PRESENCE, ABSENCE, AND ROOSTING ECOLOGY OF THE SOUTHEASTERN MYOTIS AND RAFINESQUE'S BIG-EARED BAT IN THE CACHE RIVER NATIONAL WILDLIFE

REFUGE

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ABSTRACT

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Four bat species, 2 of special concern, the southeastern myotis (*Myotis austroriparius*), and Rafinesque's big-eared bat (*Corynorhinus rafinesquii*) and 2 threatened and endangered species may occur in one of Arkansas' largest bottomland hardwood forests, the Cache River National Wildlife Refuge. However, inventory of bat species throughout the refuge is lacking and management plans may not be adequate in promoting the conservation of bats. My goal was to inventory the bats of the Cache River National Wildlife Refuge and determine roosting habits of the southeastern myotis and Rafinesque's big-eared bat. During summers 2014 and 2015, surveys were conducted throughout the refuge. Six species, Rafinesque's big-eared bat (35%), eastern red bat (*Lasiurus borealis*, 24%), southeastern myotis (18%), evening bat (*Nycticeius humeralis*, 12%), tri-colored bat (*Perimyotis subflavus*, 11%), and big-brown bat (*Eptesicus fuscus*, < 1%), were documented through mist-netting. Occupancy analysis of acoustic data suggests a different pattern of species presence, with tri-colored bats being the most dominant. Occupancy of *Myotis* bats was highest at cypress-tupelo tracts, whereas it was higher for big-brown bats in managed forests. Using radiotelemetry, 19 roost trees were found for the

southeastern myotis and 20 for Rafinesque's big-eared bat. Southeastern myotis did not seem to have a strong preference for any of the measured roost tree characteristics, whereas Rafinesque's big-eared bats selected for trees with higher diameter at breast height.

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CHAPTER I General Introduction

Bats (Order Chiroptera), represent 21.8% of all species of the class Mammalia and are the 2nd most specious group of mammals next to rodents (Order: Rodentia). Bats can be separated into 2-suborder groups -- the megachiropteran, primarily fruit-eating bats, and microchiropteran, whose diets are variable (Merritt, 2010). Suborder megachiroptera has approximately 175 species, whereas microchiropterans are composed of over 900 species (Neuweiler, 2000a).

Bats do not follow the trend of other small mammals in their evolutionary strategy of being r-selected, but instead are typically K-selected (Fleming, 1988), i.e., long life spans with generally low reproductive outputs. This life history is exemplified by the Brandt's myotis (*Myotis brandtii*) which has been documented to have lived over 40 years (Seim, 2013). Such long life spans and low reproductive output make bats vulnerable to environmental changes.

Bats are experiencing declines in population due to anthropogenic threats such as habitat loss or degradation (O'Shea *et al.*, 2016), invasive diseases (U.S. Fish and Wildlife, 2016), and renewable energy sources. Wind turbines, are estimated to be responsible for more than 800,000 bat mortalities per year (Smallwood, 2013). These fatalities occur from single-unit to large-scale wind farms (Jordan, 2014; Smallwood, 2013). Furthermore, *Pseudogymnoascus destructans*, the invasive fungus that causes

White-Nose Syndrome (WNS) causes mortality up to 100% in WNS-positive caves (U.S. Fish and Wildlife, 2016). WNS has been responsible for more than 5.7 million bat mortalities since its introduction in 2006 and has since been officially reported in Arkansas (U.S. Fish and Wildlife, 2016). Loss of bat populations can have a detrimental impact on agricultural practices. For example, bats lost from WNS are estimated to cost from \$3.7 to \$53 billion a year in pest control for farmers across the continental United States (Boyles *et al.*, 2011) and disease control through consumption of vectors of diseases such as mosquitoes (Gonsalves *et al.*, 2013). In areas where WNS is present, it has been estimated that 660 to 1320 metric tons of insects are not being consumed each year alone (Boyles *et al.*, 2011).

Bats of North America and Arkansas

There are 45 species of bats in North America. Arkansas alone has 16 species of bats, 3 of which are federally endangered (*Myotis sodalis, M. grisescens, Corynorhinus townsendii ingens*), and 1 threatened species (*M. septentrionalis*). Some of these species, such as *M. gricescens* are cave-obligate (i.e., hibernate and roost in caves year round). Other species such as Indiana bats (*M. sodalis*), and northern long-eared bat (*Myotis septentrionalis*) roost in caves in the winters and forests in the summer (USFWS, 2007; Sealander and Heidt, 1990; Timpon *et al.*, 2006). Overall, 55% of all bats in North America use forests as a place to roost at some point in their lives (Hayes and Loeb, 2007). Forests provide shelter and habitat for food sources such as insects and moths, cover from predators, and trees in which bats can roost (Hayes and Loeb, 2007; Kunz, 1982).

Southeastern myotis (Myotis austroriparius)

The southeastern myotis (Fig. 1.1; Order: Chiroptera, Family: Vespertillionidae) is a small *Myotis* bat that occurs in the southeastern United States, along the east coast into Virginia, and north into Illinois and Indiana (Fig. 1.2; Arroyo-Cabrales and Álvarez-Castañeda, 2008a). Although it is listed as a species of special concern in Arkansas (AGFC 2013), it is not federally listed as either threatened or endangered. Southeastern myotis weigh 5–12 g, and have a forearm length of 33–42 mm. It occurs with several colors molts such as gray, orange or a combination of gray and orange (Sealander and Heidt, 1990). Females store and dehydrate sperm after mating in a process called delayed fertilization. Sperm becomes rehydrated during the spring for ovulation to occur (Neuweiler, 2000a; Oxberry, 1979). Females typically give birth to twins with a 1:1 sex ratio. Young become volant 5–6 weeks after birth (Jones and Manning, 1989; Schmidly, 1997).

The species roosts in trees inside cavities (Rice, 2009; Stuemke *et al.*, 2014), manmade structures such as houses, bridge joints (Sherman, 2004), in caves in the southeastern part of their range (Rice, 1957), and mines (Harvey *et. al.*, 2011). Cavernicolous southeastern myotis will form maternity colonies reaching up to 250,000 individuals (Texas Parks and Wildlife, 2013) and over 300,000 in Florida (Lacki and Bayless, 2013). Southeastern myotis are often associated with areas that have permanent water sources such as streams and bayous (Jones and Manning, 1989) with tree stands in the bottomlands being water tupelo (*Nyssa aquatica*), bald cypress (*Taxodium distichum*), black gum (*Liquidambar styraciflua*), water oak (*Quercus nigra*), willow oak (*Q. phellos*), and swamp chestnut oak (*Q. michauxii*; Horner and Mirowski, 1996). A water

tupelo tree in Louisiana housed ca. 5,000 individuals (Lacki and Bayless, 2013). Southeastern myotis have been recorded roosting with other species such as Brazilian free-tailed bat (*Tadarida brasiliensis*), evening bats (*Nycticeius humeralis*), tri-colored bats (*Perimyotis subflavus*), gray bats, and Rafinesque's big-eared bat (Loeb *et al.*, 2011; Sasse *et al.*, 2011).

Additionally, echolocation calls of the southeastern myotis range around 50-60 kHz or around 100 kHz depending on what type of call (i.e., communication, feeding buzz). Southeastern myotis calls are similar to those of other bats within the genus *Myotis*, especially northern long-eared bats, and little brown bats (*M. lucifugus*).

Although there is documentation on roosting ecology and habitat preference for the southeastern myotis, knowledge gaps still exist for the species such as home range and foraging ecology. Further species-specific research needs can be found in Lacki and Bayless (2013).

Rafinesque's big-eared bat (Corynorhinus rafinesquii)

The Rafinesque's big-eared bat (Order: Chiroptera, Family: Vespertillionidae) has a range similar to but larger than the southeastern myotis (Arroyo-Cabrales and Álvarez-Castañeda, 2008a, 2008b; Fig. 1.3). Originally, the Rafinesque's big-eared bat was placed in the genus *Plecotus*. However, further examination prompted to change the North American big-eared bats from *Plecotus* to *Corynorhinus* (NatureServer, 2015). Rafinesque's big-eared bats are also listed as a species of special concern in Arkansas (AGFC, 2004). However, it is not federally listed as either threatened or endangered.

This bat is identifiable by its large ears and large pinnae, its toe hairs that extend

beyond the toes, and has wooly gray fur (Fig. 1.4). The Rafinesque's big-eared bat weigh 7–13g and has a forearm length of 39–44 mm (Sealander and Heidt, 1990). Like the southeastern myotis, female Rafinesque's big-eared bats go through the process of delayed fertilization (Neuweiler, 2000b; Oxberry, 1979). . Females give birth to a single pup from May to June depending on their location and young are volant 5–6 weeks after birth (Harvey *et al.*, 1999; Schmidly, 1991)

The type of summer day roosts of Rafinesque's big-eared bats are range dependent, and are associated with bottomland hardwood forests in the Mississippi Alluvial Plain, roosting in hollows of water tupelos, bald cypress, or gum trees (Carver and Ashley, 2007; Johnson and Lacki, 2013a, 2013b; Rice 2009; Stuemke, 2014), or in caves where caves are available, in colonies of ca. 900 individuals (Bayless *et al.*, 2011). Kentucky, North Carolina, and Tennessee currently hold some of the largest congregations of this bat, and all are in caves (Bayless *et al.*, 2011). In addition to using natural roosts, Rafinesque's big-eared bats utilize artificial structures such as bridges, and houses as roosting sites as well as cisterns, and mines (Harvey *et al.*, 2011; Martin *et al.*, 2013). The use of artificial structures may be related to a lack of suitable habitat due to habitat loss or destruction (Bennett *et al.*, 2008).

The echolocation of Rafinesque's big-eared bats is lower than others with a frequency of under 40 kHz. Echolocation of Rafinesque's big-eared bats is generally more difficult to detect on bioacoustics devices as compared to other species (Lacki and Bayless, 2013).

Bottomland hardwood forests and the Cache River National Wildlife Refuge

Both the Rafinesque's big-eared bat, southeastern myotis, Indiana bats (*Myotis sodalis*), and northern long-eared bats (*M. septentrionalis*) have ties to bottomland hardwood forests (Carter and Feldhamer, 2005; Fokidis *et al.*, 2005; USFWS, 2007; Rice, 2009; Stuemke, 2015). America's bottomlands have been greatly reduced and converted for agricultural use. Upon colonization by Europeans, the United States had over 158 Mha of wetland habitat, with 89 Mha present in the lower 48 states. Over a 200-year time span (1780-1980; Dahl, 1990). The state of Arkansas has lost over 70% of their original wetland habitat to agriculture (Hank and Gosselink, 1990). The Mississippi Alluvial Plain, has consisted of 8,498,398–10,117,141 ha of habitat prior to the colonization of European settlers; less than 10% remains today (Stanturf, 2005).

In addition, Arkansas has 2 large bottomland habitats, the White River National Wildlife Refuge and the Cache River National Wildlife Refuge (CRNWR). The CRNWR is comprised of palustrine wetland and bottomland, mature hardwood forest surrounded by agricultural fields and has primarily been used for waterfowl conservation since it was founded in 1986. The 28,000-ha refuge is located within Jackson, Woodruff, Monroe, and Prairie counties, Arkansas (Fig. 1.5). The CRNWR is composed of 17,951 ha of bottomland hardwood forests, 6,000 ha of reforested land, 1,124 ha palustrine and riverine habitats, and 455 ha of cropland and moist-soil units. Agricultural land has been reforested since 1999 to link together portions of the fragmented bottomlands of the CRNWR (U.S. Fish and Wildlife, 2013). The CRNWR shares the border with several state wildlife management areas (WMA) such as Dagmar WMA, Black Swamp North WMA and Black Swamp South WMA as well as land owned by Arkansas Natural

Heritage Commission. The CRNWR is listed on The Ramsar Convention of Wetlands (RCW) as one of the Wetlands of Importance for the United States. The RCW notes that there are a total of 510 species of birds, mammals, reptiles and amphibians, and mussels and 120 species of trees and shrubs within the refuge (The Annotated Ramsar List: United States of America, 2013).

Cave dwelling is more common in the winter among communities of *C*. *Rafinesquii* and *M. Austroriparius* in the northern and southeastern (Florida) part of their range (Jones and Manning, 1989; Rice, 1957; Sealander and Heidt, 1990). However, during the summer, both the *C. rafinesquii*, and *M. austroriparius* use bottomland hardwood forests in Arkansas (Fokidis *et al.*, 2005) and thus the CRNWR may be important habitat for the southeastern myotis and Rafinesque's big-eared bat as one of the largest continuous tracts of wetlands in the Mississippi Alluvial Plain (Wetlands of International Importance, 2014).

Problem statement and Objectives

Several species of special concern and federally threatened and endangered bats use bottomland hardwood forests in the summer for roosting. The CRNWR is the 2^{nd} largest contiguous tracts of wetlands in Arkansas, and among one of the largest in the United States. Although there are several smaller studies (Fokidis *et al.* 2005; Medlin, 2006; Medlin *et al.* 2006, Medlin and Risch, 2008) that broadly surveyed the bottomlands of Arkansas. In addition, several small surveys conducted by U.S. Fish and Wildlife officials on the refuge resulted in several calls identified as Indiana bat in Jackson County. However, no large-scale study has focused on the CRNWR or characterized

roost trees selected by southeastern myotis and Rafinesque's big-eared bat. These data will help to provide a baseline for future monitoring, and help to guide decisions made by land managers. My objectives were to (1) inventory the bat species of the CRNWR, (2) estimate species-specific occupancy in different habitat types on the refuge, and (3) characterize roost trees used as day roosts by the southeastern myotis and Rafinesque's big-eared bat.

Hypothesis and predictions

Objective 1: Inventory bat species of the CRNWR.

Hypothesis 1.1: The range of non-cave-obligate threatened and endangered (T&E) bats does not extend into Jackson, Woodruff, Prairie, and Monroe counties (Sealander and Heidt, 1990). I thus predicted no T&E species would be captured on the refuge.

Hypothesis 1.2: Alternatively, the Indiana bat captured acoustically within the CRNWR prior to this study was a vagrant individual. If so, Indiana bats could be captured on the refuge, but in lower numbers as compared to non-T&E species (Sealander and Heidt, 1990).

Objective 2: Estimate species-specific occupancy in different habitat types on the refuge.

Hypothesis 2.1: Habitat usage of the *Myotis* and Rafinesque's big-eared bat is similar, both in cypress-tupelo habitat, as found in previous studies (Jones and Manning, 1989; Rice, 1957; Rice, 2009; Stuemke, 2014). Thus, I predicted that occupancy of *Myotis* bats (likely dominated by southeastern myotis), and

Rafinesque's big-eared bat would be highest in cypress-tupelo habitat compared to other habitats

Hypothesis 2.2: Habitat usage for other bat species is more flexible because of their wider distribution. Therefore, occupancy should be similar and reflect availability of other habitat types (Sealander and Heidt, 1990).

Objective 3: Characterize roost trees used as day roosts by the southeastern myotis and Rafinesque's big-eared bat.

Hypothesis 3: Habitat associations and roost trees between the southeastern myotis and Rafinesque's big-eared bat are similar and dominated by tupelo and cypress cavities (Barclay and Kurta, 2007; Hein *et al.*, 2008; Jones and Manning, 1989; Rice, 2009; Stuemke *et al.*, 2014). If true, roost trees will be dominated by water tupelo and bald cypress trees with basal openings.

This thesis contains 4 chapters. In chapter 2, I investigate the distribution of bat species and occupancy among habitats within the refuge. In chapter 3, I investigate the day-roosts of the Rafinesque's big-eared bat and southeastern myotis within the refuge and identify patterns of selection for both species. Finally, in chapter 4, I synthesize my findings and discuss limitations and perspectives for future research.

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Figure 1.1 – Southeastern myotis (*Myotis austroriparius*), captured in the Cache River National Wildlife Refuge in 2014.

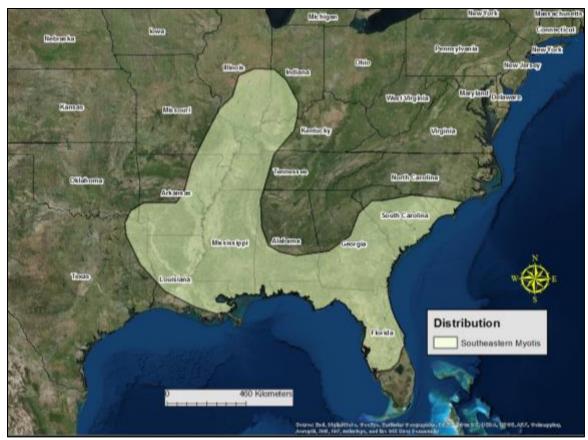


Figure 1.2 – Distribution of the southeastern myotis (*Myotis austroriparius*) from IUCN Red List (2016).

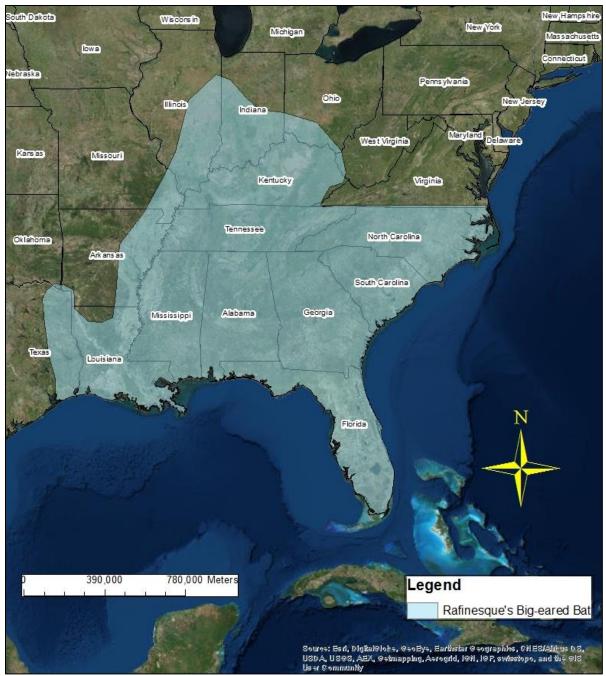


Figure 1.3 - Distribution of the Rafinesque's big-eared bat (*Corynorhinus rafinesquii*) from IUCN Red List (2016).



Figure 1.4 – Rafinesque's big-eared bat (*Corynorhinus rafinesquii*) with ears erect taken in 2015 at Cache River National Wildlife Refuge.

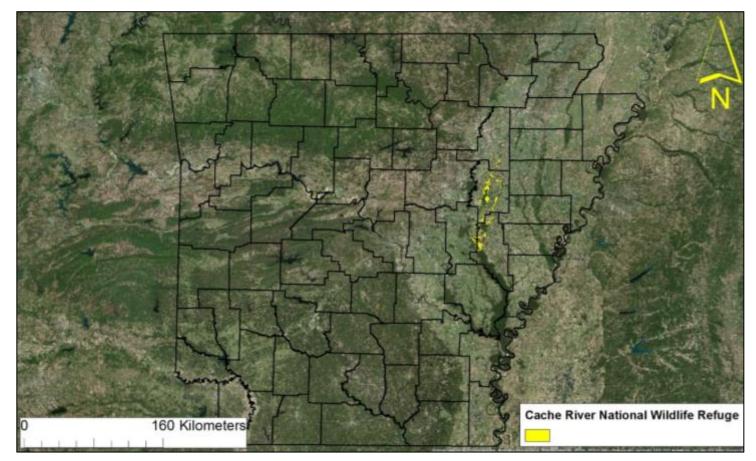


Figure 1.5 – Location of the Cache River National Wildlife Refuge, Arkansas.

CHAPTER II

PRESENCE, ABSENCE, AND OCCUPANCY OF BAT COMMUNITIES OF THE CACHE RIVER NATIONAL WILDLIFE REFUGE

ABSTRACT – Several studies have been conducted on the bats of Arkansas' bottomlands. However, none have focused exclusively on one of Arkansas' largest bottomlands habitats, the Cache River National Wildlife Refuge. The objectives of this study were to 1) inventory bat species present throughout the Cache River National Wildlife Refuge via mist-netting and bioacoustics, and 2) estimate the occupancy of bats among five habitat types (cypress-tupelo, emergent wetland, mature forest, reforestation, and managed hardwood). I mist-netted throughout the boundaries of the refuge from sunset for 5 hours and deployed bioacoustic devices from May-August 2014 and 2015. In addition, separate bioacoustics devices were deployed throughout the aforementioned habitat types from May through August, 2015. Four hundred and sixty bats were capture via mist-netting with Rafinesque's big-eared bats (Corynorhinus rafinesquii; n = 156) being the most common capture, followed by eastern red bats (*Lasiurus borealis*; n = 104), southeastern myotis (*Myotis austroriparius*; n = 91), evening bats (Nycticeius humeralis; n = 58), tri-colored bats (Perimyotis subflavus; n = 54), and a bigbrown bat (*Eptesicus*, fuscus; n = 1). Evening bats and big-brown bats tended to occupy managed hardwood forests more than any other habitat with the occupancy probability $\Psi = 0.81$ and 0.58, respectively, tri-colored bats tended to be more present in mature forest habitats ($\Psi = 0.89$), and Myotis species tended to have highest occupancy rates at cypress-tupelo stands ($\Psi = 0.59$).

INTRODUCTION

Arkansas is home to 16 species of bats, 10 of which occur in the same counties as the Cache River National Wildlife Refuge (CRNWR; Sealander and Heidt, 1990). Additionally, 4 of the 16 species have some level of federal protection: the Indiana bat (*Myotis sodalis*), gray bat (*Myotis grisescens*), Ozark big-eared bat (*Corynorhinus townsendii ingens*) are all listed as federally endangered and northern long-eared bat (*Myotis septentrionalis*) is listed as federally threatened. Several studies have focused on the distribution of bats throughout Arkansas and specifically the bottomlands of Arkansas (Fokidis *et al.*, 2005; Medlin, 2006; Medlin, *et al.* 2006). However, comprehensive presence/absence data of the CRNWR are severely lacking. Prior to this 2015, no study had documented occupancy in the refuge. Filling in knowledge gaps will give land managers at the CRNWR insight on how to manage their land to protect bat populations.

There are several methods to conducting population studies of bats such as point-count surveys in caves, mist-netting, harp-trapping, banding and emergence counts from roosts (O'Shea and Bogan, 2003). These methods can be costly in both time and money. Monitoring bats passively with bioacoustics gives insight to presence/absence of species in an area and temporal activity levels and occupancy but cannot provide population estimates (Byrnes, 2013; Hayes *et al.*, 2009; Weller, 2008). Passive presence/absence methods have the benefit, however, of being more cost effective compared to traditional methods (Royle and Nichols, 2003). Other surveying methods such as mark-recapture can be conducted as well to determine population size and distribution. However, bat recapture rates are generally low, likely because bats avoid areas where they were previously captured (Kunz and Brock, 1975: Perry, 2011). Therefore, the most informative method to assess bat populations is via presence/absence data.

My 1st objective was to inventory bat species of the Cache River National Wildlife Refuge using mist nets and bioacoustic devices. My 1st hypothesis focused on the distribution of non-cave-obligate species within the refuge. I predicted no threatened and endangered species (T&E) would be captured on the refuge (Sealander and Heidt, 1990). My 2nd prediction was that if the Indiana bat detected from previous acoustic surveys was not a vagrant individual, Indiana bat captures would be lower than for non-T&E species. My 2nd objective was to estimate occupancy of bats (via bioacoustics) in different habitats within the refuge. My 1st hypothesis was that habitat usage between the southeastern myotis and Rafinesque's big-eared bat are similar, as found in other studies (Jones and Manning, 1989; Rice, 1957; Rice, 2009; Stuemke, 2014), so occupancy should be highest in cypress tupelo habitat. My 2nd hypothesis was that habitat usage among other bat species is more flexible because of their wider distribution (Sealander and Heidt, 1999). I thus predicted occupancy would reflect availability of available habitat.

METHODS

Study site

The CRNWR is located within the Mississippi Alluvial Plain, surrounded by agricultural fields, and has primarily been used for waterfowl conservation since it opened in 1986. The 28,000-ha refuge is located within Jackson, Woodruff, Monroe, and Prairie counties, Arkansas. The CRNWR is composed of 17,951 ha of bottomland hardwood forests, 6,000 ha of reforested land, and 1,124 ha of marshes, oxbow lakes, bayous, and rivers with 455 ha of cropland and moist-soil units. See Chapter 1 for further details.

Mist-netting and Species Diversity Index

I used 1 of 4 sizes (4–12 m high) of 38-mm meshed mist-nets (AviNet Incorporated, New York, USA). Net sizes were chosen on a site-by-site basis. Netting locations were chosen based on canopy closure to provide suitable corridors (roadways, streams) to funnel bats into nets. Three to 4 mist-net set-ups were erected at each site. Nets were opened at sunset and were checked for bats every 10 min for a total of 5h as per U.S. Fish and Wildlife Indiana bat (*Myotis sodalis*) protocol.

From mist-netting data, I calculated the overall and year-specific species diversity for the CRNWR. I measured species diversity of the refuge using Simpson's Index (SI):

$$D = \frac{\sum n(n-1)}{N(N-1)}$$
(Krebs, 1999)

where *n* is the total number of organisms for each species and *N* is the total number of organisms. The value of *D* is subtracted from 1, which gives us Simpson's Diversity Index (SDI), always between 0 and 1. The higher the SDI value, the more diverse the population is. For example, if SDI = 0.90, there is a 90% chance that 2 bats taken from the population will be of a different species, or a 10% chance that they will be of the same species.

Acoustic surveys and analysis

Acoustic surveys I conducted using AnaBat SD2 units (Titley Electronics, Ballina, Australia) complemented netting efforts. Units were placed in a modified ammo box with a PVC fitting attached to direct echolocations to the microphone of the SD2 unit. Ammunition boxes were placed on a 1-m tall PVC pipe anchored to the ground within 75-m of net-sites in fields, corridors or the interior of the forest. Additionally, in 2015 only, the U.S Fish and Wildlife Service surveyed each of 5 habitats of the CRNWR i.e., emergent wetland covering 3% of the

refuge, managed hardwood (4%), cypress-tupelo (7%), reforested (21%), and mature forest (65%) using 3 SD2 units for 2–5 nights.

I analyzed call data using Bat Call Identification version 2.7c (BCID 2015, Ryan Allen, Kansas City, Missouri). I set the parameters of the software to narrow down the list of bat species to bat species present in the state of Arkansas and to exclude the Ozark big-eared bat, the eastern small-footed bat (*Myotis leibii*), and the gray bat whose distribution does not overlap the study site. Seminole bat (*Lasiurus seminolus*) and Brazilian free-tailed bat (*Tadarida brasiliensis*) echolocation calls are not available in the BCID library. BCID was set to only retain calls with an 85% probability of accuracy or higher.

BCID produces an Excel worksheet and gives calls a unique I.D in the mddttt format. For example, if it logs a call on August 1st at 0314, it is labeled as 8010314. The data sheets also identify possible species, with a probability that the call is of a certain species, and provides number of pulses, the probability that all the pulses are of the identified species, and the discrete probability that a specific call is of the identified species. I used Analook version 4.1 (Titley Electronics, Columbia, Missouri) to visually vet pulses based on the minimum and maximum frequency, length of the call, slope and slope change, and overall shape of the calls. For example, Indiana bat calls start approximately at 90 kHz and have a steep drop to 50 kHz from 1 to 2 ms before a second steep drop slightly until 6 ms before a second steep drop from 40 to 30 kHz. If the software identified a pulse with completely different characteristics than those of Indiana bats, I reassigned it to a more appropriate species if applicable or called it unidentifiable; otherwise, it was not reassigned.

In addition to analyzing calls in BCID, calls were analyzed in EchoClass version 3.1 (Britzke, Vicksburg, Mississippi) using Species Set 1 which includes big-brown bats, eastern

small-footed bats (*Myotis leibii*), little brown bats (*M. lucifugus*), silver-haired bats (*Lasionycteris noctivagans*), hoary bats (*L. cinereus*), eastern red bats (*L. borealis*), Seminole bats, Indiana bats, southeastern myotis (*M. austroriparius*), evening bats (*Nycticeius humeralis*), gray bats, northern-long eared bats (*M. septentrionalis*) and tri-colored bats (*Perimyotis subflavus*).

Occupancy analysis

Data from mist-netting and bioacoustics allow for the construction of a matrix of 0s and 1s to create occupancy detection histories, the basis for occupancy models. For example, a bigbrown bat with a detection history of "01010" indicates that species was detected on the 2nd, and 4th night of a sampling effort period. This pattern means that the species is present but not always detected during a sampling effort period. These matrices can be used with analytical programs to assess occupancy across a sampling area. Occupancy, denoted by psi (ψ), is the probability that a site selected at random or sampling unit in a single area is occupied by a species (MacKenzie *et al.*, 2006). Occupancy models provide naïve ψ , which is the ratio calculated using the following equation (Lancia *et al* 1996; MacKenzie *et al.*, 2006):

$$\hat{\psi} = \frac{x}{S}$$
,

where x is the number of occupied sites and s is the total number of surveyed sites. x is a conservative number since not capturing or detecting an animal at a site does not mean the site is not occupied (henceforth known as false absence). The probability of detection (p) is the expected proportion of animals present that is actually detected. True ψ is estimated by correcting naïve ψ with the probability of detection. A ψ of 0.15 indicates a 15% probability that a species is present at a site and 85% probability the site is not occupied by said species.

Management practices can be better informed if occupancy is reported along with detectability. Estimates of naïve ψ , ψ , and the probability of detection are reported with their standard errors.

Single-season occupancy models have 3 assumptions to consider before analysis (MacKenzie *et al.*, 2002): 1) sites are closed to changes in occupancy, 2) species are never falsely detected when absent, and 3) detection of a species at a site is assumed to be independent of detecting the species at all the other sites. Assumption 1 was met by short sampling periods in which changes in occupancy are least likely to occur, i.e., volancy, death, and recruitment. I ensured assumption 2 by visually vetting calls and assumption 3 by considering life histories and ecologies of the bat species that may occur within the CRNWR (Sealander and Heidt, 1990).

The occupancy analysis focuses on 8 selected species: the southeastern myotis, Indiana bat, northern long-eared bat, eastern red bat, evening bat, tri-colored bat, Rafinesque's big-eared bat, and the big brown bat. Occupancy was analyzed in Program PRESENCE v. 10.5 for all bat species. Due to similarities of echolocation structure of four *Myotis* bats whose range falls within the refuge, i.e., the northern long-eared bat, southeastern myotis, Indiana bat, and little brown bat, all *Myotis* bats were placed into 1 *Myotis* group for analysis. A model was run for each bat species for each of the 5 habitat types.

Next, I ran 2 models in Program PRESENCE, the Constant P model which determines the highest probability of detection and assigns each survey effort the same probability of detection. Survey-specific P models assign probabilities of detection for each night of each survey effort. Models that did not converge or did not have sufficient enough data to run effectively were excluded from further analyses. Program PRESENCE provides the Akaike information criterion (AIC) for both constant P and survey-specific P. This allowed me to determine which model was the best model. Finally, I compared occupancy estimates among

habitats, using Program CONTRAST version 2. (Hines and Sauer 1989) to determine which habitat was associated with the highest ψ for each species.

RESULTS

Mist-netting and diversity

Physical presence/absences were recorded via mist-netting from 23 May 2014 through 4 August 2014 and from 6 June 2015 through 1 August 2015 for a total of 45 nights and 21 sites. Six species were caught over 2 years: Rafinesque's big-eared bat, eastern red bat, southeastern myotis, tri-colored bat, evening bat, and big-brown bat (*Eptesicus fuscus*). Rafinesque's bigeared bat was the most dominant species captured via mist-netting (n = 156; Fig 2.1), followed by eastern red bat (n = 104), southeastern myotis (n = 91), evening bat (n = 58), tri-colored bat (n = 54), and big brown bat (n = 1, < 1%) for a total of 464 individuals. Species diversity was similar in 2014 and 2015 at 0.76 and 0.79, respectively. The SDI of the CRNWR was 0.77, suggesting a diverse bat community in the CRNWR (Table 2.1; Fig. 2.1).

Acoustics and occupancy

Out of 887,853 call files collected by U.S. Fish and Wildlife Service, BCID identified 3,896 calls with an 85% probability to the species level (Fig. 2.1): by decreasing order 3,470 files (89%) were identified as tri-colored bat, 277 (7%; Fig 2.1) as *Myotis* species, 166 (4.7%) as evening bats, 36 (<1%) as silver-haired bats, 34 (<1%) as big-brown bats, 9 (<1%) as hoary bats, and 3 (<1%) as eastern red bats. Similarly, out of 64,681 call files collected by Arkansas State University (Fig. 2.1), BCID identified 744 calls with an 85% probability to the species level. In decreasing order, 77% (n = 576) as tri-colored bats, 20% (n = 149) as *Myotis* species, 2% (n = 14) as evening bats, <1% (n = 5) as big-brown bats, <1% (n = 2) as Rafinesque's big-eared bats,

and <1% (n = 1) as eastern red bats. Both tri-colored bats and *Myotis* bats had the highest number of identified calls among both collections.

Constant probability of detection was the best model for most bats in most habitats. Occupancy for *Myotis* bats tended to be higher at cypress-tupelo stands ($\Psi = 0.59 \pm 0.15$; $P = 0.42 \pm 0.10$) and lowest at emergent wetland habitats ($\Psi = 0.17 \pm 0.09$; $P = 0.0.49 \pm 0.20$; Table 2.2). Occupancy of tri-colored bats tended to be high in all sites ranging from cypress-tupelo ($\Psi = 0.75 \pm 0.10$; $P = 0.84 \pm 0.05$) to mature forest ($\Psi = 0.90 \pm 0.09$; $P = 0.69 \pm 0.07$; Table 2.2). Occupancy of evening bats was lowest at mature forest ($\Psi = 0.44 \pm 0.15$, $P = 0.46 \pm 0.11$) and tended to be highest at managed hardwood stands ($\Psi = 0.81 \pm 0.17$; $P = 0.37 \pm 0.09$; Table 2.2). Occupancy was lowest for big-brown bats at cypress-tupelo stands ($\Psi = 0.25 \pm 0.18$; $P = 0.26 \pm 0.19$) and tended to be highest at managed hardwood ($\Psi = 0.58 \pm 0.50$; $P = 0.13 \pm 0.12$ Table 2.2). Too few data were available to estimate occupancy of eastern red bat. No one habitat type was significantly higher than the refuge as a whole (Table 2.3).

Results from EchoClass's output of *Myotis* bats in cypress-tupelo habitat initially showed 5 out of 18 sites occupied. After visual vetting of these calls, I removed 3 calls which were falsely identified as *Myotis* bats. These 3 calls were in fact background noise and had no characteristics that could justify them as even being bat calls in the first place. Two of 18 sites were occupied by *Myotis* bats.

DISCUSSION

The CRNWR has a diverse bat community with 6 physically confirmed species throughout the refuge. None of which are listed as federally threatened or endangered. Even though acoustic detected all 6 confirmed species, the most common capture, the Rafinesque's big-eared bat, had only two calls make it past the 85% minimum probability and dissimilarly the most uncommon capture (big brown bat) had twenty confirmed calls which shows disproportion between physical and acoustic monitoring. These data support my first prediction on that status of T&E species on the CRNWR.

Furthermore, occupancy results show that *Myotis* bats tended to have higher occupancy at cypress-tupelo stands more than any other habitat type even though cypress-tupelo stands comprise 6.6% of the refuge. The high occupancy of *Myotis* bats in cypress-tupelo habitat could reflect the high numbers of southeastern myotis captured via mist-netting throughout the CRNWR and strong associations with bottomland habitats where caves are absent (Gooding and Landford, 2004; Jones and Manning, 1989; Rice, 1957; Stuemke *et al.* 2014). Other *Myotis* bats such as the Indiana bat, small-footed bat, and gray bat are more common in the highland areas of the state (Sealander and Heidt, 1990) and will unlikely be found in CRNWR. Little brown bats have a wider distribution than southeastern myotis (Sealander and Heidt, 1990) and are not restricted to the bottomlands of the state.

Additionally, the tri-colored bat was the most common identified call with over 4,000 calls identified with an 85% minimum probability despite being the second least common capture via mist-netting. This is similar to the findings of Jordan (2014) who had low numbers of tri-colored bat captures but high numbers of confirmed calls. The probability of detection for tri-colored bats has a small range (0.69–0.91), but probability of detection was higher for tri-colored bats than all other species. The calls of tri-colored bats are of a higher amplitude as compared to other bat species that were identified on the refuge and may be easily detectable by bioacoustic devices (Ryan Allen, [BCID] personal communication, [May, 2016]; MacDonald *et al.*, 1994) and could have biased my results.

Conversely, Rafinesque big-eared bats and other bats within the genus *Corynorhinus* echolocate on low amplitudes causing them to be undetectable, hence their nickname of "whispering bats" (Lacki and Bayless, 2013; Loeb *et al.*, 2015; Stihler, 2011). Although Rafinesque's big-eared occupancy could not be determined, data supported the first prediction for the 2nd objective: *Myotis* bats tended to be more present in cypress-tupelo habitats.

The loudness of tri-colored bats and quietness of Rafinesque's big-eared bat may lead to overestimated and underestimated occupancy estimates, respectively. Similarly, the eastern red bat had the second highest physical capture rate among our six species, but it was also among the least common identified bat calls, despite higher frequencies than Rafinesque's big-eared bat. Eastern red bats are sometimes confused with tri-colored bats using automatic identification software and possibly during the vetting process too.

Additionally, the rarest capture, big-brown bats, in 2014 and 2015 also had low naïve and true Ψ , with the exception of the managed hardwood habitat; the probability of detection for this species had a low range, which seems to correspond with the overall rarity of the species on the refuge. This low occurrence was also reflected by mist-netting data. Big-brown bats tended to be most common on managed hardwood which may be related to the relative openness of forest structure. Evening bats tended to occupy managed hardwood forest more than any other habitat. This could be related to evening bats' preference towards habitats that have more opened canopy (Istvanko, 2015). Similarities of habitat usage between big-brown bats and evening bats have been described in studies of both species (Timpone *et al.*, 2006). Results did not support my 2nd hypothesis of availability reflecting available habitat throughout the refuge.

Furthermore, due to similarity of echolocation calls among members of the genus *Myotis*, bats identified to *Myotis* were placed into one group for analyses. While I can confirm high

numbers of southeastern myotis from harp-trapping events and netting efforts, improvements on bioacoustics software, call recognition, and a more robust call library are needed on all *Myotis* bats before acoustic-only surveys can be reliable. Likewise, due to current inability of bioacoustic devices to detect low frequency calls of big-eared bats and inability of software to accurately identify big-eared bats, it is suggested that surveys require mist-netting to confirm presence or absence (Kaiser and O'Keefe, 2015). While reliance on species identification is questionable, acoustic surveys are novel ways to identify periods of bat activity throughout the landscape and deployment of devices is cheaper than the manpower of physical surveys. Similarly, mist-netting for some species may not be very productive because some bats are known to be high-fliers such as hoary bats, which are common amongst wind-turbine fatalities (Johnson *et al.*, 2004). However, mist-netting provides the researcher with a physical bat inhand.

Based on previous literature (Fokidis *et al.*, 2005; Sealander and Heidt, 1990), I expected to catch 9 of Arkansas' 16 species via mist-netting. However, little brown bats, hoary bats, and Brazilian free-tailed bats were not captured throughout the study, despite distributions and capture history within the counties that the CRNWR encompasses. Because an Indiana had previously been detected acoustically in Jackson county, I could not exclude its presence in the Refuge. However, no Indiana bat was caught via mist-netting and the calls were too similar to other *Myotis* to conclude on its presence or absence in the Refuge. I was able to confirm the presence of the 6 other bat species. The presence of 4 of these 6 species (red bats, big brown bats, evening bats, and tri-colored) was not surprising. Indeed, they are fairly common throughout the state (Fokidis *et al.*, 2005; Sealander and Heidt, 1990). Studies in the southeastern portion of the state where bottomland forests are present (Baker and Ward, 1967), and highlands

of the Ouachita mountains (Saugey *et al.*, 1989), showed low capture rates of both Rafinesque's big-eared bats and southeastern myotis, whereas Fokidis *et al.* (2005) and Medlin *et al.* (2006) showed higher captures of both species in the east-central portion of the state. Higher numbers in the CRNWR may relate to overall suitability of the refuge for these two species. The CRNWR is also on the westward edge of their distribution (Arroyo-Cabrales, and Álvarez-Castañeda, 2008a, 2008b) and represent their core population in the state.

Finally, the inability of EchoClass to positively identify calls coupled with the software's' lack of option to select target bat species, makes it inferior to other acoustic analysis programs such as BCID. Analysis of the entire refuge's raw using EchoClass took over 30 days with software failures, whereas BCID handled analysis in less than 1 hour per habitat type. The naïve ψ of cypress-tupelo habitat for BCID for *myotis* species was 0.50. Conversely, EchoClass identified 5 calls on cypress-tupelo habitat as *myotis* species, after visual vetting that was reduced to 2 calls (naïve ψ of 0.1).

This chapter provides data of occupancy and presence/absence of bat species on the refuge and provides land managers information that can be used for the construction of future management regimes. In addition, this information may be able to be used on similar habitats i.e., White River National Wildlife Refuge (WRNWR), which is the largest bottomland habitat in Arkansas. Surveying on the WRNWR may provide comparative data that can help identify characteristics of suitable habitat for these species in both refuges in what might be, their core population in Arkansas.

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Year	CORA	LABO	MYAU	NYHU	PESU	EPFU	Total (N)	SDI
2014	99	67	41	32	39	0	278	0.76
2015	56	37	50	26	15	1	191	0.79
Total (N)	162	111	83	58	54	1	469	0.77

Table 2.1 – Bat Species Simpson's Diversity Index (SDI) for Cache River National Wildlife Refuge. Rafinesque's big-eared bat (CORA), eastern red bat (LABO), southeastern myotis (MYAU), evening bat (NYHU), tri-colored bat (PESU), big-brown bat (EPFU).

Table 2.2 – Occupancy estimates ($\Psi \pm SE$) for myotis species, tri-colored bats, evening bats, and big-brown bats in each pre-defined habitats of the Cache River National Wildlife Refuge for 2015. AIC = Akaike Information Criterion. AICwt = Relative support of the model. Naïve Ψ = Number of sites at which species is present / Total surveyed sites. Ψ = Overall occupancy. *P* = Probability of detection

Habitat	Model	AIC^1	AICwt ³	Naïve Ψ^4	$\Psi^2 \pm S.E^5$	$P^3 \pm S.E^6$
Myotis species						
Cypress Tupelo	Constant P	72.19	0.69	0.50	0.59 ± 0.15	0.42 ± 0.10
Emergent Wetland	Constant P	32.46	0.97	0.16	0.17 ± 0.09	0.49 ± 0.20
Mature Forest			Conver	rgence failed		
Reforestation	Constant P	41.1	0.95	0.2	0.23 ± 0.11	0.47 ± 0.17
Managed Hardwood	Constant P	62.62	0.88	0.37	0.45 ± 0.15	0.39 0.18
Tri-colored bat						
Cypress Tupelo	Constant P	64.83	0.22	0.83	0.83 ± 0.09	0.84 ± 0.05
Emergent Wetland	Constant P	59.96	0.68	0.89	0.89 ± 0.07	0.88 ± 0.04
Mature Forest	Constant P	77.06	0.96	0.87	0.90 ± 0.09	0.69 ± 0.07
Reforestation	Constant P	70.36	0.87	0.75	0.75 ± 0.10	0.83 ± 0.05
Managed Hardwood	Constant P	54.07	0.96	0.84	0.84 ± 0.08	0.91 ± 0.04
Evening bat						
Cypress Tupelo	Constant P	72.88	0.86	0.50	0.55 ± 0.14	0.49 ± 0.10
Emergent Wetland	Constant P	73.59	0.90	0.53	0.72 ± 0.21	0.31 ± 0.10
Mature Forest	Constant P	58.3	0.96	0.36	0.44 ± 0.15	0.46 ± 0.11
Reforestation	Constant P	72.32	0.88	0.45	0.51 ± 0.13	0.49 ± 0.11
Managed Hardwood	Constant P	80.79	0.42	0.63	0.81 ± 0.17	0.37 ± 0.09
Big-brown bat						
Cypress Tupelo	Constant P	32.12	0.49	0.17	0.25 ± 0.18	0.26 ± 0.19
Emergent Wetland	Constant P	40.87	0.97	0.21	0.29 ± 0.16	0.31 ± 0.16
Mature forest						
Reforestation	Constant P	41.71	0.71	gence failed 0.21	0.32 ± 0.19	0.27 ± 0.15
Managed Hardwood	Constant P	38.94	0.96	0.21	0.58 ± 0.50	0.13 ± 0.12

Table 2.3 – Comparison of species- and habitat-specific occupancy ($\Psi \pm SE$) from Table 2.2. Df denotes degrees of freedom. $\alpha = 0.05$. *Myotis* include *M. austroriparius*, *M. sodalis*, *M. septentrionalis*, and *M. lucifugus*; big brown bat (EPFU), tri-colored bat (PESU), evening bat (NYHU).

Bat species	df	р	Chi ²
Myotis	3	0.07	7.2
EPFU	3	0.93	1.7
PESU	4	0.79	3.5
NYHU	4	0.48	0.1

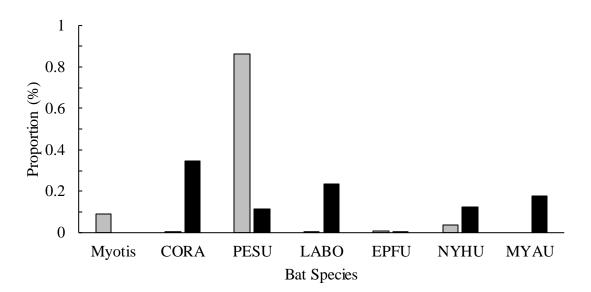


Figure 2.1 – Proportions of counts of individual bats captured through mist-netting (black) and count of call files collected through bioacoustics (gray) for Rafinesque's bigeared bat (CORA), eastern red bat (LABO), southeastern myotis (MYAU), *Myotis* species, evening bats (NYHU), tri-colored bat (PESU), and big-brown bat (EPFU). Note: *Myotis* species were pooled together for the bioacoustics count because these species have similar calls and could not be distinguished with certainty.

CHAPTER III:

ROOST TREE CHARACTERISTICS OF SOUTHEASTERN MYOTIS AND RAFINESQUE'S BIG-EARED BATS IN THE CACHE RIVER NATIONAL WILDLIFE REFUGE

ABSTRACT – Two species of special concern in Arkansas, the southeastern myotis and Rafinesque's big-eared bat, are known inhabitants of bottomland hardwood forests in other regions, where caves are absent. However, their roosting ecology in the bottomlands of Arkansas is unknown. The objective of this study was to characterize roost tree used by these species in the Cache River National Wildlife Refuge so land managers can develop management plans that promote the conservation of both species in the refuge. I affixed 23 transmitters to southeastern myotis in 2014 and 2015 and 9 to Rafinesque's big-eared bat in 2015. Bats were tracked daily to identify roost trees. I measured diameter at breast height, canopy cover, basal area, and tree height of all identified roost trees and a paired random tree. In addition, I measured diameter at breast height and recorded tree species of all trees within a 0.5-m and 11.3-m plot around each roost and random tree. I identified 19 roost trees for the southeastern myotis and 20 for Rafinesque's big-eared bat. Both bat species roosted primarily in water tupelos and bald cypress trees. Roost trees of both the southeastern myotis and Rafinesque's big-eared bat tended to be larger in diameter with higher canopy cover and in thicker stands as compared to random trees.

INTRODUCTION

There are 45 species of bats in North America, 55% of which use forests as a place to roost at some point in their lives. Arkansas has 16 species of bat, of which 8 will use a forest some time during their life, i.e., from hibernation to reproductive season, and 6 that use forests exclusively (Hayes and Loeb, 2007; Lacki *et al.*, 2007; Sealander and Heidt, 1990). Forest-dwelling bats roost in tree cavities, exfoliating bark (Johnson and Lack, 2013), in foliage (Sealander and Heidt, 1990), and underneath leaf litter (Perry and Thill, 2008). Tree roosts can be separated into 2 types: day roosts and night roosts, both of which serve similar functions as a place that provides protection from predators and adverse weather conditions. Night roosts may serve as resting area between feeding bouts, or to provide social interaction (Kunz, 1982). Long periods are spent in day roosts i.e., from sunrise to sunset, and are used as a place to raise young. Additionally, females can form maternity colonies by gathering in numbers, up to several hundred in a single tree, with juveniles possibly benefitting from the microclimate (Kunz, 1982; Kunz and Lumsden, 2003).

Two of Arkansas' species of special concern bats, the southeastern myotis (*Myotis austroriparius*) and Rafinesque's big-eared bat (*Corynorhinus rafinesquii*), are known to roost in bottomland hardwood forests within tree of water tupelos (*Nyssa aquatica*) and bald-cypress (*Taxodium distichum*) cavities (Clark, 1990; Fokidis *et al.*, 2005; Jones and Manning, 1989; Rice, 1957, Rice, 2009; Stuemke *et al.*, 2015). These roost trees are often associated with streams and lakes that provide bats with water, or a flyway in which to reach their foraging area (Hein *et al.*, 2008; Kunz, 1982). However, these roosting preferences of these species have never been confirmed in the Cache River National

Wildlife Refuge (CRNWR). Yet, the refuge represents one of the largest bottomland hardwood forest and could be managed for the conservation of these species.

Several studies have been conducted analyzing roost selection among bat species. Similarities between these studies include qualitative variables such as tree species and life stage (live/snag), and quantitative variables such as diameter at breast height (DBH), canopy cover, tree height, and basal area (Barclay and Kurta, 2007; Clement and Castleberry, 2013; Klotz, 2012). Basal area may indicate tree competition (Contreras *et al.*, 2011; Goelz and Meadows, 1999) which may influence tree diameter and height which in turn, may influence the size of the basal cavity of the tree and provide space for communal roosting and a more stable microclimate (Kunz and Lumsden, 2003). Similarly, canopy cover dictates amount of sunlight reaching under understory of the tree which also may provide stable microclimates for bats (Carver and Ashley, 2008; Jennings *et al.*, 1999). Identifying these variables can help determine how the southeastern myotis and Rafinesque's big-eared bats select their trees during the reproductive period (Jones and Manning, 1989; Harvey *et al.*, 1999; Kunz, 1982).

My objective was to characterize day roost trees of the southeastern myotis and Rafinesque's big-eared bat. My hypothesis focused on habitat associations and roost trees characteristics between the southeastern myotis and Rafinesque's big-eared bats. I predicted that both southeastern myotis and Rafinesque's big-eared bat roost trees would be associated with water systems, and roost trees would be dominated by water tupelos and bald cypress with basal openings, as found in previous studies (Barclay and Kurta, 2007; Hein *et al.*, 2008; Jones and Manning, 1989; Rice, 2009; Stuemke *et al.*, 2014).

METHODS

Study site

Arkansas has two of the largest tracts of bottomland hardwood forest, the White River National Wildlife Refuge and Cache River National Wildlife Refuge (CRNWR). The CRNWR is located within Woodruff, Prairie, Monroe and Jackson Counties. The refuge is dominated by mature hardwood forests, reforested hardwood, emergent wetland, cypress-tupelo, and managed hardwood comprising almost 14% of the refuge. See Chapter 1 for more details on the CRNWR.

Mist-netting, harp-trapping and banding

Aluminum mast poles (The Mast Company, North Carolina) systems were used to hold mist-nets of varying sizes (4–12 m) in place. Nets were placed in a double or triple arrangement. Netting locations were chosen based on canopy closure to provide suitable corridors (roadways, streams) to funnel bats into nets. Nets were opened at sunset and were checked for bats every 10 min for a total of 5h as per U.S. Fish and Wildlife Indiana bat (*Myotis sodalis*) protocol. For each bat, we measured forearm length, mass, and determined species, sex, age, reproductive status

I used a G7 Forest Strainer (Bat Conservation and Management, Carlisle, PA) to harp-trap roost trees in 2014 and 2015. Tarps were secured to the tree and duct-taped around the harp-trap to funnel bats into the trap (Fig. 3.1). I banded all bats caught during harp-trapping events using a 2.4-mm and 2.9-mm metal ring bands provided by Arkansas Game and Fish Commission. Each band has an etched unique I.D. that is specific for the

state of Arkansas. Traps were set up before sunset and closed approximately 2 hours after sunset.

Radiotelemetry

I affixed transmitters to southeastern myotis in 2014 and both southeastern myotis and Rafinesque's big-eared bat in 2015. Bats were considered for affixing transmitters if the radio was $\leq 5\%$ of their body weight following American Society of Mammologists guidelines (Sikes and Gannon, 2011). In 2014, I used a LB-2X (Holohil, Inc., Ontario, Canada) transmitter with a frequency range of 150-150.999 MHz and a nominal life of 21 days and a mass of 0.32g. In 2015, I used 0.27-g LB-2X radios with a shorter nominal life of 12 days. The shorter the battery life of the transmitter, the longer range it has. Radios had a range of detection 0.5–1 km in open habitats when used with a 3 or 5element Yagi antenna.

After transmitter activation, I trimmed the fur between the bats' scapulars to provide a good surface on which to apply transmitters. A thin layer of Perma-Type Surgical Cement was applied to the skin. I applied another layer of glue to the exposed surface of the radio-transmitters and folded the hair back onto the transmitter. Bats were then held in a cloth bag (AviNet Inc. Dryden, New York) for 20 min to allow the glue to dry. Bats were tracked to their roost every day for the life of the battery using a TRX-1000s receiver (Wildlife Materials, Murphysboro, Illinois).

I searched for transmittered bats along roads, levees, forest roads, and by walking (approximately 800-m x 300-m) transects through the woods. On several occasions once signal was detected, I biangulated or triangulated the signal to narrow down my search

area by drawing where bearings intersected on Google Earth (Google Inc.) and searching in proximity to the intersection. I opportunistically used aerial-tracking to locate bats twice in 2014. A Cessna 182 Skylane, flown by Arkansas Civil Air Patrol and equipped with an ATS R4500 scanner-receiver and two 4-element Yagi antennas attached to wing struts, flew over capture sites. For detailed methods on aerial-tracking see Moore (2015). I used a handheld GPS unit (Garmin, Schaffhausen, Switzerland) to mark roost trees once identified.

Emergence counts

Emergence counts were conducted outside roost trees by 2–3 observers. Bats were tallied using a clicker-counter and adding one for every bat exiting the tree and subtracting one for every bat reentering the tree. Once bats stopped exiting the tree for at least 15 min, a timer was set for 10 min with 1 min added to the countdown for every bat that exited the tree until the timer expired. Counts from all observers were averaged. I then listened for bat activity in cavity trees and added one to our emergence count if bats were still present within the tree.

Habitat Characteristics and Vegetation Sampling

For each identified roost tree, I recorded tree species, diameter at breast height (DBH; cm) using a metric diameter tape (Forestry Suppliers, Inc., Jackson, Mississippi), canopy cover (%) using a convex spherical crown mirror (Forestry Suppliers, Inc.), basal area (ha) using a factor-10 base prism (Forestry Suppliers, Inc.), and tree height (m) using a clinometer (Forestry Suppliers, Inc.). True DBH of some trees was difficult to obtain

because basal swelling did not decrease at a low enough height that would allow for optimal measuring (i.e., measuring 0.6-m above any basal swelling).

Each roost tree was paired with a random tree. Random trees were selected by using a random number generator to determine distance and direction of the random tree from the roost tree. Distance was determined by generating a random number between 40-100 m without replacement, and direction was determined by generating a random number from $0-360^{\circ}$ without replacement. These ranges were chosen to ensure plots for roost and random trees did not overlap. Trees were not counted twice if two radioed bats of the same sex used the same tree in a single season. However, if different sexes used the same tree, this roost tree was counted twice, but each sex received its own random tree. I used a modified Breeding Biology Research and Monitoring Database protocol (BBIRD; Martin *et al.*, 1997) to characterize microhabitat around roost trees and random trees by recording tree species and diameter at breast height of each tree within 5-m (roost-site scale) and 11.3-m (roost-patch scale) plots.

Data Analysis

I analyzed roost and random tree measurements in Program R v. 3.2.2 (R Core Team, 2016). I checked for correlation among all variables (DBH, canopy cover, basal area, and tree height). Variables were discarded if the correlation coefficient was above 0.70 (Perry and Thill, 2007). I built generalized linear mixed models with a binomial error distribution, with the binary (roost or random) variable as the response, the tree and plot variables as predictors, and the bat ID as a random effect to account for pseudoreplication of bats using multiple trees. Model selection was performed using the backwards stepwise approach to identify patterns of selection for both species. I used an

information-theoretic approach based on Akaike Information criterion corrected for small sample size (AICc; Burnham and Anderson, 2002). The best model was the model with the lowest AICc. If models had a Δ AICc < 2, I followed the principle of parsimony, keeping the model with the fewest parameters.

RESULTS

Mist-netting, harp-trapping and banding

I documented six species of bats within the CRNWR. The most common capture via mist-netting was Rafinesque big-eared bat (n = 156; 33.9% of total captures) followed by eastern red bat (*Lasiurus borealis*; n = 104; 22.6%), southeastern myotis (n = 91; 19.7%), evening bat (*Nycticeius humeralis*; n = 58; 12.6%), tri-colored bat (*Perimyotis subflavus*; n = 54; 11.7%), and big brown bat (*Eptesicus fuscus*; n = 1; <0.01%). Harp-trapping known southeastern myotis roost trees yielded 423 additional captures of southeastern myotis (Table 3.1). Harp-trapped trees contained both sexes, and both age classes. There were no pregnant southeastern myotis in the roost trees, but some females were lactating, or post-lactating.

Radiotelemetry

Thirty-two transmitters were affixed to southeastern myotis and Rafinesque's bigeared bats during the study (Table 3.3). I successfully tracked 8 of 9 affixed Rafinesque's big-eared bats and identified 20 trees in which they roosted. I successfully tracked 12 of 23 radioed southeastern myotis affixed from which I identified 19 roost trees. Aerialtracking resulted in the location of one *M. austroriparius* roost tree.

Water tupelo (Fig. 3.2 and Fig 3.7) were used most frequently by the southeastern

myotis (n = 15 trees; Table 3.2 and 3.4), followed by black gum (*N. sylvatica*, Marsh, n = 2), bald cypress (*Taxodium distichum*, L) Rich, n = 1), red maple (*Acer rubrum*, L, n = 1), and sweet gum (*Liquidambar styraciflua*, L, n = 1). Rafinesque's big-eared bat primarily roosted in water tupelos (n = 15; Table 3.2 and 3.4; Fig. 3.7), followed by bald cypress (n = 4), and American hornbeam (*Carpinus caroliniana* (Mill.) Koch, n = 1). Tree species within 11.3-m roost plots were similar between southeastern myotis and Rafinesque's big-eared bat with the top three tree species being *Nyssa aquatica* (n = 821; 55%), *Liquidambar styraciflua* (n = 118; 7.8%), and *Taxodium distichum* (n = 104; 6.9%).

Sixty percent of located trees for both bat species had basal openings (n = 24), 17.5% (n = 7) had chimney openings, 12.5% had neither chimney, window nor basal openings (i.e., a small opening on the basal swelling of the tree), 7.5% had small window openings (n = 3), and 2.5% (n = 1) had both a chimney and a basal opening. Ninety percent of roost trees were mostly (> 50%) surrounded by water. The remaining 10% of roost trees were on slightly higher ground and away from water. Roost trees used by both southeastern myotis and Rafinesque's big-eared bat were located by flowing bayous or within or on the periphery of Beaver Lake and were sometimes clustered on the landscape (Figs. 3.3–3.6).

Emergence counts

Eleven emergence counts were conducted over two years. Six water tupelos with basal openings were used by southeastern myotis in groups of 62–467 bats; one black tupelo used by southeastern myotis had 68 bats. One water tupelo had a mixture of southeastern myotis and Rafinesque's big-eared bats with 35 individuals. Trees in which only Rafinesque's big-eared bats roosted were all bald cypress and counted 23–63

individuals. One hollow bald-cypress tree used by Rafinesque's big-eared bats had an emergence count of 56 individuals.

Patterns of roost selection

No variables for either species had a correlation coefficient of over 0.70 (Table 3.5). Although DBH, canopy cover and basal area tended to be higher for roost trees than for random trees (Table 3.4; Fig. 3.8), the best parsimonious model for southeastern myotis was the null model (Table 3.6), suggesting that southeastern myotis did not select roost trees based on the considered habitat characteristics. For the Rafinesque's big-eared bats, DBH and basal area seemed to differ between roost and random trees (Fig. 3.9), but the model that best explained roost selection was with DBH only (Table 3.6).

DISCUSSION

Bottomland hardwood forests may be important for the conservation of both the southeastern myotis and Rafinesque's big-eared bat because they are often associated with tree species that can be used as roosts by both species. Despite the null model being the best for the southeastern myotis, characteristics of roosts i.e., larger diameter, higher canopy cover, may still be important as they can provide thermal benefits to bats roosting in the tree. DBH influenced roost selection for Rafinesque's big-eared bats. The use of larger diameter trees that are in proximity to water by both southeastern myotis and Rafinesque's big-eared bats may be related to stable microclimates through isolative properties and promoting a humid environment which may promote juvenile growth during critical stages of development (Kunz, 1982). Larger diameter trees are correlated with larger cavity height (Miller *et al.*, 2011; Rice, 2009), which may provide more space

for communal living (Kunz and Fenton, 2003) and a stable microclimate for juvenile development (Kunz, 1982; Neuweiler, 2000). Furthermore, prevention of solar radiation from reaching the tree because of high canopy cover may also help in creating an ideal microclimate and preventing the bat's body temperature from going outside their thermal-neutral-zone (Gooding and Langford, 2004; McNabb, 1982; Menzel *et al*, 2002; Neuweiler, 2000; Rice, 2009; Sedgeley, 2001; Vonhof and Barclay, 1996).

Also, mixed communal living was observed in most roost trees of both bat species through emergence counts and harp-trapping. This suggests the habitat is suitable for large numbers of bats to group together or that clustering together may be crucial behavior in regards to maintaining an ideal microclimate (Kunz and Fenton, 2003; Menzel *et al.* 2002; Sedgeley, 2001). The mixing of males in roost during the reproductive season is similar to other studies where male southeastern myotis were found roosting with reproductive females (Reed, 2004; Rice, 1957). Alterations of canopy cover manually or naturally may influence the microclimate within the tree (Carver and Ashley, 2008).

Additionally, use of *N. aquatica*, *T. distichum*, *L. styraciflua*, and *N. sylvatica* by southeastern myotis is consistent with similar studies of the species (Carver and Ashley, 2008; Hoffman *et al.*, 1999; Lucas *et al.* 2015; Rice, 2009; Stuemke *et al*, 2014). Likewise, use of *N. aquatica* and *T. distichum* has been well documented for the Rafinesque's big-eared bat (Carver and Ashley, 2008; Clement and Castleberry, 2013; Gooding and Langford, 2004; Johnson and Lacki, 2013; Martin *et al.*, 2011; Lucas *et al.*, 2015, Rice, 2009; Stuemke *et al.* 2014). Findings are also similar to other studies in that they are surrounded by or in proximity to permanent water sources both lentic and lotic,

such as lakes and bayous (Johnson and Lacki, 2013; Lucas *et al.* 2015; Medlin and Risch, 2008). In 2014, record rainfall (~ 30.5 cm) in June may have forced some bats to use trees without cavities or chimney openings due to rising water levels obstructing the basal opening. One bat roosted on the outside of tree immediately following a rain event. Other bats that roosted on the outside of trees were not located following a rain event, and basal openings were exposed in surrounding trees. One Rafinesque's big-eared bat (150.376) was located on the exterior of two different trees, one of which contained an accessible basal opening. Other studies of the roosting ecology of both southeastern myotis and Rafinesque's big-eared bat (Gooding and Langford, 2004; Rice, 2009; Stuemke, *et al.* 2014; Trousdale, 2011) noted the use of basal and chimney openings on roost trees. However, no study has documented window openings in trees. Window openings were approximately 1 m or higher from the base of the tree. These openings may be beneficial to both species during times of flooding as the opening may be less likely to be obstructed by water during floods.

Both *N. aquatica* and *T. distichum* are strongly associated with habitats that are permanently flooded or experience long periods of inundation (Mitsch and Gosselink, 1993) and the roost plots of both species were dominated by both tree species. Use of roost trees in these lentic and lotic systems may present a problem to the bats when extreme rain events cause water levels to rise above and obstruct basal openings. The problem may be circumvented if a chimney or window opening is present. However, I found that 60% of roost trees in our study did not possess any other opening than basal.

The southeastern myotis and Rafinesque's big-eared bat both show site fidelity and preference to habitats dominated by water tupelo and bald cypress trees (Trousdale,

2011). This suggests that mature cypress-tupelo forests play an important role in the roosting ecology of both species where caves are absent. Land managers should give consideration to larger water tupelos and bald cypresses with basal and chimney openings when cruising timber and managing trees not only in cypress-tupelo stands but in adjacent tree stands. Several identified roost trees were adjacent to small patches of clear cutting within the Biscoe Bottoms tract of the CRNWR. Knowing that some bat of both species selected tree species (i.e., red maple, and black tupelo) in proximity to cypress-tupelo stands, land manager should consider buffers around cypress-tupelo before implementing any silviculture practices.

Although mixed communal roosting has been documented for the southeastern myotis, it has never been documented in such high numbers. The thermal requirements between male and female are likely different in the summer, hence formation of maternity colonies (Benton and Scharoun, 1958; Davis *et al.*, 1965; Sedgeley. 2001; Stegeman, 1954). Although summers in Arkansas are warm (27–33°C; Arkansas Weather, 2016), this study did not investigate temperature as a predictor variable. Since temperature and microclimate are suggested to play an important role during the summer months, I suggest future studies should measure other variables that may explain roosting preference, such as internal temperature and humidity of the tree cavity and investigate differences between males and females and perhaps give insight to potential reasons for such high mixed communal roosting. In addition, true DBH was difficult to obtain for some larger trees with basal swelling occurring approximately 3.7 m from the ground. This was seen with roost trees used by both species. Development of methods to obtain true DBH may give better insight to what degree DBH is truly influential.

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,	0	,	U				
	Μ	ale		Female			
	NR	REP	NR	PREG	LAC	PL	Total
Juvenile	76	0	40	0	0	0	
Adults	69	78	123	0	5	35	
Total	2	23		203			423

Table 3.1 – Southeastern myotis counts at harp-trapped tree during summers 2014 - 2015 in Cache River National Wildlife Refuge. NR = Non-reproductive, REP = Reproductive male, PREG = Pregnant, LAC = Lactating, PL = Post-lactating.

Table 3.2 – Transmittered bat characteristics: species i.e., southeastern myotis (MYAU) and Rafinesque's big-eared bat (CORA), sex, age, reproductive status (Repro.). Reproductive status includes non-reproductive (NR), pregnant (PREG), lactating (LAC), and post-lactating (PL), weight, left forearm length (LFA) and Bat I.D. (associated with transmitter frequency).

							Bat	Successfully
Year	Species	Sex	Age	Repro.	Weight (g)	LFA (mm)	I.D.	Located
2014	MYAU	Μ	А	NR	7.0	36.00	150.948	Yes
	MYAU	Μ	А	NR	6.5	38.80	150.906	Yes
	MYAU	F	А	LAC	9.0	38.00	150.676	No
	MYAU	Μ	А	NR	7.5	35.50	150.631	No
	MYAU	F	А	PREG	8.5	38.00	150.550	Yes
	MYAU	F	А	LAC	7.5	37.70	150.435	No
	MYAU	Μ	А	NR	7.0	35.90	150.307	No
	MYAU	F	А	LAC	8.0	38.50	150.150	No
	MYAU	Μ	А	NR	7.0	36.20	150.212	Yes
	MYAU	F	А	PL	8.0	36.10	150.190	Yes
	MYAU	F	J	NR	8.0	37.80	150.868	Yes
	MYAU	Μ	J	NR	6.5	36.50	150.218	No
	MYAU	Μ	А	NR	7.0	37.10	150.148	Yes
2015	CORA	F	А	PREG	14.0	42.25	150.027	Yes
	CORA	F	А	PREG	14.5	41.34	150.009	Yes
	MYAU	F	J	NR	7.5	38.33	150.068	Yes
	MYAU	F	А	LAC	8.0	39.16	150.108	No
	MYAU	Μ	А	NR	8.0	38.10	150.150	No
	CORA	Μ	А	NR	11.0	40.75	150.550	Yes
	CORA	Μ	А	NR	9.0	42.88	150.211	No
	CORA	Μ	А	SCR	9.0	39.62	150.376	Yes
	CORA	F	А	LAC	10.5	44.18	150.394	Yes
	MYAU	Μ	А	NR	6.0	37.14	150.351	Yes
	MYAU	F	А	NR	7.0	37.80	150.949	Yes
	CORA	Μ	J	NR	7.5	44.96	150.912	Yes
	MYAU	Μ	А	NR	7.0	34.78	150.867	Yes
	CORA	F	J	NR	9.0	44.49	150.829	Yes
	MYAU	Μ	J	NR	8.0	37.45	150.795	Yes
	MYAU	Μ	А	NR	6.5	37.47	150.748	No
	CORA	Μ	А	NR	11.0	42.56	150.713	Yes
	MYAU	Μ	А	NR	7.0	33.70	150.671	No

Table 3.3 – Roost tree data of both the southeastern myotis and Rafinesque's big-eat bat. Bat. I.D. signifies bats from Table 3.2. Species includes black tupelo (NYSY), red maple (ACRU), sweetgum (LIST), water tupelo (NYAQ), bald cypress (TADI), and American hornbeam (CACA). Tree measurements include diameter at breast height (DBH), basal area (BA), and canopy cover (CC).

Bat I.D.	Plot No.	Species	DBH (cm)	BA (m ² /ha)	CC (%)	Height (m)
150.948	1	NYSY	160.02	8.36	93	19.9
150.906	1	ACRU	105.66	10.68	74	18.9
150.550	1	LIST	40.89	18.58	96	18.9
150.190	1	NYAQ	194.82	27.87	95	31.1
150.212	1	NYAQ	252.48	25.55	97	47.6
150.212	2	NYAQ	216.15	12.07	97	28.0
150.868	2	NYAQ	211.33	14.86	47	32.0
150.868	3	NYAQ	178.86	14.40	46	28.0
150.148	1	NYAQ	298.96	14.86	95	29.9
150.148	2	NYAQ	208.03	18.58	87	25.9
150.868	4	NYAQ	220.47	12.54	93	29.6
150.027	1	NYAQ	355.98	19.04	25	22.6
150.068	1	NYAQ	121.92	14.86	93	19.5
150.027	2	NYAQ	314.96	8.83	51	7.6
150.027	3	TADI	384.05	8.36	85	39.6
150.351	1	NYAQ	529.59	6.87	98	39.0
150.394	1	NYAQ	529.59	14.40	98	39.0
150.376	1	CACA	97.79	8.36	91	29.0
150.376	2	NYAQ	313.69	21.37	92	21.9
150.351	2	NYAQ	400.05	14.40	88	30.2
150.394	2	NYAQ	495.3	18.12	92	22.9
150.376	3	NYAQ	101.35	16.72	92	15.2
150.351	3	NYAQ	406.65	15.30	81	29.0
150.394	3	NYAQ	367.79	30.66	84	18.3
150.394	4	TADI	457.71	15.79	86	50.3
150.394	5	NYAQ	242.82	14.40	84	9.1
150.949	1	NYAQ	378.71	14.40	87	40.5
150.912	1	TADI	762.51	11.61	86	30.2
150.912	2	NYAQ	431.8	15.30	85	46.6
150.829	1	NYAQ	322.58	19.51	80	25.9
150.829	2	NYAQ	431.8	15.30	85	46.6
150.829	3	TADI	637.54	6.04	74	46.6
150.867	1	NYAQ	378.71	14.40	87	40.5
150.867	2	NYAQ	435.61	14.40	85	39.6
150.912	3	NYAQ	292.61	13.00	78	16.8
150.912	4	NYAQ	377.19	13.00	88	29.9
150.912	5	NYAQ	508.64	9.29	84	9.1
150.975	1	TADI	518.16	16.26	76	38.1

Table 3.4 – Characteristics (\pm SE) of roost and random trees for southeastern myotis (MYAU; n = 19) and Rafinesque's big-eared bats CORA; (n = 20) during summers 2014 and 2015 in the Cache River National Wildlife Refuge, AR. The sample size for roost and random trees is denoted by n

	MYAU roost	MYAU random	CORA roost	CORA random
Diameter at breast height (cm)	276.69 ± 32.3	134.04 ± 29.8	387.03 ± 35.4	139.1 ± 20.7
Canopy Cover (%)	84.94 ± 3.5	75.97 ± 5.8	79.47 ± 3.9	72.2 ± 2.5
Basal area (m ² /ha)	163.89 ± 12.3	123.61 ± 7.2	158.1 ± 14.3	155 ± 13.8
Tree height (m)	35.44 ± 5.3	24.84 ± 4.0	$27.7\pm~3.2$	26.75 ± 3.2

Table 3.5 – Spearman correlation coefficients among variables of both southeastern myotis and Rafinesque's big-eared bat roost and random trees. Variables include diameter at breast height (DBH), canopy cover (CC), basal area (BA), and tree height (H). *r* indicates correlation coefficient.

(a) <i>M. austroriparius</i> $(n = 19)$	r	(b) <i>C. rafinesquii</i> $(n = 20)$	r
DBH, CC,	0.03	DBH, CC,	0.41
DBH, BA	0.26	DBH, BA	0.34
DBH, H	0.65	DBH, H	0.21
CC, BA	0.05	CC, BA	0.29
CC, H	0.03	CC, H	0.28
BA, H	0.33	BA, H	0.09

Table 3.6 – Top 5 linear, mixed-effects models of roost trees selection of *M. austroriparius* (a) and (b) *Corynorhinus rafinesquii* (b) with the response variable being roost tree or random tree and diameter at breast height (DBH), canopy cover (CC), basal area (BA) and tree-height (H) as predictor variables for 2014 and 2015 in the Cache River National Wildlife Refuge. K represent number of parameters. AICc = Akaike Information Criterion. Δ AICc = Difference in AICc of most parsimonious model and current AICc. AICwt = Weight of the model/relative support of the model.

Model Predictors	Κ	AICc	ΔAICc	AICwt
(a) M. austroriparius				
Null model	3	67.95	0.00	0.68
BA	4	67.94	1.79	0.28
DBH	4	75.27	7.31	0.02
Н	4	76.84	8.88	0.01
CC	4	76.88	8.92	0.01
(b) C. rafinesquii				
DBH	4	57.54	0.00	0.97
DBH + H	5	64.80	7.26	0.03
Null	3	64.95	10.42	0.01
BA	4	71.57	14.03	0.00
DBH + BA+H	6	72.79	15.25	0.00



Figure 3.1 – Black tupelo (*Nyssa sylvatica*). Identified as roost tree of *M. austroriparius* tree in Cache River National Wildlife Refuge. Harp-trapped in July, 2015.

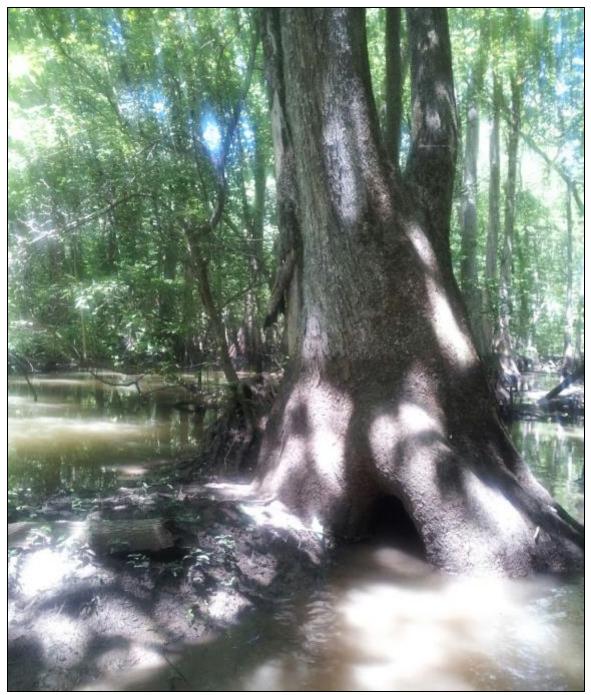


Figure 3.2 - Water tupelo (*Nyssa aquatica*) with basal opening. Roost tree of *M. austroriparius*, 2014.

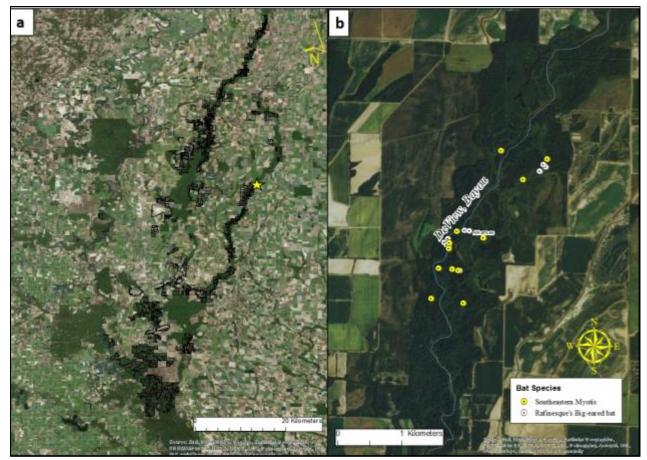


Figure 3.3 - Distributions of southeastern myotis and Rafinesque's big-eared bats on Bayou de View tract, Cache River National Wildlife Refuge, 2014 - 2015. Star on map A indicates location of map B on Refuge. Yellow dots indicated location of southeastern myotis roost trees, white does indicated Rafinesque's big-eared bat roost trees.

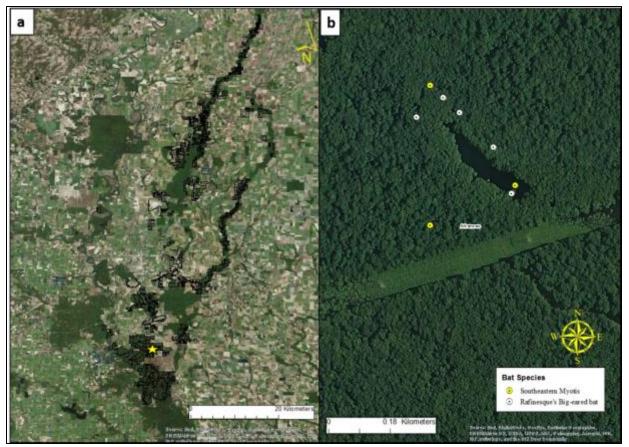


Figure 3.4 – Distributions of southeastern myotis and Rafinesque's big-eared bats on Biscoe Bottoms North tract of Cache River National Wildlife Refuge, 2015. Star on map A indicates location of map B on Refuge. Yellow dots indicated location of southeastern myotis roost trees, white does indicated Rafinesque's big-eared bat roost trees.

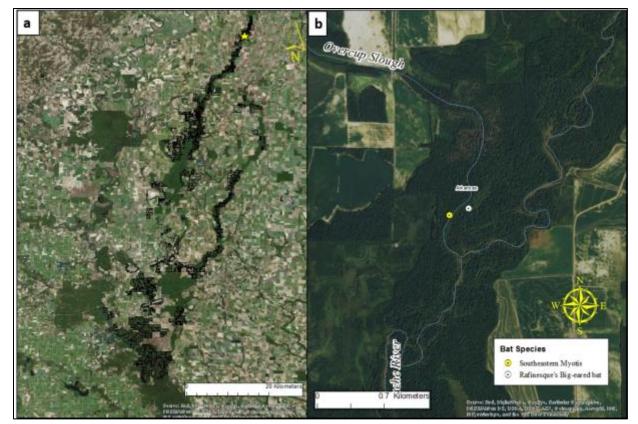


Figure 3.5 - Distributions of southeastern myotis and Rafinesque's big- eared bats on Nicholson tract of Cache River National Wildlife Refuge, 2014 - 2015. Star on map A indicates location of map B on Refuge. Yellow dots indicated location of southeastern myotis roost trees, white does indicated Rafinesque's big-eared bat roost trees.

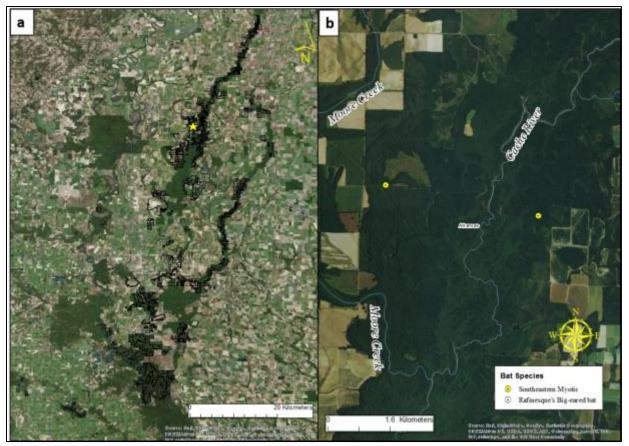


Figure 3.6 – Distributions of southeastern myotis on Penn's Bay / Walker Cemetery of Cache River National Wildlife Refuge, 2014. Star on map A indicates location of map B on Refuge. Yellow dots indicated location of southeastern myotis roost trees, white does indicated Rafinesque's big-eared bat roost trees.

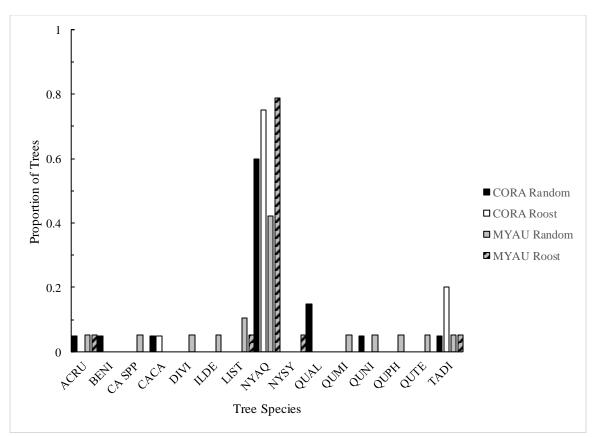


Figure 3.7 – Proportion of roost and random trees of the southeastern myotis (MYAU) and roost and random trees of the Rafinesque's big-eared bat (CORA). Tree species include red maple (ACRU), river birch (BENI), hickory species (CA Spp), American hornbeam (CACA), persimmon (DIVI), sweet gum (LIST), water tupelo (NYAQ), black tupelo (NYSY), white oak (QUAL), swamp chestnut oak (QUMI), water oak (QUNI), willow oak (QUPH), Nuttall oak (QUTE), and bald cypress (TADI).

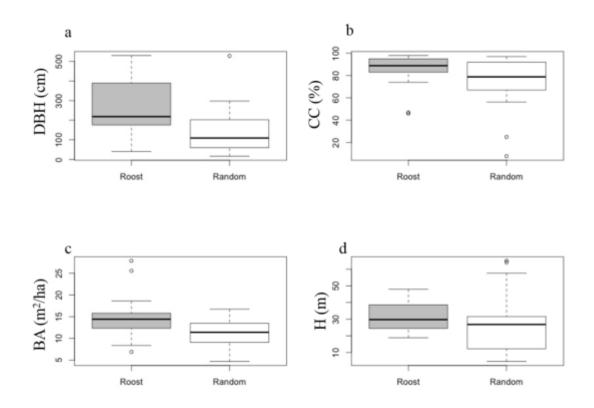


Figure 3.8 – Characteristics of roost and random trees of the southeastern myotis: a) diameter at breast height (DBH), b) canopy cover (CC), c) basal area (BA), and d) tree height (H).

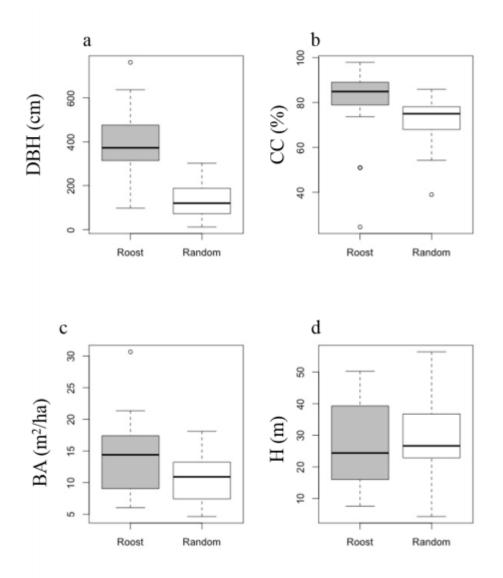


Figure 3.9 – Characteristics of roost and random trees of the Rafinesque's big-eared bat: a) diameter at breast height (DBH), b) canopy cover (CC), c) basal area (BA), and d) tree height (H).

CHAPTER IV

CONCLUSION

There is a considerable lack of knowledge regarding the bat communities in the bottomlands of Arkansas, specifically the distribution of bats of Cache River National Wildlife Refuge (Fokidis *et al.*, 2005; Medlin *et al.*, 2006; Medlin and Risch, 2008; Medlin *et al.*, 2010). The purpose of this research was to increase knowledge of the distribution of bats, document occupancy of bats throughout 5 predefined habitats, and roosting ecology of the southeastern myotis and Rafinesque's big-eared bat of the Cache River National Wildlife Refuge (CRNWR).

First, mist-net surveys suggest a diverse bat community with 6 species of bats on the CRNWR out of the 16 species present in the state of Arkansas. The Rafinesque's bigeared bat (*Corynorhinus rafinesquii*) was the most common capture followed by eastern red bats (*Lasiurus borealis*), southeastern myotis (*Myotis austroriparius*), evening bats (*Nycticeius humeralis*), tri-colored bats (*Perimyotis subflavus*), and big brown bats (*Eptesicus fuscus*). In addition to capture data, no threatened and endangered (T&E) species were detected acoustically. Capture and acoustic data support my first prediction: no T&E species would occur on the CRNWR.

Myotis bats tended to have higher occupancy estimates in cypress-tupelo habitat as compared to other habitat types. High *Myotis* occupancy in cypress-tupelo habitats

may be related to high numbers of mist-net captures of the southeastern myotis within the refuge and their affinity for cypress-tupelo habitat (Hoffman, 1999; Rice, 1957; Rice, 2009; Stuemke, 2015). No Rafinesque's big-eared bat calls made it past the 85% confidence filter and no occupancy models could be run. Results partially support my prediction for the first hypothesis for objective 2: *Myotis* bats had highest occupancy in cypress-tupelo habitats.

Tri-colored bats tended to have highest occupancy throughout all habitats but highest occupancy in mature forest habitat, which cover over 65% of the refuge. Tricolored bats have higher amplitude calls and are easily detectable by bioacoustic devices, which may explain the high overall occupancy detection rates, which may have biased results. It is difficult to say if habitat type determines tri-colored bat occupancy based on the results. Evening bats and big brown bats tended to have highest occupancy ratings at managed hardwood habitats, which is the second smallest habitat on the refuge, similarities, may be due to similarities in overlap of habitat usage.

This study started because an Indiana bat (*Myotis sodalis*) was detected in the CRNWR in Jackson County on a bioacoustic device prior to the study. Before implementing any management practices, an inventory of bat species was needed for the refuge. As seen with this study, relying on just one inventory method (i.e., mist-nets or bioacoustics) can give spurious results and lead to management decisions that are detrimental instead of beneficial to bat species.

Finally, using radio-transmitters I radio-tracked 23 southeastern myotis in 2014 and 2015 and 9 Rafinesque's big-eared bats in 2015. I identified 19 roost trees for southeastern myotis and 20 for Rafinesque's big-eared bat. Both species used bald

cypress (*Taxodium distichum*) and water tupelo (*Nyssa aquatica*). However, the Rafinesque's big-eared bat was the only one to roost in American hornbeam (Carpinus *caroliniana*) and the southeastern myotis was the only one to roost in black gum (N. sylvatica), sweet gum (Liquidambar styraciflua), and red maple (Acer rubrum). Southeastern myotis bats congregated in higher numbers (up to 467 individuals) than Rafinesque's big-eared bats (up to 63 individuals). Roost plots were associated with high numbers of water tupelo, sweetgum (Liquidambar styraciflua), and bald cypress trees with basal openings, both of which are associated with wetland habitat. These results are similar with other studies of both species (Carver and Ashley, 2008; Clement and Castleberry, 2013; Johnson and Lacki, 2013; Lucas et al. 2015; Stuemke et al., 2014) and support my third hypothesis: roost trees and habitat of the southeastern myotis and Rafinesque's big-eared bat are similar. Out of all the predictor variables (i.e., diameter at breast height, canopy cover, basal area, and tree height), diameter at breast height was the only variable that predicted roosting for the Rafinesque's big-eared bat, whereas none of the variable influenced roosting for the southeastern myotis.

In conclusion, the preference of both bat species to roost in bald cypress and water tupelo cavities, combined with high occupancy rates of *Myotis* bats in cypress-tupelo habitat provide strong support that both species have high affinity towards bottomland hardwood forests, particularly with cypress-tupelo stands, in absence of cave systems. Other bat species were using the Refuge, although I could not confirm the presence of Indiana bats.

This study was conducted during the breeding season of bats in Arkansas. I suggest exploring roosting ecology of the southeastern myotis and Rafinesque's big-eared

bat in fall and winter months since both species use floodplains, and cypress-tupelo swamps and roost in tree cavities and exhibit activity (i.e., roost switching, emergence for water) during winter months, however, the Rafinesque's big-eared bat experiences light torpor (Johnson, 2012). This shallow torpor may explain why arousal during the winter is more frequent (Boyles *et al.*, 2006; Clement and Castleberry, 2014; Johnson, 2012; Johnson and Lacki, 2012; Harvey *et al.* 1999; Neuweiler, 2000, Rice, 2009). Management practices should focus on trees used both breeding and non-breeding season. Selection of roost trees in the summer may be related to microclimates that promote growth of pups, whereas selection for trees in fall and winter may differ due to different biological pressures i.e., reproductive status, hibernation, and selecting trees that fulfill potential thermal requirements.

In addition, long periods of flooding during winter months may trap bats within the tree if chimney openings are absent and basal openings are obstructed by water, therefore, trees that provide other openings other than basal openings may be used more to avoid potentially becoming trapped and starving within the tree. Documenting winter months will help fill in knowledge gaps concerning winter roosting ecology of both species. In addition, documenting variables such as internal temperature and humidity of cavity trees in the summer in concert with other predictor variables may expose patterns of selection for both species. Having both summer and winter roosting requirements of both bats will help land managers develop strategies that will benefit both species throughout the year.

Several southeastern myotis with radio affixed to them were never successfully located but were detected close by capture sites following initial capture. Therefore, I

suggest aerial-telemetry as an alternative approach to locate bats on the landscape for use in foraging studies. Little is known of foraging strategies of the southeastern myotis and Rafinesque's big-eared bat (but see Lacki and Bayless 2013). In addition to moving far from roosting sites, natural and manmade barriers (i.e., levies) may prohibit or hinder studies that focus on foraging of the southeastern myotis and difficulties may be alleviated with the use of aerial-telemetry (e.g., Moore 2016). Future studies should also include more variables (i.e., humidity and temperature within the tree) and a method to obtain true DBH to not bias results.

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