 2013 and 2014 for the 6 species in the multi-species hierarchical model. *M. lucifugus* - Mylu*, M. evotis* - Myev*, M. ciliolabrum* - Myci*, T. brasiliensis* -Tabr *, L. cinereus* - Laci*, and E. fuscus* - Epfu.

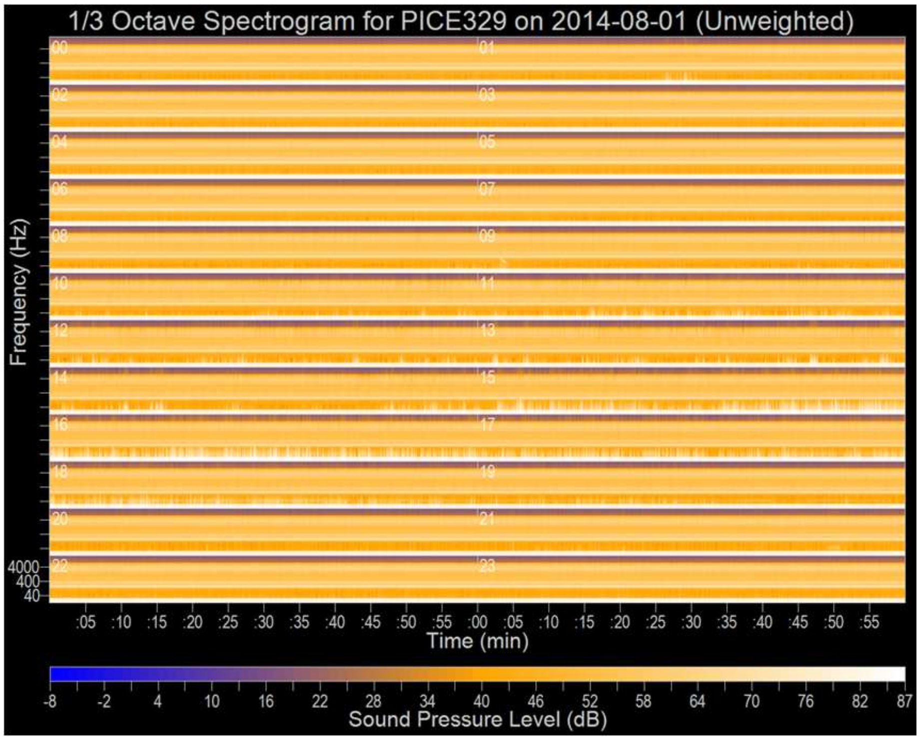

Figure 2.8. 24-hour spectrogram during a day of noise treatment. Spectrogram generated from the MP3 recording made 10 m from the noise source (the outdoor speaker). At this distance, noise from the treatment masks (visually and audibly) other sounds at the site.

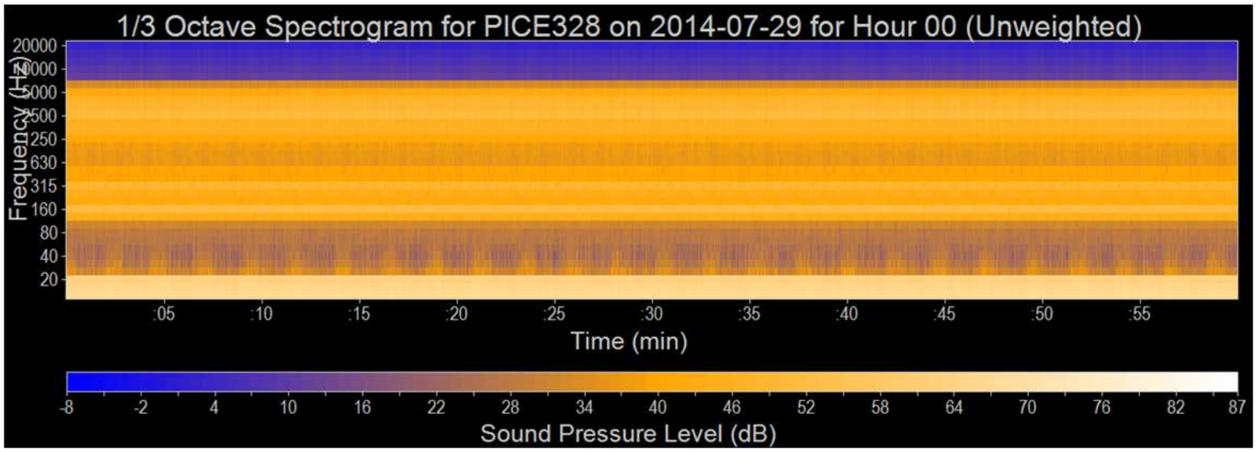

Figure 2.9. 1-hr spectrogram during noise treatment. Spectrogram generated from the MP3 recording made 10 m from the noise source (the outdoor speaker). This spectrogram highlights the consistent broadband properties of the noise signature.

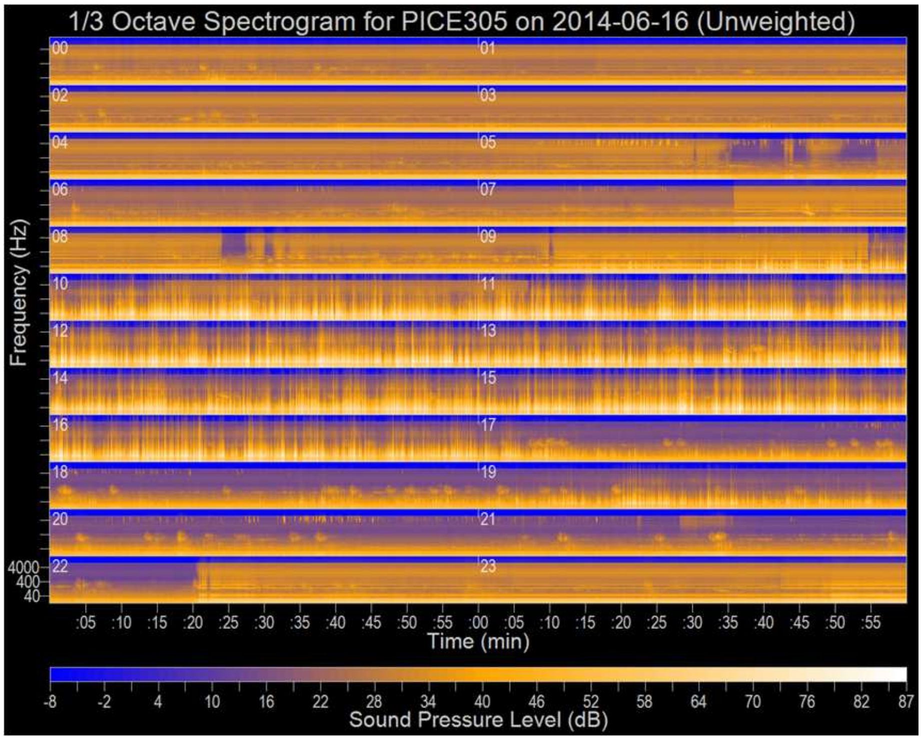

Figure 2.10. 24-hour spectrogram from site 305, where the overall nighttime L_{eq} was 38 during the recovery period (no added noise). Each line of the spectrogram represents 2 hours of audio recording. There is visual evidence of buzzing / hum on and off throughout the day. For example, the start of the hum is visible approximately 22 minutes into the 2200 hour. This type of buzzing / hum elevates the estimated L_{eq} and the duration is difficult to quantify, especially at sites with added noise (where this subtle hum is not visually identifiable in the spectrogram).

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CHAPTER 3

TEMPORAL TRENDS IN BAT ACTIVITY DURING AND AFTER EXPERIMENTAL NOISE EXPOSURE

Introduction

Understanding how wildlife respond to different types of anthropogenic disturbance has been an active area of research for decades. There is often interest in understanding how land use change and urbanization affect biodiversity, species of concern, and ecosystem services. In addition to the issues involving physical habitat destruction, there is a growing acknowledgement that wildlife habitat can be degraded and fragmented by disrupting the sensory environment (Barber et al. 2010, Van Dyck 2012).

Anthropogenic light pollution and noise pollution are two of the most commonly studied sensory disturbances affecting wildlife. These sources of disturbance are often associated with urbanization and large scale physical alteration of wildlife habitat, but can also be more temporary. Recent review papers highlight the wide range of taxa that are affected by noise and light pollution. Newport et al. (2014) addressed both noise and light as sources of sensory disturbance within the context of urbanization, focusing on impacts to biodiversity. Laiolo (2010) looked more broadly at bioacoustics within the context of conservation, reporting that many human activities including chemical pollution and noise pollution affect animal acoustic communication and behavior. Slabbekoorn et al. (2010) emphasized underwater noise and resulting effects on aquatic species, while Barber et al. (2010) reviewed the impacts of chronic exposure to anthropogenic noise and effects on terrestrial species. Kight and Swaddle (2011) studied the impacts that noise can have on the neuroendocrine system, and the importance of considering physiological responses, along with behavioral responses to noise. In summarizing studies of anthropogenic noise pollution on both aquatic and terrestrial studies, Shannon et al.

(2015) emphasized gaps in the literature and provide suggestions for future research with an emphasis on standardizing acoustical metrics and assessing wildlife response to noise-source removal.

Noise and other sensory disturbances can be temporary, and in some instances, mitigation may be able to reduce the spatial extent of these disturbances. So, what happens when these sensory disturbances end? Most studies have emphasized the response of different species during a period of noise exposure (Williams et al. 2015), and the handful of experiments that have attempted to address the response of wildlife post-exposure did not find long-lasting impacts.

Jong et al. (2016) studied the effect of nighttime light exposure on great tits (*Parus major*) in a captive setting using the following exposure process: a two-week initial period of no nighttime lighting, followed by a month of nighttime light exposure, and finally a three-week recovery period. Jong et al. observed both behavioral and physiological responses to the light treatment, as well as a dose-response to varying light intensities with birds increasing nocturnal activity in response to the nighttime light treatment over several days. Interestingly, the transition from higher nighttime activity levels during the light treatment disappeared immediately when birds entered the third phase and supplemental light was turned off. This type of rapid recovery has been observed in response to cessation of experimental noise treatments as well.

Bruintjes et al. (2016) studied the ventilation rate of European eels and European seabass during and after exposure to experimental noise treatments. Increased ventilation rate is a sign of stress, and in both species, ventilation rates increased during the two-minute period of noise exposure. In the seabass, ventilation rates returned to normal within two minutes after the noise exposure ended. For the eels, ventilation rates began decreasing immediately after the noise treatment ended, but had not fully recovered after two minutes. In addition to these changes in physiology during and after noise treatments, Bruintjes et al. (2016) also observed changes in behavioral response to predator stimuli.

Another recent study (Klett-Mingo et al. 2016) found that great tits (*Parus major*) responded to aircraft take-off events by increasing vigilance during the noise event, and reducing vigilance afterward. They also observed that the percent time spent feeding decreased during the noise events, and increased immediately after. Observations were limited to 20-seconds prior to aircraft take-off, during the noise event, and for an additional 20-seconds post-exposure.

In 2012, Le Roux and Waas conducted a field experiment to investigate whether long-tailed bats in New Zealand reduced activity levels in the presence of aircraft noise. They conducted a series of five-minute playback experiments that projected the sound of aircraft taking off once per minute, and monitored bat activity with hand-held bat recorders. They also monitored bat activity prior to the playback, and for 10-minutes following the noise exposure. They found some evidence of reduced activity in the first five minutes after the noise treatment ended, but by 10 minutes post-exposure, an activity reduction was no longer significant (Le Roux and Wass 2012). Bats are highly mobile, and the brief reduction in activity may have been the result of localized and short-term noise avoidance.

The artificial light experiment by Jong et al. (2016) took place over several weeks, but the noise experiments by Bruinjes et al. (2016), Klett-Mingo et al. (2016), and Le Roux and Wass (2012) were only a matter of seconds or minutes in duration, followed by short-term observation post-exposure. These short-term recovery periods were long enough to observe recovery for the species being studied, but this is not always the case. There have been multiple recent calls for more studies that include longer noise treatments and that continue to investigate the post-exposure period (Shannon et al. 2015, Radford et al. 2016, Tennessen et al. 2016).

In this field experiment, I investigated the effect of a six-day continuous noise treatment on bat activity levels, coupled with a six-day recovery period post-exposure. To my knowledge, this is the first noise experiment to monitor a multi-day recovery period for any species in the field. Based on the results of the noise experiment described in Chapter 2, this chapter focuses on the three species of bats that responded negatively to a multi-day noise treatment (*Myotis ciliolabrum*, *Lasiurus cinareus*, and *Tadarida brasiliensis*) and one species (*Myotis evotis*) that did not show reduced activity levels in the presence of noise (see Chapter 2). In this chapter, I investigate the ability of these species to rebound in the six-day period following noise cessation. As part of a study examining the effects of noise from natural gas development on bat activity, I used a cross-over design to experimentally study the post-exposure response in activity levels of bats following a noise treatment. Specific questions focused on whether evidence of an effect existed and how activity levels rebounded post-treatment (e.g., was there an immediate, first night response or a temporal trend of increasing activity level during the recovery period?). The crossover (aka change-over) experimental design has been widely used in clinical trials and biomedical research (Grizzle 1965) as well as ecological field studies (e.g., Johnson et al. 2002, Swarthout and Steidl 2003, Tuttle et al. 2006). The classic two-period crossover experiment consists of two treatments, where each site is exposed to both treatments, and the order of the treatments is randomly assigned (Wallenstein and Fisher 1977).

Methods

Study area – Study sites were located in the Piceance Basin, on Bureau of Land Management lands in Rio Blanco County, Colorado, U.S.A. (39.98°N 108.20°W). The dominant vegetation included mixed native grasses and forbes, non-native grasses, common rabbitbrush (*Chrysothamnus nauseosus*), and big sage (*Artemisia tridentata*). The nearby landscape included a mosaic of grasses and shrubs, mixed with pinyon pine (*Pinus edulis*) and juniper (*Juniperus* sp.). Due to the combination of more open grass and shrub land habitat, pinion-juniper woodlands, and abundant cliff and rock outcropping for roosting, this area supports at least 16 species of bats (Pers. comm. Heather Sauls, BLM White River Field Office, Nov. 2014). Elevation throughout the study area ranged from 1750 m to 2150 m. Primary land uses included cattle ranching, hunting, natural gas extraction, and recreation. The landscape is semi-arid with a 30-year average annual rainfall of 44.3 cm from 1981 - 2010 (WRCC 2016), and topographically diverse with many dry drainages and ephemeral streams. In the study area, flowing water was uncommon in summer months, except in the White River, Piceance Creek, and Yellow Creek. Most other drainages were dry, but would occasionally collect moisture after a rain event.

Experimental design – Between June 7 and August 4, 2014, I used a two-period crossover design to test the effects of a noise treatment, drilling operations during natural gas well development, on bat activity levels over time. The order of the treatment sequence can be described as A:B or B:A. For this study, 'A' refers to no added noise, and 'B' refers to a noise treatment consisting of the projected recording of drilling noise (see section on 'noise treatment' below for details). A total of 12 sites were randomly assigned to the A:B sequence, and 13 sites were assigned to the B:A sequence. A criticism of the crossover design is that there can be effects from the first treatment that carry over into the second period of the trial (Johnson et al. 2002, Mills et al. 2009). The 'washout' time is essentially a post-exposure recovery period. Understanding the bat activity dynamics during this recovery period was of central interest, making this application of the crossover study a bit different from the traditional application of crossover designs.

Site selection – To improve the chances of encountering commuting and foraging bats, the 28 sites were located along 13 drainages in the study area. Drainages were identified in ArcGIS, and ground-truthed to ensure they met the following criteria: 1) located on BLM land, 2) drilling for natural gas was not currently taking place along the drainage, 3) over 1 km in length, and 4) at least one vehicle access point along the drainage for equipment drop off. Site locations were determined by drawing a random center point along the drainage in ArcGIS, and establishing sites 500 m in either direction (creating paired sites 1 km apart). Due to the high concentration of small 4x4 roads throughout the area, access to sites was not a significant limitation. A pair of A:B and B:A sites was assigned to each drainage, but due to equipment failures results are based on twenty-five sites (12 A:B and 13 B:A).

Noise treatment – Using a Type I calibrated Larson Davis Sound Pressure Level (SPL) Meter, I took SPL readings and recorded audio of the noise 113 m from an active natural gas drill rig. Access to drill sites is limited, and at this distance, I was at the edge of the physically disturbed construction site. I quantified the equivalent continuous sound pressure level (L_{eq}) in A-weighted decibels (dBA) for the duration of the recordings (all sound pressure metrics re 20 μPa). At each treatment site, I broadcast the drill rig noise recording, so the noise levels reaching the central microphone (10 m from the speaker) were an approximate match to noise levels 100 m from a drill rig. Community brand R.5 outdoor speakers, in conjunction with Kemo electronics 6-16-volt 40-watt amps projected the drill rig playback, and were powered using two Power-Sonic 12-volt 103-Amp-hour rechargeable batteries connected in parallel, which could power the speakers continuously for approximately 48 hours. To simulate actual drill rig activity, the noise playback was designed to run continuously for the duration of the treatment, other than during the brief time needed to change batteries (see Table 3.1 for summaries of noise levels). Site checks and battery changes always took place during the day, when bats were not active.

Acoustical monitoring – At all sites, I deployed two SM2BAT+ ultrasound detector-recorders ('bat detectors') with SMX-US omnidirectional microphones (Wildlife Acoustics, Maynard, MA) to monitor nightly bat activity. SM2BAT+ units recorded in 'triggered .wav mode.' In this recording mode, the bat detector monitors the rolling background ambient sound levels, and when an ultrasound signal exceeds a user-defined threshold level, the detector initiates a recording and creates a file in .wav format. The central microphone was connected to a bat detector using the highest available sampling rate of 384 kHz. The second bat detector recorded in stereo (two channels) at the maximum stereo sampling rate of 192 kHz. Using microphone extension cables, the left and right stereo microphones were spaced 5-6 m in opposite directions from the central mic to extend the acoustical sampling space. Other than the sampling rate, the settings were identical. For the digital settings, I used an 18-dB trigger level, 16-kHz digital high pass filter, a 2.0-second window minimum, and a 5.0-second window maximum. Using the analog filter switches, I followed Wildlife Acoustics' recommended settings (Wildlife Acoustics 2011), +36-dB gain, the analog high-pass filter set to 1 kHz, and microphone bias off. When using triggered .wav mode, the digital high pass filter defines the frequency band of interest, but does not actually filter out all lower frequencies (see Wildlife Acoustics 2011 for more details). Recordings were directly saved to SD cards in the SM2BAT+ units and transferred to external hard drives for later analysis. All bat detectors were scheduled to power on at 2000 hrs (before sunset) and run until 0600 hrs (after sunrise). For the units recording in stereo, the left and right channels monitor the background and trigger independently, however, once either channel triggers, a stereo recording from both channels is created. I used external 6V sealed lead acid 36-amp-hour batteries to power all SM2BAT+ recorders. These batteries consistently ran for at least 6 days before needing to be recharged and replaced. Ultrasound microphones were mounted at the top of large tripods approximately 2.15 m above ground. At this height, the ultrasound mics were above the vegetation. At each site, ultrasound recordings from both detectors were combined for analysis. See Appendix 3.1 for full details on recording equipment settings.

In addition to monitoring bat activity at the sites, I continuously recorded the audible acoustical spectrum using Zoom H2 digital audio recorders (Zoom North America, Ronkonkoma, NY), in MP3, 64-kbps mode. As described by Mennitt and Fristrup (2012), sound pressure levels can be estimated using Zoom H2 units in the field.

No animals were handled in this study, and all research described followed protocols approved by Colorado State University Institutional Animal Care and Use Committee (Protocol #12-3574A).

Audio processing – Ultrasound recordings (.wav files) were processed using SonoBat 3.2.1 West Suite (sonobat.com, Szewczak et al. 2011), which includes several tools for sorting and processing ultrasonic. Following the examples of others (e.g., Kalcounis et al. 2013, Kennedy et al. 2014, Parkins and Clark 2015), I passed all triggered .wav recordings through the SonoBat Batch Scrubber 5.4 before automated classification. The Batch Scrubber removed low quality calls, and files that did not contain bat call sequences. I used the "high grade" setting, and included calls in the 5-20 kHz frequency range. Prior to running the stereo files through the scrubber, I split the stereo files into separate left and right channel recordings using custom scripts written in R (R Core Team, 2013). The single-channel 384-kHz .wav recordings were batch scrubbed in their raw form. Call sequences that were not removed by the scrubbing process were passed to the SonoBat 3.2.1 West Batch Classifier and identified using two methods as described in Clement et al. (2014): 1) "by evaluating mean parameter values of acceptable calls within a sequence" and 2) "by evaluating classification agreement among individual calls within a sequence". To avoid misclassification, I followed the lead of other researchers (e.g., Kalcounis et al. 2013, Bunkley et al. 2015), and only accepted the classification decisions for call sequences when SonoBat reached a "consensus" decision, i.e., both methods agreed on the classification. Additionally, I required at least 4 pulses from a sequence to be included in all identification decisions. A ≥3 pulse criterion has been used in studies where calls were manually identified (e.g., Johnson et al. 2002; Hourigan et al. 2009). The addition of the 4-pulse criterion is more conservative than using the "consensus" decision alone and is intended to further reduce misclassification during the automated classification process. For species with fewer than 50 recordings identified using SonoBat, each call was manually vetted to visually identify diagnostic call features. For species with more than 50 identified recordings, a minimum of 20 call sequences were verified by manually vetting. For species that were confirmed through manual vetting, the results and models include all automated identifications, not only the subset of calls that were visually vetted.

MP3 files from the Zoom H2 units were processed using Program AUDIO2NVSPL and Acoustical Monitoring Toolbox (AMT, Damon Joyce, National Park Service Natural Sounds and Night Skies Division). Zoom H2 recordings were used to calculate aggregate acoustical metrics including A-weighted L_{90}, L_{50}, and L_{10} values during nighttime hours (2000 hrs to 0600 hrs) over the duration of each treatment. L_x levels refer to percentile-exceeded sound levels, for example, the L_{90} for a site represents the sound level that is exceeded 90 percent of the time (NPS 2013). For these types of wide bandwidth hourly metrics, Zoom H2 units can be accurate to within 1 dBA (Mennitt and Fristrup 2012).

Statistical analysis – After the automated species identification process in SonoBat, count data for *M. ciliolabrum*, *L. cinareus*, *T. brasiliensis,* and *M. evotis* were summarized into nightly encounter histories for each site. I incorporated these species-specific nightly counts, where each identified recording is considered a count, into the model described below.

Using a Bayesian modeling approach (Hobbs and Hooten 2015), I modeled bat activity levels over time from spatially and temporally replicated counts using a hierarchical binomial-Poisson mixture model. This model implementation first appeared in the ecological literature to estimate abundance from point counts, while accounting for imperfect detection (Royle 2004, Kéry and Schaub 2012). An assumption is that individuals are not counted twice during a survey, but individual bats are not identifiable using current acoustical monitoring techniques. For my study, I interpret the results as 'bat activity levels' rather than 'abundance.'

In my model, bat activity levels (N_{ij}) for each site (i) and survey night (j) were described as $N_{ij} \sim$ Poisson (λ_{ij}). Due to imperfect detection of bats via acoustical monitoring methods, true activity level cannot be observed directly, thus observed counts (y_{ij}) of each species were incorporated as $y_{ij} \sim$ Binomial(N_{ij}, p), where p is detection probability. To incorporate the effects of noise treatment over time, the effect of the treatment, and potential interactions between the two, I used a log link to model mean activity level, λ_{ij} as

$\log(\lambda_{ij}) = \beta_0+\beta_1*\text{night}_j+\beta_2*\text{ treatment}_j +\beta_3*\text{ night}_j * \text{treatment}_j$.

Vague normal priors were used for all coefficients. I used Beta(a,b) priors for detection, where 'a' and 'b' are the shape parameters that define the beta distribution. Although vague or 'non-informative' priors are often suggested (e.g., Royle and Dorazio 2008, Kéry 2010, Kéry and Schaub 2012), there is a growing number of ecological studies that incorporate existing ecological knowledge (Morris et al. 2013, Hooten and Hobbs 2015, Linden and Roloff 2015, Morris et al. 2015). Using an informative prior from pilot studies can increase the precision of a model, without reducing model accuracy (Morris et al 2015), thus species-specific shape parameters ‘a’ and ‘b’ were assigned based on information from a 2012 pilot study (Appendix 3.2).

The binomial-Poisson mixture models were implemented using the software program WinBUGS (Spiegelhalter et al. 2003) and the R2WINBUGS package in R (R Core Team 2013). Data from each species was analyzed separately, using the same model structure. To fit the above model, I obtained 5 parallel MCMC chains for 300,000 iterations, with a thinning rate of 5, and the first 200,000 iterations were discarded as burn-in. Chain mixing and convergence were assessed using the Gelman-Rubin statistic ($\hat{R}$) provided by WinBUGS (Brooks and Gelman 1998). $|\hat{R}|$ deviated from 1 by no more than 0.05 for all parameters. The WinBUGS specifications for the models described above are in Appendix 3.3.

Covariates –The nightly noise treatment covariate was included in the models as either 0 (nights without noise treatment) or 1 (nights with noise treatment). Survey night (standardized) over the entire length of the A:B or B:A sequence was also included, along with the noise*night interaction term.

Derived metrics – Within the WinBUGS model code, nightly bat activity level estimates at each site (*N*[i,j]) are saved for each model iteration, after burn-in. I summarized the average nightly activity levels for all control nights (N.ctl), treatment nights (N.trt), and recovery nights (N.rec). N.trt was calculated separately for the A:B and B:A sequences. For the A:B sequence, I calculated $\Delta N_{A:B}$ = N.trt – N.ctl to estimate the magnitude of the difference in activity levels between the two periods in the sequence. Using the same approach, I calculated $\Delta N_{B:A}$ = N.rec – N.trt during the B:A sequence.

Results

Nighttime (2000 hrs to 0600 hrs) ambient sound levels at control sites ranged from 25 to 45 L_{eq} in dBA over the duration of the control period, and at recovery sites levels ranged from 22 to 41 L_{eq} in dBA. At treatment sites, nighttime levels ranged from 56 to 77 L_{eq} in dBA over the duration of the experiment (Table 3.1). The drill rig noise recording that was projected at all treatment sites had a 15 minute wideband L_{eq} of 70.6 dBA, as measured 113 m from the source.

Based on the derived summary statistics from the binomial-Poisson mixture models, the greatest average number of nightly bat calls for all species was recorded during the control period in the A:B sequence. At sites in the A:B sequence, $\Delta N_{A:B}$ was -5.37, -4.15, and -2.58 for *M. ciliolabrum*, *L. cinareus*, and *T. brasiliensis*, respectively; however, 95% credible intervals (CIs) overlap in all cases (Table 3.2, Figure 3.1). There was no change in activity level for *M. evotis*, $\Delta N_{A:B}$ of 0.0 (Table 3.2). During the B:A sequence, $\Delta N_{B:A}$ was 2.19, 0.40, 0.88, and 0.20 for *M. ciliolabrum*, *L. cinareus*, *T. brasiliensis*, and *M. evotis*, respectively (Table 3.3), but 95% CIs overlap between the 'B' and 'A' periods (Table 3.3, Figure 3.1).

In the A:B model, 95% credible intervals did not overlap with zero for any of the covariate estimates for *M. ciliolabrum*, suggesting that the noise treatment ('noise'), the chronological time throughout the entire A:B sequence ('night'), and the interaction between 'night' and 'noise' all affected activity levels (Table 3.4, Figure 3.2). For *L. cinareus* and *M. evotis*, the interaction term was the only covariate estimate that did not include 0 in the 95% CIs. This interaction was negative for *L. cinareus*, and positive for *M. evotis* (Table 3.4, Figure 3.3 and Figure **3.4**). Although the nightly average activity level appears to decline over time (Figure 3.4), none of the covariate estimates were significant for *T. brasiliensis* (Table 3.4). The mean estimate for detection rate (*p*) was 0.40, 0.20, 0.38, and 0.30 for *M. ciliolabrum*, *L. cinareus*, *T. brasiliensis*, and *M. evotis*, respectively (Table 3.4).

In the B:A model, the interaction term 'night*noise' (0.76) was the only covariate estimate that did not have a 95% CIs overlapping zero for *M. ciliolabrum* (Table 3.5, Figure 3.2). The 95% CIs for 'noise,' 'night,' and the interaction term all overlapped with zero for *L. cinareus*, *T. brasiliensis*, and *M. evotis* (Table 3.5, Figure 3.3,Figure **3.4**, andFigure **3.5**).

Discussion

A few wildlife field studies have isolated noise experimentally in the lab (Schaub et al. 2008, Siemers and Schaub 2011, Bunkley and Barber 2015, Luo et al. 2015) and in the field (Blickley et al. 2012, McClure et al. 2013, Ware et al. 2015), to separate noise from confounding sources of disturbance. Fewer have monitored a post-exposure period (but see Le Roux and Wass 2012, Bruinjes et al. 2016, Klett-Mingo et al. 2016). This study provides novel information about a multi-day post-exposure period following noise cessation. In previous studies that addressed the post-exposure period, noise treatments or events were relatively short in duration (a few minutes), and the corresponding recovery period was monitored over a similarly brief timeframe. This study differs in both the duration of the noise treatment (continuous noise over six days/nights), and in the duration of the post-exposure monitoring period (also six days/nights).

Of the four bat species included in this study, not all responded to the noise treatment, and for some species, the activity level patterns are not easily interpreted. There was little indication that noise affected the activity levels of *M. evotis* at the time scale monitored in this study. *M. evotis*, or other species in the area, possibly avoid noise over a much shorter duration, as observed by Le Roux and Waas (2012). Similar to Bunkley et al. (2015) who found a 40%

decrease in activity for *T. brasiliensis*, *T. brasiliensis* activity was 44% lower during the treatment period in the A:B sequence. However, estimated activity levels for *T. brasiliensis* steadily declined throughout the control period, as well as throughout the treatment period, making it difficult to determine what role noise was playing in *T. brasiliensis* activity. *T. brasiliensis* activity levels did increase slightly after the noise treatment ended, but none of the treatment covariates were significant for *T. brasiliensis* during the A:B or B:A sequences.

M. ciliolabrum activity levels also declined over the control period in the A:B sequence. This initial decline occurred prior to the noise treatment, indicating that other factors were influencing *M. ciliolabrum* activity levels. At the onset of the noise treatment, *M. ciliolabrum* activity levels did decline more steeply, but then increased throughout the treatment period. For *M. ciliolabrum*, the 'night' and 'noise' parameter estimates were both negative in the A:B sequence. The negative 'night' parameter indicates that activity levels decreased from the beginning of the 'A' period to the end of the 'B' period, and the negative 'noise' parameter indicates that activity levels were lower during the noise treatment period, as compared to the control period. During the A:B sequence, the 'night*noise' interaction term was positive, indicating that within the noise treatment period, activity levels increased. During the B:A sequence, the 'night*noise' interaction term was also positive, with a 95% CIs that did not overlap with zero. The steady increase in activity level throughout the treatment period in both the A:B and B:A sequences may be a sign of temporary avoidance of a novel stimulus, as suggested by Luo et al. (2015).

The activity patterns of *L. cinareus* provide the most convincing evidence of noise avoidance. Activity levels of *L. cinareus* were fairly constant throughout the control period, dropped at the onset of the noise treatment in the A:B sequence, and continued to decline throughout the ‘B’ treatment period. *L. cinareus* uses relatively low frequencies for echolocation (<35kHz), and bats in the ‘low frequency’ group may be more affected by anthropogenic noise (Bunkley et al. 2015). Furthermore, *L. cinareus* activity levels did not show signs of recovery after the noise treatment ended in the B:A sequence. This suggests that after the six-day post-exposure period, there were still lingering effects of noise on *L. cinareus* activity levels**.**

Playback experiments are a useful way to isolate and replicate the effects of anthropogenic noise (Holles et al. 2016), and understanding the noise levels that elicit a response from each species can help inform management and mitigation efforts (Shannon et al. 2015). Accurately describing the ambient sound conditions and noise exposure levels at treatment sites is often neglected or inadequate in terrestrial studies involving noise and wildlife (Shannon et al. 2015). Leaving costly sound pressure level meters in the field for long durations is not common in wildlife studies, but it is possible to estimate sound levels using less costly digital audio recorders as I did in this study. However, weatherproofing (and livestock proofing) audio equipment in the field remains a challenge. Although I outfitted all digital audio recorders with a rain covering and a windscreen to prevent damage from weather events, some of the audio files contain distorted recordings that included electrical buzzing (Figure 3.6 andFigure **3.7**). This buzzing is visually (and audibly) identifiable at times when noise or high wind is not present, but is not possible to identify during treatment periods when the noise treatment overwhelms the audio and the spectrograms (see Figure 3.8 for spectrogram during noise treatment). Since these audio anomalies affect the acoustical metrics that were estimated using the digital audio recordings, the sound levels reported in Table 3.1 can only be considered rough estimates of the true acoustical conditions, and the experimental noise was treated as binary (noise or no noise) rather than a continuous variable.

Experimental manipulations make it possible to separate the impacts of various sensory stressors, which is not typically possible in observational studies (Lewandowski et al. 2016, Slabbekoorn 2016), but noise playback experiments using small outdoor speakers cannot replicate the full range of frequencies, or the amplitude of the original source (Holles et al. 2016). Noise from an operating natural gas drill rig is continuous and intense during the drilling phase, which can last for several weeks to several months. The noise and light from these rigs can travel miles from the source, and these sensory cues are known to affect a wide range of taxa (Newport et al. 2014). For example, mule deer have been shown to avoid areas near active drilling sites by as much as 600 m (Northrup et al 2015). At this distance, it is likely that avoidance is due to sensory cues, including noise and light pollution, since this is well beyond the area of physical disturbance. After isolating noise experimentally, the results from this study suggest drilling noise as a cause for localized avoidance, at least for some bat species. In addition to responding to noise, bats also respond to artificial light. The response by bats to light pollution is species-specific, with some species avoiding light sources, while others are attracted to them (Stone et al. 2012). Further study is needed to determine how bats respond to the interaction of noise and light as experienced near actual natural gas drill rigs.

Previous studies investigating the recovery period after noise exposure found little evidence of lingering treatment effects beyond a few minutes in several taxa (Le Roux and Wass 2012, Bruinjes et al. 2016, Klett-Mingo et al. 2016). For most species, however, very little is known about the response following noise cessation (Shannon et al. 2015). In this study, *L. cinareus* activity levels dropped during the noise treatment period, and failed to rebound during the post-exposure period. While other factors may also have been influencing these activity patterns, these results suggest that *L. cinareus* may be more sensitive to the effects of noise than other focal species in this study. In addition to urbanization and other landscape level land-use changes, *L. cinareus* has been disproportionally affected by wind energy development. Tree roosting species, including *L. cinareus*, account for over three-quarters of fatalities at wind energy sites (Arnett et al. 2008, Arnett and Baerwald 2013, Cryan et al. 2014). Though the impact of noise on *L. cinareus* is non-lethal at the levels I investigated, noise is an added stressor for this species, and others. For species that do respond to noise pollution, the recovery time after noise ends will certainly differ by species, and as I show here, recovery is not always rapid. Future wildlife studies that assess periods of post-exposure could contribute meaningfully to this area of research, and aid in the development of conservation and mitigation efforts.

Table 3.1. Site summary information. All Lx values are percentile-exceeded sound levels in dBA over the duration of the A or B treatment.

year	site name	sequence	A dates	B dates	Leq.A	Leq.B	L90.A	L50.A	L10.A	L90.B	L50.B	L10.B
2014	300	A:B	7/1-7/8	7/8-7/14	25	77	23.2	23.6	25.6	76.7	77.4	78.1
2014	302	A:B	6/24-6/30	6/30-7/6	31	74	24.9	26.2	28.2	73.3	73.8	74.4
2014	304	A:B	6/8-6/14	6/14-6/20	27	74	23.7	24.3	27	73.4	74.2	74.8
2014	309	A:B	6/22-6/28	6/28-7/4	30	76	23.7	24	25.4	75.1	75.6	76.3
2014	310	A:B	6/7-6/14	6/14-6/20	36	74	23.9	24.8	27.2	73.4	74	74.6
2014	312	A:B	7/6-7/13	7/13-7/19	37	73	23.9	24.3	26.3	72.1	72.7	73.3
2014	317	A:B	6/23-6/29	6/29-7/5	26	72	23.7	24.5	27.2	71.6	72.1	73
2014	321	A:B	7/16-7/22	7/22-7/28	39	76	30.5	35.5	39.3	74.8	75.3	76.1
2014	322	A:B	7/8-7/14	7/14-7/20	43	72	26.7	27.5	28.4	71.8	72.2	72.5
2014	325	A:B	7/9-7/15	7/15-7/21	44	63	24.1	24.4	25.7	61.9	62.7	63.4
2014	326	A:B	7/22-7/28	7/28-8/3	36	71	31.8	34.2	37.1	70.4	70.8	71.3
2014	329	A:B	7/23-7/29	7/29-8/4	33	69	27.1	28.1	29.4	69	69.9	70.6
2014	301	B:A	7/8-7/14	7/2-7/8	37	73	26.3	27.2	28.8	72.7	73.2	73.7
2014	303	B:A	6/30-7/6	6/24-6/30	30	75	25.8	26.9	28.9	74.9	75.3	75.8
2014	305	B:A	6/14-6/20	6/8-6/14	38	75	26.1	27.3	34.4	75	75.4	75.9
2014	308	B:A	6/28-7/4	6/22-6/28	27	76	24.4	24.8	25.8	74.8	75.5	76.1
2014	311	B:A	6/14-6/20	6/7-6/14	29	75	24.4	25.2	27.9	74.9	75.5	76
2014	313	B:A	7/13-7/19	7/7-7/13	32	77	24.9	25.6	27.8	76	76.6	77.2
2014	314	B:A	6/16-6/22	6/9-6/16	31	68	24.7	25.2	27.9	67.8	68.8	69.7
2014	316	B:A	6/29-7/5	6/23-6/29	31	75	26.9	28.2	31.1	74.3	74.7	75.2
2014	320	B:A	7/22-7/28	7/16-7/22	37	71	23.9	24.9	26.6	70.8	71.4	71.9
2014	323	B:A	7/14-7/20	7/8-7/14	22	75	18.8	19	19.3	74.1	74.6	75.2
2014	324	B:A	7/15-7/21	7/9-7/15	33	64	24.6	24.8	25.6	62.8	63.4	64.2
2014	327	B:A	7/28-8/3	7/22-7/28	41	77	35.9	38.3	40.8	76	76.7	77.5
2014	328	B:A	7/29-8/4	7/23-7/29	34	56	25.7	27.5	30.8	55	56	56.9

Table 3.2. Average nightly bat activity levels by species over all control nights (N.ctl) and treatment nights (N.trt) in the A:B model. CI.ctl is the 95% credible interval for the control period, and CI.trt is the 95% credible interval for the treatment period. $\Delta N_{A:B}$ is defined as N.trt – N.ctl.

species	N.ctl	N.trt	$\Delta N_{A:B}$	% change	95% CI.ctl	95% CI.trt
M. ciliolabrum	11.81	6.44	-5.37	-45.46	(4, 38)	(1, 16)
L. cinareus	7.75	3.60	-4.15	-53.51	(1, 21)	(0, 13)
T. brasiliensis	5.88	3.30	-2.58	-43.81	(1, 15)	(0, 9)
M. evotis	4.12	4.12	0.00	0.00	(0, 12)	(0, 12)

Table 3.3. Average nightly bat activity levels by species over all treatment nights (N.trt) and recovery nights (N.rec) in the B:A model. CI.trt is the 95% credible interval for the treatment period, and CI.rec is the 95% credible interval for the recovery period. $\Delta N_{B:A}$ is defined as N.rec – N.trt.

species	N.trt	N.rec	$\Delta N_{B:A}$	% change	95% CI.trt	95% CI.rec
M. ciliolabrum	5.09	7.28	2.19	43.21	(1, 15)	(2, 17)
L. cinareus	3.89	4.29	0.40	10.05	(0, 11)	(0, 12)
T. brasiliensis	2.51	3.39	0.88	35.01	(0, 6)	(0, 10)
M. evotis	2.49	2.69	0.20	8.04	(0, 7)	(0, 8)

Table 3.4. Mean parameter estimates from A:B models, with 95% credible intervals (CI), Estimates on the log scale, where nightly activity level is described as: $\log(\lambda_{ij}) = \beta_0+\beta_1*\mathrm{night}_j+\beta_2*\ \mathrm{treatment}_j+\beta_3*\ \mathrm{night}_j * \mathrm{treatment}_j$ for *Myotis ciliolabrum* – Myci, *Lasiurus cinareus* - Laci, *Tadarida brasiliensis* - Tabr and *Myotis evotis* –Myev.

parameter	species	estimate	95% CI
intercept	Myci	2.28	(2.05, 2.51)
night	Myci	-0.23	(-0.42, -0.04)
noise	Myci	-0.73	(-1.12, -0.34)
night*noise	Myci	0.56	(0.18, 0.94)
detection	Myci	0.40	(0.35, 0.46)
intercept	Laci	2.31	(1.59, 3.15)
night	Laci	0.27	(-0.10, 0.65)
noise	Laci	-0.15	(-0.85, 0.52)
night*noise	Laci	-1.31	(-2.14, -0.51)
detection	Laci	0.20	(0.07, 0.37)
intercept	Tabr	1.63	(1.31, 1.94)
night	Tabr	-0.17	(-0.45, 0.11)
noise	Tabr	0.00	(-0.52, 0.51)
night*noise	Tabr	-0.36	(-0.91, 0.20)
detection	Tabr	0.38	(0.32, 0.44)
intercept	Myev	1.85	(0.66, 3.59)
night	Myev	-0.44	(-0.85, -0.03)
noise	Myev	0.03	(-0.67, 0.71)
night*noise	Myev	0.8	(0.15, 1.45)
detection	Myev	0.30	(0.12, 0.53)

Table 3.5. Mean parameter estimates from B:A models, with 95% credible intervals (CI), Estimates on the log scale, where nightly activity level is described as: $\log(\lambda_{ij}) = \beta_0+\beta_1*\text{night}_j+\beta_2*\text{ treatment}_j+\beta_3*\text{ night}_j * \text{treatment}_j$ for *Myotis ciliolabrum* – Myci, *Lasiurus cinareus* - Laci, *Tadarida brasiliensis* - Tabr and *Myotis evotis* –Myev.

parameter	species	estimate	95% CI
intercept	Myci	2.12	(1.82, 2.40)
night	Myci	-0.17	(-0.44, 0.11)
noise	Myci	-0.05	(-0.43, 0.34)
night*noise	Myci	0.76	(0.32, 1.20)
detection	Myci	0.40	(0.35, 0.46)
intercept	Laci	1.29	(0.36, 2.34)
night	Laci	0.04	(-0.52, 0.61)
noise	Laci	0.05	(-0.73, 0.83)
night*noise	Laci	0.10	(-0.72, 0.92)
detection	Laci	0.19	(0.07, 0.37)
intercept	Tabr	1.09	(0.64, 1.51)
night	Tabr	0.14	(-0.27, 0.55)
noise	Tabr	-0.23	(-0.85, 0.39)
night*noise	Tabr	-0.19	(-0.41, 0.44)
detection	Tabr	0.38	(0.32, 0.44)
intercept	Myev	0.88	(0.02, 1.86)
night	Myev	0.02	(-0.54, 0.57)
noise	Myev	-0.19	(-0.98, 0.59)
night*noise	Myev	-0.17	(-0.98, 0.64)
detection	Myev	0.30	(0.12, 0.53)

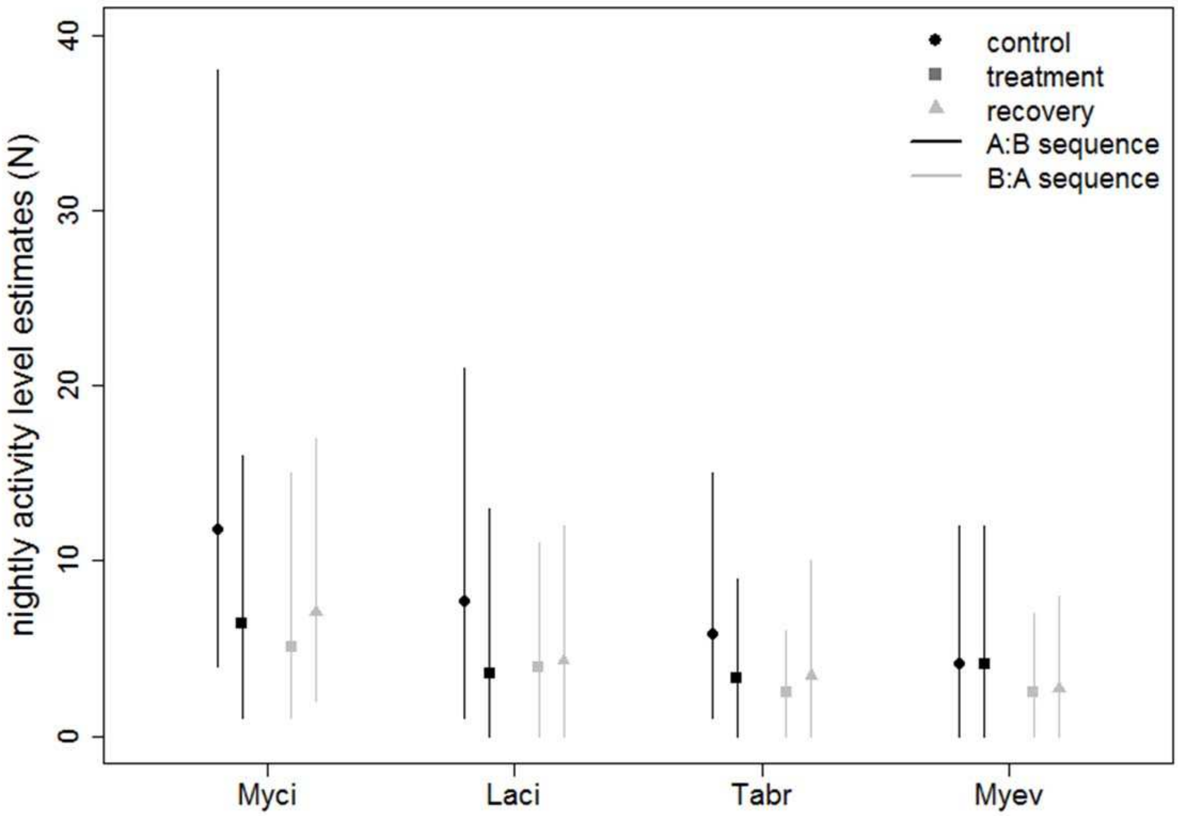

Figure 3.1. Average nightly activity levels for each treatment period, means with 95% credible intervals. Mean values calculated by species, over all sites and all night during the given treatment. Points and lines in black show the N.ctl and N.trt from the A:B sequence, and points and lines in grey show the N.trt and N.rec from the B:A sequence. *Myotis ciliolabrum* – Myci, *Lasiurus cinareus - Laci*, *Tadarida brasiliensis* - Tabr and *Myotis evotis* –Myev.

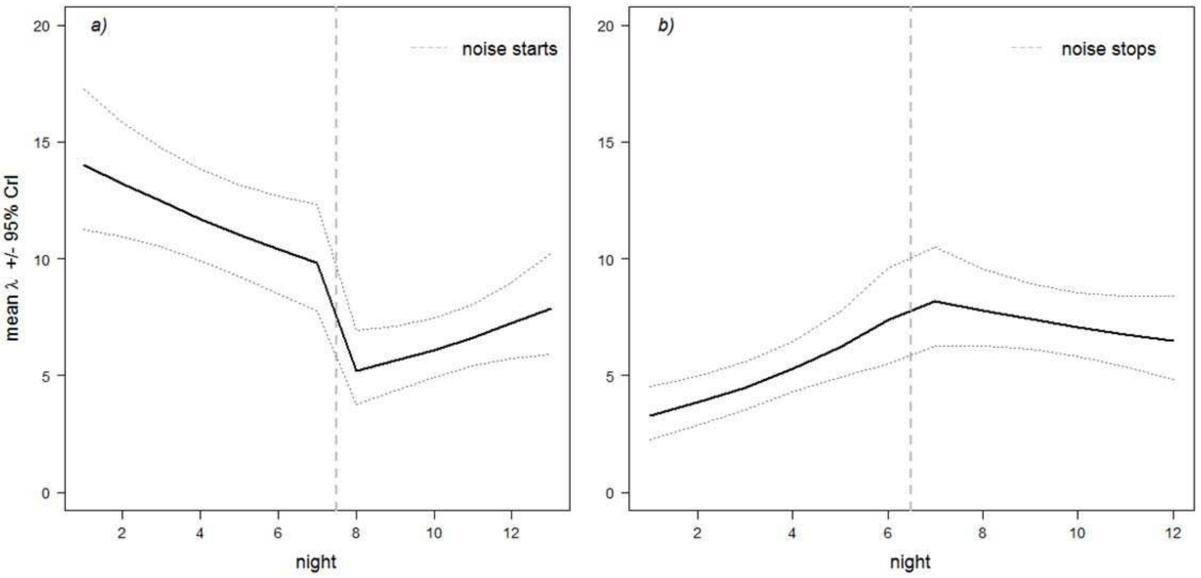

Figure 3.2. Estimated mean nightly activity levels averaged over all sites (mean $\lambda_{.j}$) with 95% credible intervals (CI) for *Myotis ciliolabrum*. Estimates from the A:B model show control nights prior to the dashed line, and treatment nights after (a), estimates from the B:A model show treatment nights before to the dashed line, and recovery nights after (b).

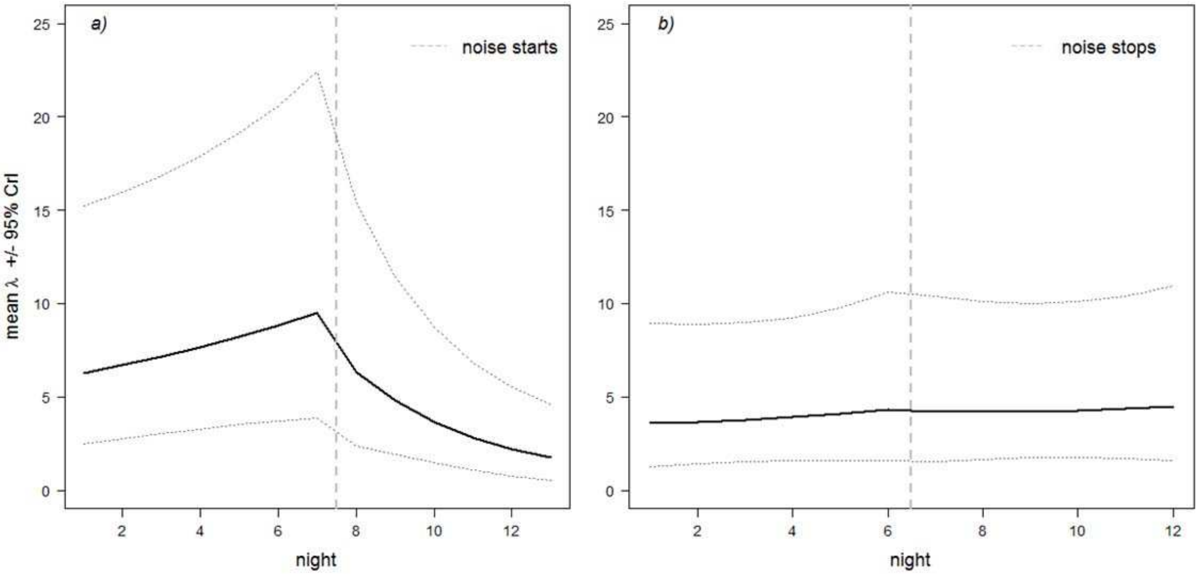

Figure 3.3. Estimated mean nightly activity levels averaged over all sites (mean $\lambda_{.j}$) with 95% credible intervals (CI) for *Lasiurus cinereus*. Estimates from the A:B model show control nights prior to the dashed line, and treatment nights after (a), estimates from the B:A model show treatment nights before to the dashed line, and recovery nights after (b).

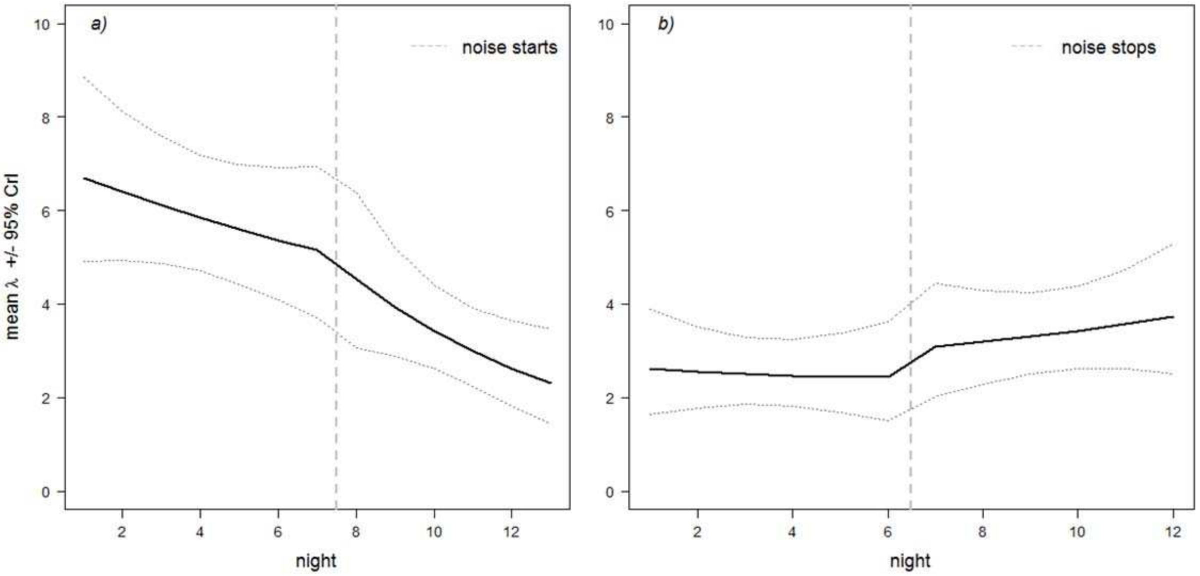

Figure 3.4. Estimated mean nightly activity levels averaged over all sites (mean $\lambda_{.j}$) with 95% credible intervals (CI) for *Taderida brasiliensis*. Estimates from the A:B model show control nights prior to the dashed line, and treatment nights after (a), estimates from the B:A model show treatment nights before to the dashed line, and recovery nights after (b).

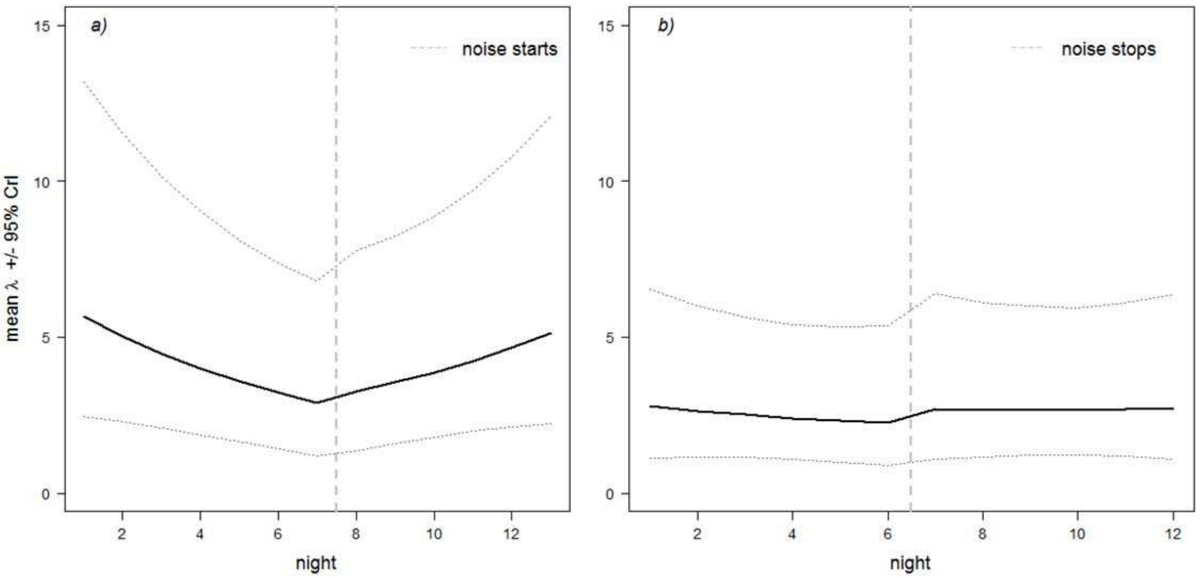

Figure 3.5. Estimated mean nightly activity levels averaged over all sites (mean $\lambda_{.j}$) with 95% credible intervals (CI) for *Myotis evotis*. Estimates from the A:B model show control nights prior to the dashed line, and treatment nights after (a), estimates from the B:A model show treatment nights before to the dashed line, and recovery nights after (b).

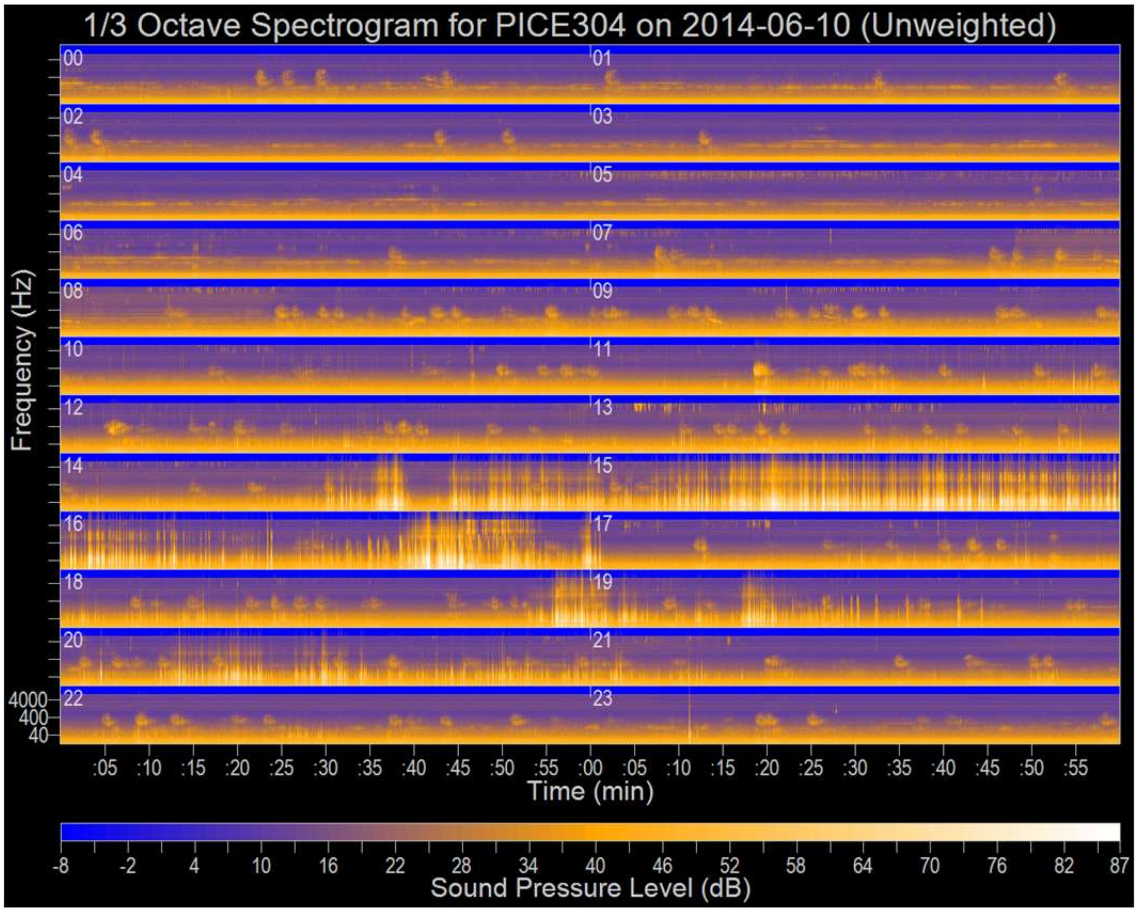

Figure 3.6. 24-hour spectrogram at site 304. During the control period the overall nighttime Leq was 27 dBA

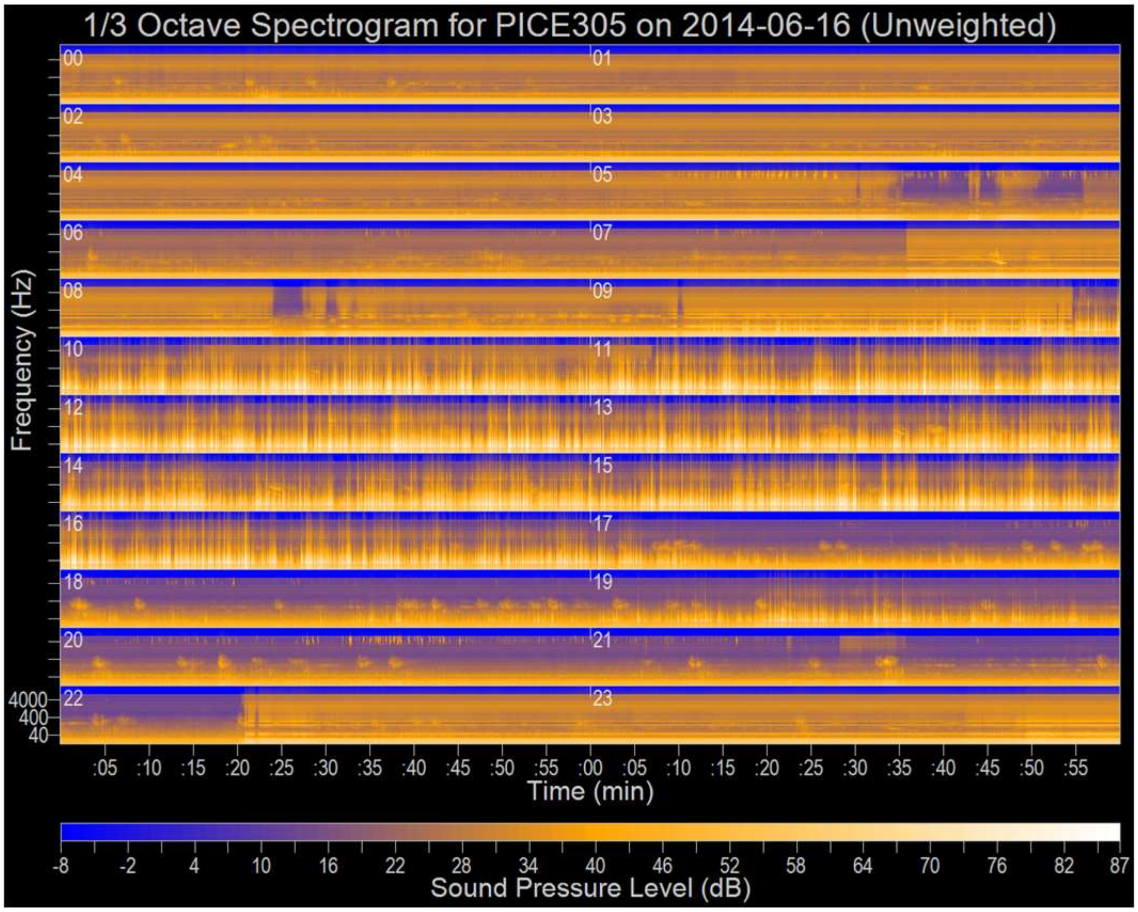

Figure 3.7. 24-hour spectrogram from site 305, where the overall nighttime equivalent continuous sound pressure level (Leq) was 38 during the recovery period. Evidence of electrical buzzing off and on throughout the day. This would elevate the estimate Leq. However, natural events like high wind can also contribute to higher Leq.

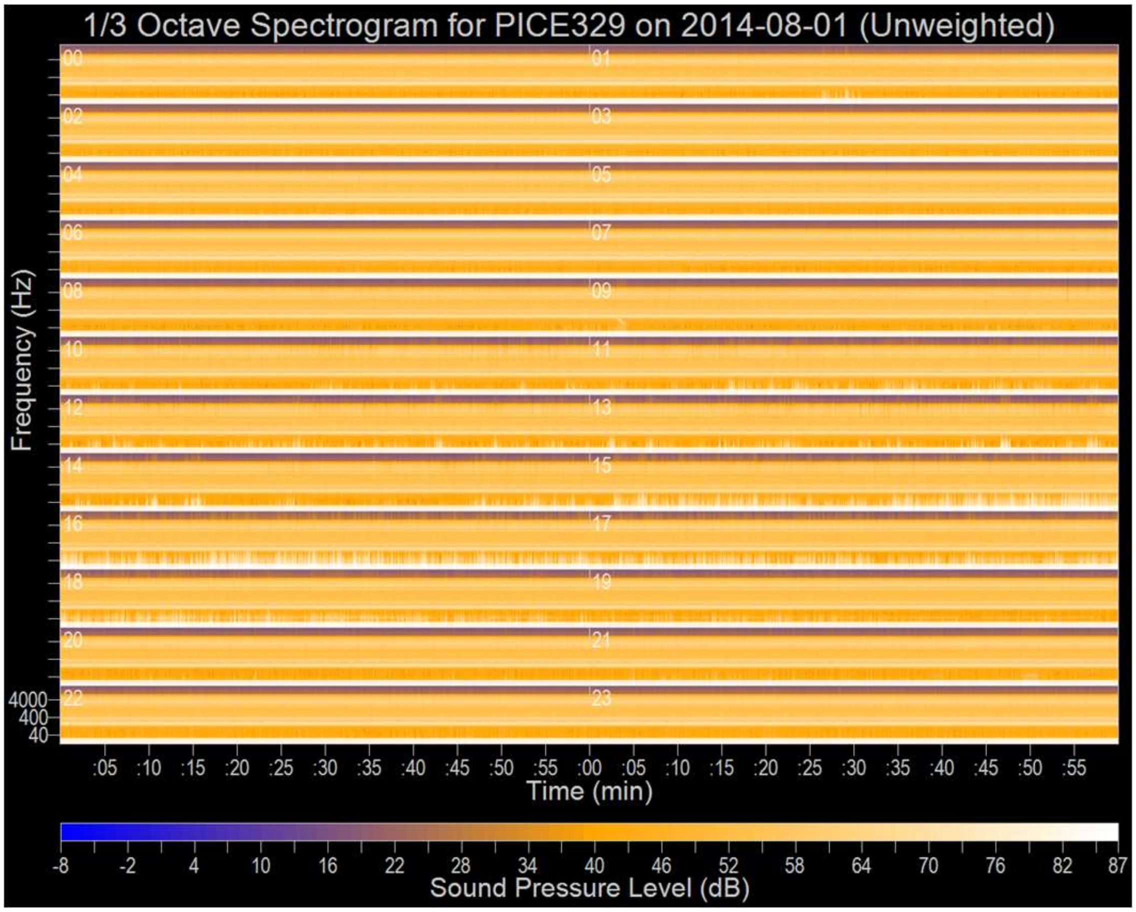

Figure 3.8. 24-hour spectrogram from site 329, where the estimated overall nighttime equivalent continuous sound pressure level (Leq) during the treatment was 69.0 dBA.

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APPENDIX 2.1 – CHAPTER 2 SITE INFORMATION

pair	Year	trt	#nights	dates	#IDed calls	# spp	L90	L50	L10	site name
1	2013	control	5	7/11-7/16	302	8	24	24.5	26.2	202
1	2013	noise	5	7/11-7/16	20	5	76.5	77	77.8	203
2	2013	control	7	6/19-6/26	77	6	23.6	24.2	26.3	204
2	2013	noise	7	6/19-6/26	85	7	71.3	71.8	72.5	205
3	2013	control	4	7/5-7/9	4	4	24	24.7	27.6	206
3	2013	noise	4	7/5-7/9	36	5	73.3	73.9	74.7	207
4	2013	control	6	7/18-7/24	262	9	26.3	28.2	31.5	210
4	2013	noise	6	7/18-7/24	136	6	76.9	77.5	78.3	211
5	2013	control	6	7/19-7/25	623	11	26.1	27.1	29.6	212
5	2013	noise	6	7/19-7/25	426	7	73.9	74.6	75.4	213
6	2013	control	6	7/20-7/26	340	7	24.1	25.2	28	214
6	2013	noise	6	7/20-7/26	242	7	63.2	63.8	64.6	215
7	2013	control	6	7/26-8/1	129	7	31.9	34.4	38	216
7	2013	noise	6	7/26-8/1	66	6	76.4	76.9	77.6	217
8	2013	control	6	7/27-8/2	200	9	25.1	26.9	29.6	218
8	2013	noise	6	7/27-8/2	115	7	74.3	74.9	75.8	219
9	2013	control	6	7/28-8/3	204	7	24.1	25.1	28.4	220
9	2013	noise	6	7/28-8/3	92	7	79.3	79.9	80.6	221
10	2013	control	6	8/3-8/9	373	9	31.9	33.9	38.3	222
10	2013	noise	6	8/3-8/9	150	8	71.4	72.3	73.1	223
1	2014	control	6	6/24-6/30	565	9	24.9	26.2	28.2	302
1	2014	noise	6	6/24-6/30	151	10	74.9	75.3	75.8	303
2	2014	control	6	6/8-6/14	99	7	23.7	24.3	27	304
2	2014	noise	6	6/8-6/14	69	7	75	75.4	75.9	305
3	2014	noise	6	6/22-6/28	63	7	74.8	75.5	76.1	308
3	2014	control	6	6/22-6/28	182	8	23.7	24	25.4	309
4	2014	control	7	6/7-6/14	240	7	23.9	24.8	27.2	310
4	2014	noise	7	6/7-6/14	184	7	74.9	75.5	76	311
5	2014	noise	6	6/23-6/29	287	8	74.3	74.7	75.2	316
5	2014	control	6	6/23-6/29	239	8	23.7	24.5	27.2	317
6	2014	noise	6	7/16-7/22	279	10	70.8	71.4	71.9	320
6	2014	control	6	7/16-7/22	248	7	30.5	35.5	39.3	321
7	2014	control	6	7/8-7/14	265	8	26.7	27.5	28.4	322
7	2014	noise	6	7/8-7/14	249	8	74.1	74.6	75.2	323
8	2014	noise	6	7/9-7/15	88	7	62.8	63.4	64.2	324
8	2014	control	6	7/9-7/15	77	6	24.1	24.4	25.7	325
9	2014	control	6	7/22-7/28	229	7	31.8	34.2	37.1	326
9	2014	noise	6	7/22-7/28	115	8	76	76.7	77.5	327
10	2014	noise	6	7/23-7/29	51	8	NA	56	56.9	328
10	2014	control	6	7/23-7/29	51	7	27.1	28.1	29.4	329

APPENDIX 2.2 – CHAPTER 2 EQUIPMENT SETTINGS

SM2BAT+ settings:
Microphones: SMX-US by Wildlife Acoustic
Sampling rate: 192 kHz (for stereo recorders) or 384 kHz (for single-channel recorders)
Recordings mode: Triggered .wav (no compression)
Trigger level: 18 dB
Digital high pass filter: 16 kHz
Min. recordings window: 2 seconds
Max. recording window: 5 seconds
Analog filter switches—
Gain: +36
Analog high-pass filter: 1kHz
Mic bias: OFF
Nightly recording hours: 20:00:00 – 05:59:59

Zoom H2 settings:
Mic gain: M/MID
Mics: 2-channel
LO CUT: OFF
REC MODE: MP3 64
AGC/COMP: OFF
MONITOR: OFF
PLUG-IN: OFF
PRE REC: OFF
AUTO REC: OFF
MONO MIX: OFF
REC LEVEL: 100

APPENDIX 2.3 – CHAPTER 2 WINBUGS CODE

‘All Bats’ model code in WinBUGS:

```
model{
#priors:
beta0~dnorm(0, .01) #intercept
beta1~dnorm(0, .01) #noise trt
beta2~dnorm(0, .01) #elevation
beta3~dnorm(0, .01) #percent shrub cover

alpha0p~dnorm(0, .01) #intercept
alpha1p~dnorm(0, .01) #temp
alpha2p~dnorm(0, .01) #rain

#for sites/ecological model
for(i in 1:M){
N[i]~dpois(lambda[i])
log(lambda[i])<-beta0+beta1*B1[i]+beta2*B2[i]+beta3*B3[i]

#for survey nights/observation model
for (j in 1:J){
y[i,j]~dbin(p[i,j],N[i])

p[i,j]<-exp(lp[i,j])/(1+exp(lp[i,j]))
lp[i,j] <- alpha0p + alpha1p * A1[i,j] + alpha2p * A2[i,j]
} #J
} #M
} #model
```

‘Multi-Species’ model code in WinBUGS:

```
model {

    # Priors
    for (s in 1:6){
    beta0[s]  ~ dnorm(0, .01)
    beta1[s]  ~ dnorm(0, .01)
    beta2[s] ~ dnorm(0, .01)
    beta3[s] ~ dnorm(0, .01)
    p[s] ~ dbeta(1, 1)
    }

    # Likelihood
    # Ecological model for true abundance
    for (s in 1:6){                  # Loop over all species
    for (i in 1:M){                  # Loop over M sites
    N[i,s] ~ dpois(lambda[i,s])      # Abundance
    log(lambda[i,s])<-
beta0[s]+beta1[s]*B1[i]+beta2[s]*B2[i]+beta3[s]*B3[i]

    # Observation model for replicated counts
    for (j in 1:J){           # Loop over surveys/nights
    y[i,j,s] ~ dbin(p[s], N[i,s])   # Detection, varies by species

    } #j
    } #i
    } #s
    } #model
```

APPENDIX 3.1 – CHAPTER 3 EQUIPMENT SETTINGS

SM2BAT+ settings:
Microphones: SMX-US by Wildlife Acoustic
Sampling rate: 192 kHz (for stereo recorders) or 384 kHz (for single-channel recorders)
Recordings mode: Triggered WAV (no compression)
Trigger level: 18 dB
Digital high pass filter: 16 kHz
Min. recordings window: 2 seconds
Max. recording window: 5 seconds
Analog filter switches—
Gain: +36
Analog high-pass filter: 1kHz
Mic bias: OFF
Nightly recording hours: 20:00:00 – 05:59:59

Zoom H2 settings:
Mic gain: M/MID
Mics: 2-channel
LO CUT: OFF
REC MODE: MP3 64
AGC/COMP: OFF
MONITOR: OFF
PLUG-IN: OFF
PRE REC: OFF
AUTO REC: OFF
MONO MIX: OFF
REC LEVEL: 100

APPENDIX 3.2 – CHAPTER 3 PILOT STUDY

In 2012, I conducted a pilot study on BLM land in Rio Blanco County, Colorado. The goal of the pilot study was to determine which bat species were present in the area, and to test ultrasound recording equipment and analysis software using field recordings. A total of 40 sites (*i*) were surveyed for 3 nights (*j*) each, using SM2BAT+ ultrasound detector-recorders. All site locations were established by randomly generating coordinates in ArcGIS.

I incorporated the encounter data from this pilot season into a simple binomial-Poisson mixture model to estimate bat activity levels at each site, and to obtain species-specific estimates of detection probability. Posterior distributions for detection probability were produced for the four species (*s*) discussed in this chapter (*M. ciliolabrum*, *L. cinareus*, *T. brasiliensis*, and *M. evotis*). Using the package 'fitdistr' in R, I fit the posteriors to Beta distributions and estimated species-specific shape parameters 'a' and 'b'. These shape parameters were then used to define the Beta distributions that were used as prior distributions for detection in this Chapter (Figure 3.9).

Model for 2012 pilot data:
$N_{is} \sim \text{Poisson}(\lambda_{is})$
$y_{ijs} \mid N_{is} \sim \text{Binomial}(N_{is}, p_s)$
$\log(\lambda_{is}) = \beta_{0s}$

Priors:
$\boldsymbol{\beta_{0s}} \sim$ Normal (0,100)
$\boldsymbol{p_s} \sim \text{Beta}(1,1)$

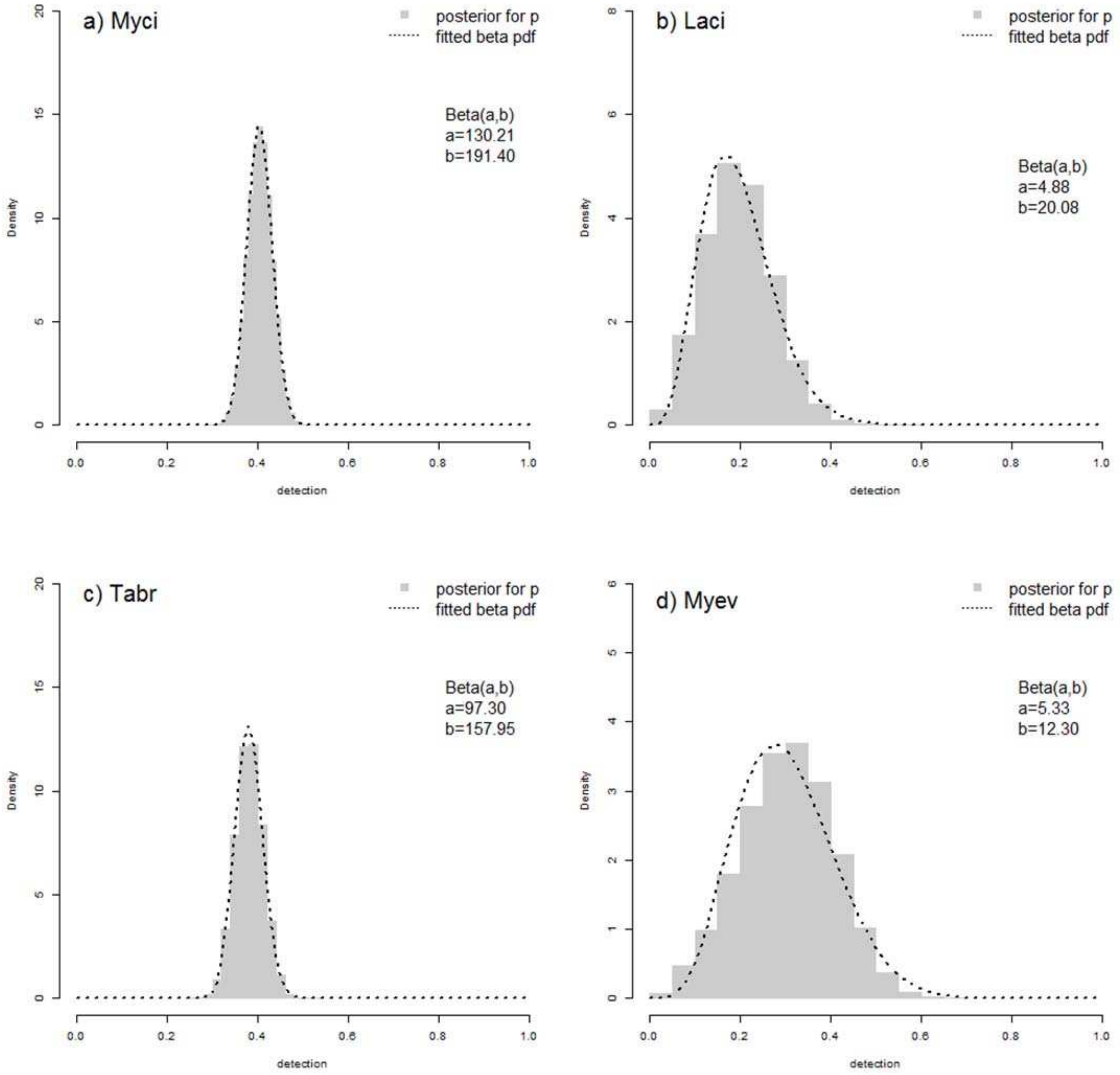

Figure 3.9. Posterior distributions for detection from 2012 pilot season data for (a) *M. ciliolabrum*, (b) *L. cinareus*, (c) *T. brasiliensis*, and (d) *M. evotis*, along with their fitted Beta distributions

APPENDIX 3.3 – CHAPTER 3 WINBUGS CODE

```
model {

    # Priors
    beta0 ~ dnorm(0, 0.01) #intercept
    beta1 ~ dnorm(0, 0.01) #overall time trend
    beta2 ~ dnorm(0, 0.01) #noise treatment by night
    beta3 ~ dnorm(0, 0.01) #interaction
    p ~ dbeta(a,b) # a and b - defined from 2012 pilot data

    # Likelihood
     for (i in 1:nsite){
       for (j in 1:nnight){

        N[i,j] ~ dpois(lambda[i,j])
        lambda[i,j] <- exp(log.lambda[i,j])
       log.lambda[i,j] <-
            beta0+beta1*night[j]+beta2*noise[j]+beta3*night[j]*noise[j]
        y[i,j] ~ dbin(p, N[i,j])
          }  #j night
        }  #i site

    }
```