

FALL AND WINTER ROOSTING ECOLOGY OF SOUTHEASTERN MYOTIS AND
RAFINESQUE'S BIG-EARED BATS IN THE CACHE RIVER NATIONAL
WILDLIFE REFUGE

STACY JAE SCHERMAN

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Approved by
Dr. Virginie Rolland, Advisor
Dr. Thomas Risch, Committee Member
Dr. Yeonsang Hwang, Committee Member

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ABSTRACT

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Tree roosts in bottomland forests are critical resources required year-round for Rafinesque’s big-eared bats (*Corynorhinus rafinesquii*; CORA) and Southeastern myotis (*Myotis austroriparius*; MYAU). I characterized their roost selection and activity patterns in fall and winter in Cache River National Wildlife Refuge, Arkansas. In October-December of 2016 and 2017, I radio-tracked 19 CORAs and 14 MYAUs to 33 and 42 roost trees, respectively. Both species switched their roosting habits (e.g., MYAUs switched tree species and CORAs spent more time in roosts), but remained active when temperatures approached freezing. Additionally, CORA selected trees with large internal volume and diameter at breast height (DBH) within plots of predominately water tupelos and bald cypresses, whereas MYAU selected trees with large internal volume and high cavity openings within plots of medium DBH trees where bald cypresses were absent. Overall, CORAs and MYAUs seemed to anticipate seasonal flooding that could potentially trap them inside cavities.

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CHAPTER 1: INTRODUCTION

1.1 Impacts of Bats

Across the globe bats provide ecological services in pollination, seed dispersal, pest control, fertilization, and prey for predators (Fenton and Simmons 2014). Bats also impact their ecosystems because regardless of what they eat, they tend to eat much more per gram of body mass than other mammals to meet their energy requirements for flight (Fenton and Simmons 2014).

Almost all bat species in North America are insectivorous and directly benefit humans by providing a natural form of insecticide (Harvey et al. 2011). If bat populations decline drastically, the economic consequences could be significant (Boyles et al. 2011). Of all the species of moths eaten by members of *Corynorhinus*, 19 are considered to be crop pests in the larval stage (Lacki and Dodd 2011). A two-year study showed that big brown bats (*Eptesicus fuscus*) can eat over 200 species of insects and that little brown bats (*Myotis lucifugus*) can consume over 550 insect species (Fenton and Simmons 2014). Insectivorous bats regularly eat more than 50% of their body weight in insects each night (Harvey et al. 2011). A colony of one million Brazilian free-tailed bats (*Tadarida brasiliensis*) can consume about 500–1000 tons of insects in a single summer (Altringham 2011). Another example is a small colony of big brown bats in Indiana that can consume

1.3 million pest insects per year (Boyles et al. 2011). Although estimating the economic importance of bats to agriculture is difficult, Boyles et al. (2011) estimates the value of pest control by bats at an average of \$183/ha in a cotton-dominated agricultural landscape in south-central Texas. If this value is extrapolated to the entire nation, bat pest suppression would then range \$3.7-53 billion/year, highlighting the massive economic importance of bats in forestry and agriculture (Boyles et al. 2011).

1.2 Threats to Bats

The order Chiroptera has 78 species that are considered Endangered or Critically Endangered and 99 species that are considered Vulnerable (IUCN 2016). If a bat population experiences a severe decline, its recovery is challenged by relatively slow population growth due to their low reproductive rates, which increases the risk of local extinctions (Racey and Entwistle 2003). Therefore, it is important to evaluate the factors that impact bat populations in North America including wind turbines, white-nose syndrome (WNS), persecution due to lack of understanding and habitat degradation.

First, large numbers of bats are killed at utility-scale wind energy facilities across the United States as well as Europe and Australia (Kunz et al. 2007a, 2007b; Arnett et al. 2008). Although some fatalities are caused by direct contact with the rotor blades, up to 50% of the fatalities are caused by barotrauma, which is lethal tissue damage to internal structures resulting from extreme and rapid pressure changes (Kunz et al. 2007b; Baerwald et al. 2008). The rapid air-pressure reduction is produced by the moving turbine blades and is undetectable by sight or echolocation (Baerwald et al. 2008). Most regions

of the United States report fatality rates of 0.8–8.6 bats/MW/year (Kunz et al. 2007a). Ground searches at wind facilities specify fatalities in 11 species of bats, many of which are residents during summer months (Kunz et al. 2007b; Arnett et al. 2008). The highest numbers of bat fatalities are in late summer and early autumn, which aligns with the migration of tree-roosting bats (Kunz et al. 2007b, Arnett et al. 2008; Baerwald et al. 2008). Wind energy is one of the fastest growing sectors in the energy industry. In 2013, wind energy supplied 4.5% of the nation's energy and a U.S. Department of Energy (2015) report suggests it is possible to increase to 20% by 2030. The projected growth of the industry raises serious concerns about the cumulative impacts on bat populations (Kunz et al. 2007a, 2007b; Arnett et al. 2008).

Second, WNS was first discovered in North America in bats hibernating in Howe Cave near Albany, NY in 2006 and has been spreading rapidly across North America, causing sharp declines in bat abundance (Blenhert et al. 2009; Foley et al. 2011; Langwig et al. 2015). The disease is caused by the fungus, *Pseudogymnoascus destructans* that grows on the muzzle, ears, wing membranes, and other tissues of the bat (Blenhert et al. 2009; Frick et al. 2010; Foley et al. 2011). The fungus grows well at temperatures 3–15°C and relative humidity greater than 90%, which are common winter conditions in caves, mines, and rock crevices where more than half of North American bat species hibernate in large numbers during the winter (Blenhert et al. 2009; Foley et al. 2011; Langwig et al. 2015). The physiology of infected individuals is disrupted by the fungus and they arouse from hibernation more often than healthy bats, which drastically depletes their fat reserves and causes dehydration, often leading to mortality (Foley et al. 2011; Langwig et al. 2015). The fungus is transmitted mainly by bat to bat contact, but can also be spread by human

or animal vectors and environmental contacts (Foley et al. 2011). Infected hibernacula have experienced population decreases ranging 30–99%, with an average population decrease of 73% (Frick et al. 2010; Foley et al. 2011). WNS has killed an estimated 5–6.7 million individual bats and has been found in 11 species (Leopardi et al. 2015; BCI 2016). These unprecedented population losses pose a serious risk to the survival of North America's bat species and threaten some populations with local extinctions (Frick et al. 2010; Foley et al. 2011; Langwig et al. 2015). All hibernating bat species could be affected by WNS, but the extent to which tree-roosting bats may be affected by the syndrome remains unknown (Foley et al. 2011).

Third, because of bats' negative image with the public, human-bat conflict constitutes an additional threat. There are myths about bats being dirty, blind, or purposefully tangling themselves in people's hair (Fenton and Simmons 2014). They are also assumed to have diseases, be blood-lusting, and aggressive (Fenton and Simmons 2014; Hoffmaster et al. 2016). Bats regularly utilize human structures as roosts, which is accepted when it is a bridge, cistern, or abandoned building, but becomes problematic when it is within a person's residence (Trousdale and Beckett 2004; Hoffmaster et al. 2016). Bats that roost in human dwellings tend to be removed due to fear of bats and disease, but the stress of displacement can cause low reproductive success for that season (Hoffmaster et al. 2016). Bats can be removed safely, but many stories describe humans harming or killing bats, such as an elderly woman in Houston, TX who killed dozens of bats around her home with a cane (Benito 2016). The negative attitude of many humans towards bats is likely due to a lack of awareness and understanding of how interesting

and important these creatures really are. Public perception and understanding of bats can be improved through education programs (Hoffmaster et al. 2016).

Finally, the vandalism, disturbance to, and alteration of cave sites is a significant form of habitat degradation (McCracken 1989; Tuttle 2007). Bats in caves are vulnerable to disturbances particularly because they tend to aggregate in large colonies during hibernation or reproduction (McCracken 1989). Disturbances of any kind at a cave roost can cause bats to abandon roost sites either for the season or permanently, and decrease the survival of pups being reared at the site (McCracken 1989; Ludlow and Gore 2000). Disturbances during hibernation can cause unplanned arousals, which can prematurely deplete energy reserves (McCracken 1989). Vandals and thoughtless cave explorers caused the largest colony of Southeastern myotis (*Myotis austroriparius*) in Alabama to abandon their cave (Mount 1986). In addition, the endangered Indiana (*Myotis sodalis*) and Gray bats (*Myotis grisescens*) have abandoned some of their traditional roost caves due to commercial cave development (McCracken 1989). However, some bat colonies have benefited from the installation of cave gates restricting access to essential expeditions (Sasse et al. 2007).

Other forms of habitat degradation, are habitat loss, disturbance at roosts during hibernation or pup rearing, loss of snags and suitable cavities in large old trees, and poor forest management techniques (Tuttle 2007; Rice 2009). The functionality of ecosystems is severely reduced when there is large-scale habitat loss and fragmentation (Twedt and Best 2004). The loss of mature forest habitat has caused a reduction in foraging opportunities and changes in prey species diversity and abundance (Tuttle 2007). Continued fragmentation of forests alters travel corridors between roosting sites and

foraging sites, bat movements, and foraging patterns (Duchamp et al. 2007). Management practices that ensure tree stands of a single age or monoculture cause a change in prey insect hatching cycles and a scarcity of old trees and snags with exfoliating bark and cavities (Duchamp et al. 2007). Specifically, the Mississippi Alluvial Plain, which historically consisted of over 9.7 Mha of bottomland hardwood forest, is an example of substantial habitat loss (Tiner 1984; Twedt and Best 2004). The total amount of bottomland hardwoods in the southeastern United States has decreased by ~50% since the time of European settlement (McKee et al. 2012). Arkansas originally had 3.96 Mha of wetlands, covering 29% of the state. An overall 72% of wetlands in Arkansas were lost between 1780 and 1980, mainly as a result of conversion to agricultural land (Dahl 1990; Racey and Entwistle 2003), but also because of urban development, silvicultural practices, and hydrologic alterations (Kress et al. 1996; McKee et al. 2012).

1.3 Forest Use by Bats

Fifty-one species of bats in the U.S. use forests in varying amounts (Lacki et al. 2007). Cave-obligate bats such as Gray bats and Townsend's big-eared bats (*Corynorhinus townsendii*) use forests only for foraging (Sealander and Heidt 1990; Martin et al. 2006), whereas other bats, such as Indiana bats and Northern long-eared bats (*Myotis septentrionalis*), use caves for hibernation during the winter and then migrate into forest roosts across the landscape when temperatures increase in the spring (Kurta et al. 1993; Caceres and Barclay 2000). Twenty-seven North American bat species are considered

tree-roosting bats; these also use forests for foraging activities (Sealander and Heidt 1990; Lacki et al. 2007).

In the southeastern United States, the Gulf Coastal Plain is defined by rivers that have created and maintained broad alluvial valleys and by distinct forest communities that occur on these floodplains (Smith 1996). Floodplain forest communities are the product of thousands of years of geomorphic and hydrologic processes (Smith 1996).

Bottomlands were formed by alluvial deposits within a floodplain and are generally flat with only slight variations in elevation (Hosner and Minckler 1963). These bottomland forests are used by bats as important roosting and foraging sites (Gooding and Langford 2004; Medlin and Risch 2008). Bottomland hardwood forests characteristically contain bald cypress (*Taxodium distichum*) and water tupelo (*Nyssa aquatic*) which are roosting trees for several species of bats, particularly Rafinesque's big-eared bat (*Corynorhinus rafinesquii*) and the Southeastern myotis (Clark 1990; Rice 2009).

Arkansas consists of many protected bottomland areas including White River National Wildlife Refuge, Wapanocca National Wildlife Refuge, Little River Wildlife Management Area and Cache River National Wildlife Refuge. In northeast Arkansas, the floodplain of the Cache River maintains some of the largest contiguous tracts of intact forested wetlands that remain in the Lower Mississippi River Valley (Kress et al. 1996; Smith 1996). This area commonly experiences water level fluctuations of more than 3 m during the annual cycle (Kleiss 1996).

1.4 Roosting Ecology of Arkansas Bottomland Bats

Arkansas has 16 species of bats including at least 7 that regularly use bottomlands (Medlin et al. 2006). Two families are represented: Vespertilionidae and Molossidae. Vespertilionidae, the largest family of bats, is distributed throughout the world (Reid 2009; Sealander and Heidt 1990). The members of Vespertilionidae are known as the plain-nosed bats because they lack any skin flaps, folds, or other outgrowths on the muzzle that are representative of other bat families (Sealander and Heidt 1990; Reid 2009). These bats are aerial insectivores and scoop flying insects into their uropatagium during flight (Reid 2009).

In bottomland forests, hollow trees are a fundamental resource regularly used as day roosts (Rice 2009; Clement and Castleberry 2013). These trees provide protection from fluctuations in temperature and humidity as well as from predators and poor weather conditions, while also providing a place for social interaction, reproduction, pup rearing, and hibernation (Kunz 1982; Rice 2009; Clement and Castleberry 2013). During the winter in the temperate zone, most bats hibernate, i.e., reduce their metabolic rate and body temperature, but periodically must awaken to perform activities that maintain homeostatic balance (e.g., drink, urinate, and relocate; Foley et al. 2011). However, Southeastern myotis and Rafinesque's big-eared bats, both Vespertilionidae species, only hibernate in northern populations while southern populations remain active during much of the winter (Jones 1977; Jones and Manning 1989; Sealander and Heidt 1990), with daily torpor bouts of 2–15 hours during which the body temperature can drop to 10°C. In

contrast with torpor, hibernation can last 2–15 days with body temperature dropping to 2°C (Speakman and Thomas 2003).

Southeastern myotis and Rafinesque's big-eared bats show strong fidelity towards sites, but switch roosts frequently (Lewis 1995; Trousdale et al. 2008). Summer and winter roosts are usually in different locations, but summer and winter ranges may remain the same (Rice 1957; Barbour and Davis 1969; Lacki and Bayless 2013). Occasionally a summer roost is also used during the winter (Ludlow and Gore 2000). Data on seasonal movements are scarce across the range and there is no record of either species migrating in Arkansas. In general, there is a paucity of knowledge on both of these species with regards to roost site selection, movement patterns, site fidelity, and implications of seasonal flooding during fall and winter seasons.

Both are regularly associated with permanent slow-moving rivers and creeks or ponds and lakes that are bordered by mature bottomland forests (Jones and Manning 1989; Clark 1990; Gooding and Langford 2004; Rice 2009). In these areas, they are commonly observed roosting in cavities within water tupelos and bald cypresses among others. Other potential roosts include cisterns, wells, bridges, culverts, and old buildings (Jones 1977; Cochran 1999; Trousdale and Beckett 2005). Both of these bat species have a preference for large trees with observed diameters at breast height (DBH) >70 cm (Cochran 1999; Gooding and Langford 2004; Trousdale and Beckett 2005; Clement 2011). They both benefit from using live hollow trees as maternity roosts because they provide an insulating effect which creates stable internal temperatures that are cooler than ambient temperatures and nearby unoccupied hollow trees (Clark 1990; Lacki and Bayless 2013). A downside to tree roosts is that they constantly change until they either

rot or fall, which requires occasional relocation among fewer suitable roost trees available on the landscape (Kunz 1982).

Southeastern myotis mainly roost in water tupelo, but will also use bald cypress (Stevenson 2008), water hickory (*Carya aquatic*, Hoffman 1999), black gum (*Nyssa sylvatica*, Mirowsky and Horner 1997; Schratz 2016), sweetgum (*Liquidambar styraciflua*, Stevenson 2008; Schratz 2016), and red maple (*Acer rubrum*, Carver and Ashley 2008; Schratz 2016). Roost trees have DBH of 50–155 cm (Stevenson 2008). Hollow trees are used by this species year-round, but there is a scarcity of information describing winter roost tree characteristics. Rafinesque's big-eared bats choose trees similar to those chosen by Southeastern myotis. Their roost trees are usually 11–26 m tall with DBH of 79–155 cm (Cochran 1999; Gooding and Langford 2004; Carver and Ashley 2008). They use summer roost trees that are often mature hollow water tupelo and bald cypress (Cochran 1999; Stevenson 2008; Schratz 2016). However, they are occasionally found in other species such as black tupelo (*Nyssa sylvatica*, Clark 1990; Stevenson 2008), American beech (*Fagus grandiflora*, Stevenson 2008), American sycamore (*Platanus occidentalis*, Clark 1990; Stevenson 2008), American hornbeam (*Carpinus caroliniana*, Schratz 2016) and sweetgum (*Liquidambar styraciflua*, Stevenson 2008). The cavities of these roost trees typically have basal openings, but occasionally have a mid-section entrance or a broken top (Gooding and Langford 2004; Rice 2009).

Temperatures recorded 1 m inside several Southeastern myotis roost tree cavities ranged from 23–26°C from May through August (Lacki and Bayless 2013). Nursery colonies using caves or artificial structures (such as bridges) are documented at similarly stable but slightly higher temperatures. Cave hibernacula temperatures ranged from 4.4–

10°C in Indiana, Illinois, and Arkansas (Rice 1957). Maternity colonies of Rafinesque's big-eared bats can tolerate a wide range of temperatures, from 9–46°C (Clark 1990; Hurst and Lacki 1999; Roby et al. 2011). Rafinesque's big-eared bats hibernate at temperatures ranging from 0–14°C (Hurst and Lacki 1999; Lacki and Bayless 2013). However, little is known about winter temperature requirements or how humidity affects winter roost selection in southern populations.

From September to March, Southeastern myotis, in southern populations, enter torpor when temperatures fall below 7°C (Jones and Manning 1989; Brown 1997). Similarly, Rafinesque's big-eared bats remain active year round through much of the southern portion of their range (Jones 1977; Sealander and Heidt 1990; Johnson et al 2012b). They use shallow torpor bouts during cold spells and inclement weather to reduce energy use (Jones 1977; Johnson et al 2012b). Rafinesque's big-eared bat colonies change roost locations seasonally. Southern colonies are often smaller and more scattered across the landscape than northern colonies (Lacki and Bayless 2013). Although capable of entering hibernation, Rafinesque's big-eared bats are typically more alert and active than other species during the winter (Barbour and Davis 1969; Jones 1977). By exploiting winter foraging opportunities, this bat has likely reduced the need for migration (Boyles et al. 2006). However, further studies are likely to show a range of winter behaviors, from extensive hibernation where roosts are cooler to near continuous nightly activity and winter foraging where the climate is warmer.

In southeastern Mississippi, Rafinesque's big-eared bats showed a flexible roosting strategy and characteristically switched roosts regularly. In a multi-year study, they were found to switch roosts every 2.1 days, use 2.5 ± 1.2 roosts per tracking period, and

change roosts 2.6 ± 2.0 times per tracking period, with females generally staying in roosts longer than males (Trousdale et al. 2008). The average distance between consecutive roosts was 572.8 ± 640.3 m (Trousdale et al. 2008). Clusters of Rafinesque's big-eared bats exhibit variable group size within and between roosts, which is characteristic of the fission-fusion model of social behavior (Kerth and König 1999; Trousdale 2011; Johnson et al. 2012a). In this model, individual bats are dispersed across many different roosts, but form a social network larger than the bats of any single roost (Johnson et al. 2012a). This results in colony members having complex relationships. Other cavity-roosting species have similar social behavior and roost fidelity (Kalcounis and Brigham 1998; Willis and Brigham 2004), so Southeastern myotis may also conform to these behaviors.

1.5 Study Species

Southeastern myotis and Rafinesque's big-eared bats have been found with evidence of WNS but little is known about the extent to which tree-roosting bat populations are affected by the syndrome (Foley et al. 2011; Bernard et al. 2015). Although related species are strongly affected by the activities at wind farms, Arnett et al. (2008) did not report any fatalities of Southeastern myotis or Rafinesque's big-eared bats. The main threat facing these two species is the reduction of appropriate forest conditions for bats and their prey caused by large-scale habitat degradation (Tuttle 2007; Rice 2009). This causes a loss of snags and cavities in large trees that make suitable roosts, loss of insect diversity, alteration of travel corridors and a change in bat foraging and movement patterns (Duchamp et al. 2007; Tuttle 2007).

1.5.1 Southeastern myotis (*Myotis austroriparius*)

The Southeastern myotis is characterized by its slender, pointed tragus, un-keeled calcar, hairs on toes that extend past the tips of the claws and thick, woolly fur (Barbour and Davis 1969; Jones and Manning 1989; Sealander and Heidt 1990). There are 3 distinct color phases of this species, red, gray/brown, and a mottled mixture of colors (Lacki and Bayless 2013). Other distinguishing features of this bat are the flesh-colored nose and slight sagittal crest (Barbour and Davis 1969; Sealander and Heidt 1990). This species measures 80–100 mm in total length with a forearm length of 31–46 mm and weighs 5–12 g (Barbour and Davis 1969, Sealander and Heidt 1990).

Southeastern myotis range from southeastern North Carolina to central Florida, then across the Gulf Coastal states to eastern Texas and Oklahoma, and northward up the Mississippi River Valley to Arkansas, western Kentucky and southern Illinois and Indiana (Fig. 1.1; Barbour and Davis 1969; Jones and Manning 1989). In many portions of its distribution, data are deficient or scarce (Lacki and Bayless 2013). Caves and mines are utilized as roosting sites when they are available in the northern portion of their range, but hollow trees, mines, and abandoned buildings are used when caves are absent (Jones and Manning 1989; Gooding and Langford 2004; Mirowsky et al. 2004; Klotz 2012).

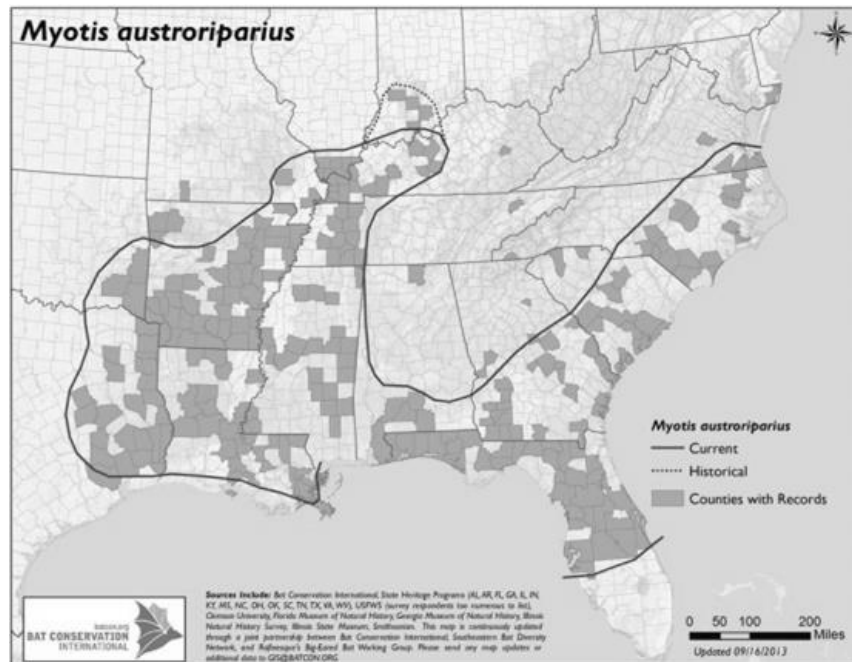


Figure 1.1. The range of Southeastern myotis. Source: Lacki and Bayless (2013).

The echolocation calls of the Southeastern myotis are easily recorded with audio bat detection equipment and are typical of the call structure of other bats in the genus *Myotis*, particularly Northern long-eared bat and the little brown bat. They are fairly easy to distinguish from other species due to the steep frequency modulated sweep with the large bandwidth and short duration (<5ms; Lacki and Bayless 2013). Most of the energy in the call appears at 50–60 kHz, but it can be common for substantial energy to be at higher frequencies so that they appear fragmented (Lacki and Bayless 2013). This species exhibits a large variability in echolocation calls depending on the habitat in which they are flying and the context of the individual producing the call.

The timing of Southeastern myotis breeding remains uncertain, but it is thought that copulation occurs in autumn (Rice 1957; Jones and Manning 1989) and females delay fertilization to give birth to twins in late April or mid-May (Rice 1957; Harvey et al.

2011). Maternity roosts in hollow trees can contain 100–300 individuals (Mirowsky and Horner 1997). The offspring are more altricial than in other bats, likely due to the physiological limitations of producing twins; they experience a high mortality rate during the first week of life (Foster et al. 1978). In the evening while the females forage, the young remain in the maternity roost until they become volant at 5–6 weeks of age, which is typically early June to July (Rice 1957; Jones and Manning 1989). The maternity roosts are abandoned between July and November for winter roosting sites (Rice 1957). Females begin to return to the maternity roosts by the second week of March (Rice 1957).

Southeastern myotis use a strategy called aerial hawking to feed on insects over bodies of water in open air (Harvey et al. 2011). They have been observed foraging once a night for just the first 3 hours after sunset or twice a night at dusk and then again between 0100 and 0300. This variability likely depends on the temperature and season since insect availability can vary heavily. Often known to forage over water, they consume a variety of prey including mosquitoes (Diptera), beetles (Coleoptera), and moths (Lepidoptera) (Rice 1957; Zinn and Humphrey, 1981; Harvey 1992). Their diet can consist of mainly mosquitoes and crane flies on cool spring nights when Diptera are abundant. This is important to help control the population of mosquitoes which can be vectors for diseases (Lacki and Bayless 2013).

1.5.2 Rafinesque's Big-Eared Bat (*Corynorhinus rafinesquii*)

The Rafinesque's big-eared bat is a small forest-dwelling bat characterized by large ears with narrow tips, a broad, long tragus, and 2 prominent lumps on the face that are

enlarged pararrhinal glands (Barbour and Davis 1969; Sealander and Heidt 1990). Other identifying features are the ventral fur that is dark black at the base and pale or white at the tips (Barbour and Davis 1969; Jones 1977), toe hairs that extend beyond tips of toes and a first upper incisor that is bicuspid (Barbour and Davis 1969; Sealander and Heidt 1990). The species measures 92–106 mm in total body length with a forearm length of 40–66 mm, and weighs 7–13 g (Sealander and Heidt 1990; Lacki and Bayless 2013).

Rafinesque's big-eared bats are found from eastern Texas to southern Missouri, north to southern Ohio, Indiana and Illinois, and east to Florida, North Carolina and West Virginia (Fig. 1.2; Barbour and Davis 1969; Jones 1977; Sealander and Heidt 1990). Although the species is widespread throughout the southeastern United States, it has never been considered common and its populations are scattered, leaving it possibly susceptible to natural threats such as hurricanes (Lacki and Bayless 2013).

Similar to the Southeastern myotis, roost choice by Rafinesque's big-eared bats varies throughout their range with individuals using caves and mines where they are available and utilizing hollow trees and manmade structures where caves are lacking in the southern portion of their range (Jones 1977; Gooding and Langford 2004; Mirowsky et al. 2004; Klotz 2012). Colonies of this bat frequently move among trees in close proximity to one another, but show high site fidelity to the same group of trees (Gooding and Langford 2004; Trousdale and Beckett 2005; Trousdale et al. 2008). They also show strong fidelity to other roost sites such as abandoned buildings and bridges (Clark 1990; Trousdale et al. 2008).

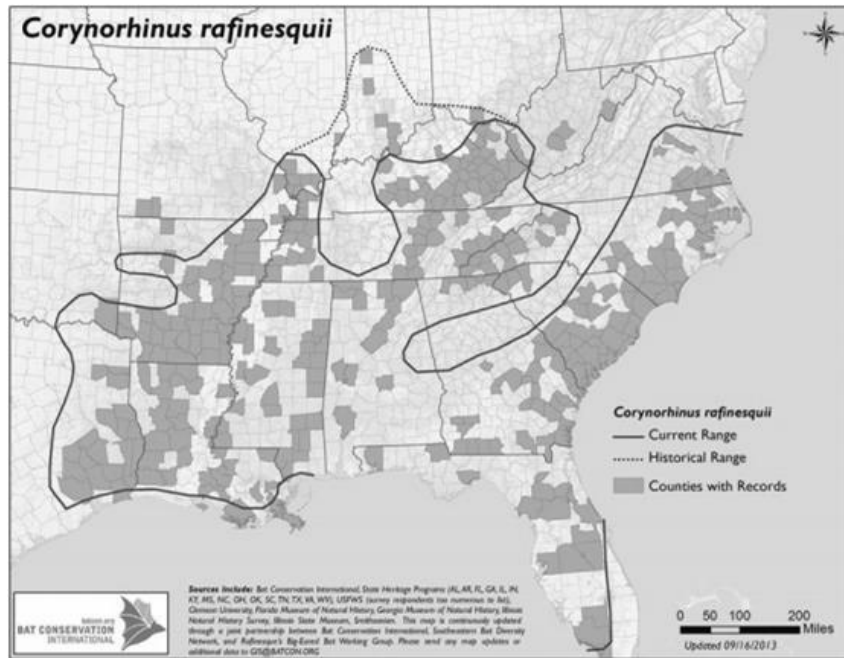


Figure 1.2. The range of Rafinesque’s big-eared bat. Source: Lacki and Bayless (2013).

Rafinesque’s big-eared bats and other bats within the genus *Corynorhinus* are known as ‘whispering bats’ because their echolocation calls have a relatively low amplitude and are therefore difficult to record on audio bat detection equipment (Clement and Castleberry 2011; Lacki and Bayless 2013). The structure of their call often varies but has a fundamental frequency around 40 kHz and can be described as a frequency modulated sweep (Lacki and Bayless 2013). The duration of the calls are less than 10 ms and include a second harmonic that distributes energy to 60 kHz (Lacki and Bayless 2013).

Not much is known about the social structure or behavior of Rafinesque’s big-eared bats. Males are presumably territorial and typically roost separately but in close proximity to nursery groups (Clark 1990). Males are rarely found in maternity colonies but may join female clusters once nursing is complete in mid-August (Barbour and Davis

1969; Hurst and Lacki 1999). Rafinesque's big-eared bats mate in the winter and fall, but ovulation and fertilization are delayed until the spring (Barbour and Davis 1969; Jones 1977). Pregnant females give birth to a single pup in late May to early June (Jones 1977; Harvey et al. 2011). Young are closely associated with females for the first 3 weeks after birth, at which time they become volant and acquire their permanent dentition (Jones 1977). Maternity colonies can consist of as few as 4 individuals or as many as 150 individuals (Jones 1977; Hurst and Lacki 1999; Martin et al. 2011). Maternity colonies disperse in the early fall and reform in spring between early April and late May (Clark 1990; Lacki and Bayless 2013).

Rafinesque's big-eared bats are versatile and agile fliers that glean insects from foliage (Barbour and Davis 1969; Johnson and Lacki 2013). They emerge after sunset and forage until midnight and then re-emerge around 0530 to continue foraging until sunrise (Lacki and Bayless 2013). The primary diet includes members of the insect order Lepidoptera, with over 80% of their prey being moths (Johnson and Lacki 2013). They eat moths from 6 families and at least 22 different species (Lacki and Ladeur 2001; Lacki and Dodd 2011). A large portion of the moths they consume feed on trees in their larval stage, but some attack agricultural crops (Hurst and Lacki 1999; Lacki and Ladeur 2001). They can also consume horse and deer flies that can transmit diseases.

1.6 Problem Statement

Arkansas bottomlands are home to two species of rare bats, Rafinesque's big-eared bats and Southeastern myotis listed as Species of Greatest Conservation Need (AGFC

2016). However, little is known about the general roosting ecology of these bats and even less is known about their behavior during fall and winter. The bats in Cache River National Wildlife Refuge occupy one of a few remaining tracts of bottomland hardwoods that have not been altered significantly by drainage or channelization (USFWS 2016). However, the volume of water that flows through Cache River and Bayou de View might be affected by levees and water discharge from fields. Understanding bats' roosting requirements is therefore crucial to ensure and manage suitable habitats during all seasons including winter when bats may be trapped by rising water. The goal of this project was to increase my understanding of the winter habitat use and requirements of these bottomland forest bat species. To characterize the activity of these bats and to locate their roost trees during the fall and winter, I used mist-netting, acoustics, and radio-tracking techniques.

1.7 Specific Objectives

This project aimed to address the research needs to study fall and winter roosting ecology of both bat species as delineated in the Arkansas Wildlife Action Plan (Fowler 2015), identified throughout their respective range (Lacki and Bayless 2013), and listed under mammals as Priority #2 in the 2015 Arkansas State Wildlife Grant Request For Proposal. My specific aims were as follows:

Objective 1- Document fall and winter activity (e.g., emergence patterns, colony size) of Southeastern myotis and Rafinesque's big-eared bat.

I hypothesized that fall and winter activity is temperature-dependent and similar between species. Southeastern myotis and Rafinesque's big-eared bats regularly associate with the same habitat types in the summer which have permanent slow-moving water sources and are bordered by mature bottomland forests (Clark 1990; Gooding and Langford 2004; Rice 2009). These species have been recorded many times to use the same summer roosts including hollow trees and bridges (Bennett et al. 2008; Stevenson 2008; Rice 2009). During the winter, clusters of Southeastern myotis and Rafinesque's big-eared bats have been observed roosting together, sometimes in a torpid state, in large hollow trees, and in water wells (Jones 1977; Stevenson 2008; Sasse et al. 2011). Therefore, I predicted that both species would share some of the same roosts in the fall and winter.

A winter study found that Rafinesque's big-eared bats regularly switched roosts during the winter and found no difference in the amount of roost-switching between sexes (Rice 1957). From November to January in southeastern Arkansas, water wells were occupied by a nearly balanced sex ratio of Rafinesque's big-eared bats (Sasse et al. 2011). During the summer, males and females of each species have different temperature requirements, particularly during pregnancy and rearing young. However, in the fall and winter, their temperature requirements are similar and they likely stay in close proximity to each other for mating during at least fall months. Accordingly, I predicted that colonies would be composed of male and female bats.

Many temperate bats gather at sites to participate in swarming (breeding behavior associated with large numbers of bats at cave entrances) between August and October (Lowe 2012; van Schaik et al. 2015). The Rafinesque's big-eared bat is closely related to the Townsend's big-eared bat which swarms between September and October at cave and mine sites (Ingersoll et al. 2010). The reproductive condition of males of many *Myotis* species peak between late August and early September, which is also when copulations have been observed (Lowe 2012). Male Rafinesque's big-eared bats join female clusters once nursing is complete in mid-August (Barbour and Davis 1969; Hurst and Lacki 1999). Therefore, I predicted that swarming activity would occur through October.

Bats in torpor reduce their metabolic rate and body temperature but periodically must awaken to perform activities to maintain homeostatic balance (drink, urinate, relocate, etc.) (Foley et al. 2011). Rafinesque's big-eared bats and Southeastern myotis are shallow hibernators, i.e., arouse regularly to forage during the winter and frequently exhibit roost-switching behaviors (Jones 1977; Jones and Manning 1989; Sealander and Heidt 1990). Furthermore, Brown (1997) reports torpor for Southeastern myotis at 7°C but they have been recorded exiting a mine at -2.2°C in Southwest Arkansas (Reed 2004). Although 0°C is a likely threshold for bat activity, activity may also increase with each degree of temperature increase, especially since insect activity increases as temperature increases. I predicted that winter flight activity would be reduced but not eliminated as temperatures decreased.

Objective 2- Characterize roost trees (e.g., cavity size, orientation) used by Southeastern myotis and Rafinesque's big-eared bats during fall and winter compared to randomly selected potential roost trees.

I hypothesized that roost trees used by both bat species had properties (e.g., size, microclimate, and surrounding basal area) that unused trees did not. Both bat species have summer preferences for large trees with observed DBH >40 cm (Cochran 1999; Gooding and Langford 2004; Trousdale and Beckett 2005; Clement 2011). Large trees with large internal chambers can likely provide a more stable environment and host larger groups of bats than smaller trees. In a summer survey conducted on the same study site in 2014 and 2015, the smallest roost tree used by Southeastern myotis and Rafinesque's big-eared bats had a DBH of 40.89 cm (Schratz 2016). Therefore, I predicted that winter roosts would have higher DBH and larger cavities than surrounding unused cavity trees.

Several studies have reported both bat species use some of the same roost sites year round and show strong site fidelity (Stevenson 2008; Rice 2009). However, preliminary observations at my site did not result in any of the summer roost trees previously identified in 2014 and 2015 being occupied, likely due to the extensive seasonal flooding that inundates tree cavities making them unavailable to bats. Also, preliminary observations relied largely on visual inspection of trees so it is possible that bats used summer trees on days other than the day of inspection. I predicted that some of the winter roost trees would be the same trees as those used in the summer.

Objective 3- Characterize habitat (e.g., basal area) surrounding confirmed roost trees compared to randomly selected locations.

I hypothesized that Cache River National Wildlife Refuge provides enough suitable roost trees to sustain populations of Rafinesque's big-eared bats and Southeastern myotis. Rafinesque's big-eared bats and Southeastern myotis frequently exhibit roost-switching behaviors (Barbour and Davis 1969; Trousdale et al. 2008; Johnson et al. 2012a). If the

Cache River National Wildlife Refuge provides enough suitable roosting trees, they should exhibit this characteristic behavior. Therefore, I predicted individual bats would use multiple roost trees.

If there are enough suitable roost trees available within Cache River National Wildlife Refuge then neither species will be forced to travel away from this important foraging area to use a manmade roost on the perimeter. Both species of bats would instead continue their roost-switching behaviors by switching only between roost trees within the interior of the refuge. Accordingly, I predicted that bats would not use bridges and other manmade structures in areas <8 km from the study site.

Objective 4- Characterize the microclimate (i.e., temperature and humidity) of confirmed fall and winter roost trees and potential roost trees.

Tree roosts provide a buffering effect that helps bats to thermoregulate without using an excessive amount of energy (Coombs et al. 2010). Bats will choose roost trees with insulative properties that provide a stable microclimate. Roost trees with the best insulative abilities will likely be trees with fewer and smaller cavity entrances, which would reduce air flow through the tree, or trees that have standing water on the inside of the cavity, which would retain heat better than a tree without water. Rice (1957) found that Southeastern myotis cave hibernacula temperatures ranged from 14–17°C from November to March, in agreement with my preliminary results. Therefore, similar temperature ranges are expected from winter tree roosts. I predicted that temperatures inside winter roost tree cavities would be higher and more stable than ambient.

Objective 5- Determine if and how seasonal flooding and freezing affects

Southeastern myotis and Rafinesque's big-eared bats.

I hypothesized that predictable seasonal flooding would not affect the winter activity of bats. Animals living in bottomland systems have behavioral strategies to avoid the hazards of flooding. Eastern wild turkeys (*Meleagris gallopavo silvestris*) choose nest sites at higher elevations to improve nesting success and coveys of Northern bobwhite quail (*Colinus virginianus*) move their range to escape rising flood waters (Applegate et al. 2002; Byrne and Chamberlain 2013). I expected that bottomland bats would also have a strategy to avoid being trapped within cavity trees when they are seasonally flooded. This strategy likely includes avoiding trees with small, low basal openings since they would be the first to be submerged by flood waters. They will then favor trees with tall basal openings or higher openings such as chimney, upper or window type openings (See chapter 2). Accordingly, I predicted that bats in the study area would select winter tree roosts with cavity openings that were high enough to remain open during flood events.

Each objective resulted in unique data that will allow State and Federal land managers to protect and conserve roost sites by tree preservation, cavity creation, and manipulation of hydrology. Additionally, determination of favorable roost trees at which winter counts can be conducted will improve estimate of population size.

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CHAPTER 2: METHODS

2.1 Study Site

I conducted my study in the Cache River National Wildlife Refuge (CRNWR), which includes 54 individual land tracts in four counties (Jackson, Woodruff, Prairie, and Monroe) in eastern central Arkansas (Fig. 2.1). CRNWR was established in 1986 to provide quality habitat for migratory ducks and to restore and manage large tracts of bottomland hardwood forest (USFWS 2016a). The CRNWR consists of bottomland forest (19,400 ha), croplands (1,400 ha), and reforested areas (6,200 ha; USFWS 2016a). The CRNWR is almost completely surrounded by agricultural fields. Some exceptions include state wildlife management areas (WMA) like Dagmar WMA, Black Swamp North and Black Swamp South WMAs, and some lands owned by the Arkansas Natural Heritage Commission. CRNWR is regarded as one of the few remaining bottomland hardwood areas in the Lower Mississippi River Valley not significantly altered by channelization and drainage (USFWS 2016b). In 1989 it was designated as a site for inclusion in the 'List of Wetlands of International Importance' by the Ramsar Convention. The Ramsar list describes CRNWR as representative of the Lower Mississippi Alluvial Valley Ecosystem and part of the remaining 20% of wetlands in the area. CRNWR supports a rich biodiversity including over 250 bird species, 60 reptile and

amphibian species, 60 mammal species, 40 mussel species, 70 fish species, 70 tree species, and 50 shrub species (Ramsar Sites Information Service 2014).

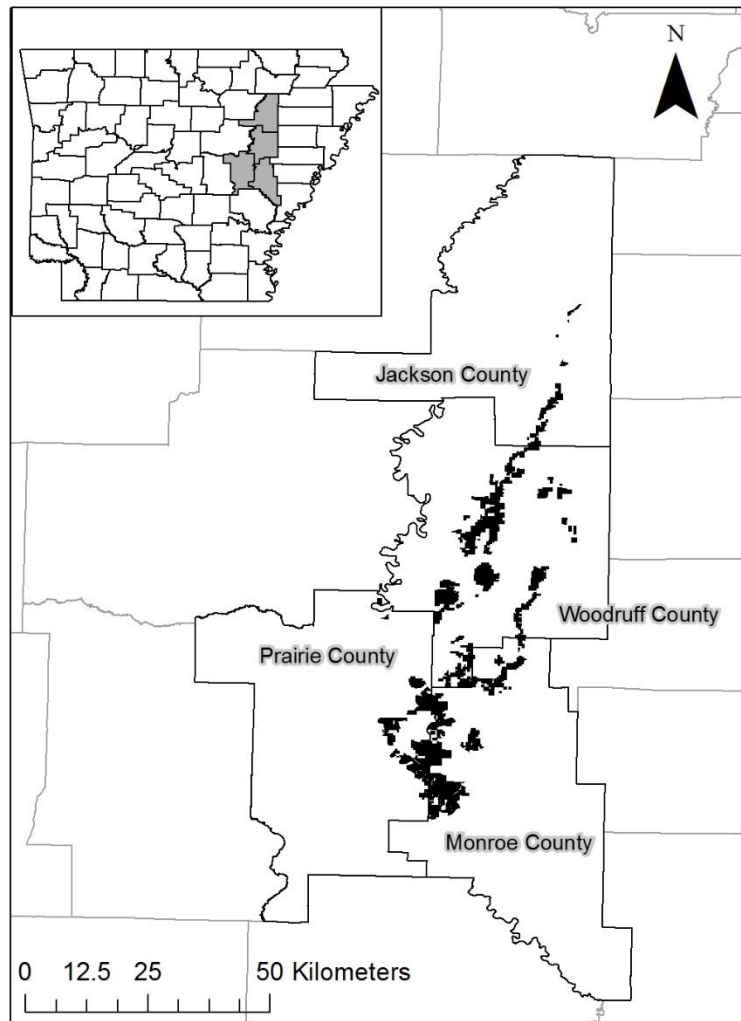


Figure 2.1. Cache River National Wildlife Refuge (black tracts), Arkansas. Inset map: State of Arkansas with Jackson, Woodruff, Prairie, and Monroe counties highlighted.

The main tract of land used for the study was the McNeil farm property, located along Bayou de View in Woodruff County. In 2014, Woodruff County planted 23,750 ha of rice, a crop that requires controlled flooding for weed control (USDA 2014). Many rice

fields surround the McNeil farm property or are located upriver along Bayou de View. When rice fields are drained in August and September, much of that water flows into drainage ditches and ultimately into Bayou de View. Even though Bayou de View is unmanaged and free-flowing, a portion of its volume is inadvertently affected by this human activity. Where Bayou de View comes close to private property, there is a series of levees that prevent the current from spreading out into farms and private lands as it normally would. These levees do not prevent the normal flow of Bayou de View, but collectively they cause the water levels to be higher due to the restricted space through which to pass.

The McNeil farm property is managed by the U.S. Fish and Wildlife Service (USFWS) who must balance the need to encourage scientific research and accomplish other management goals such as game hunting or providing waterfowl sanctuaries. Deer hunting with bows by the public was allowed on the property during the entire research season, but did not interfere with research. Deer hunting with modern guns by permit holders was allowed on the property for 9 continuous days each November and significantly restricted property access for research. The property is also a designated waterfowl sanctuary to provide a resting area for migrating waterfowl that is safe from surrounding duck hunters. USFWS limited my access to the sanctuary during hunting hours on many days in November and December of both seasons.

2.2 Bat Mist-Netting, Radio-tracking and Colony Monitoring

Bats were captured between October and December of 2016 and 2017 using different lengths (6–18 m) of 38-mm mesh nets. Net sizes and orientations were determined on a case-by-case basis depending on the netting site chosen. Netting sites were established within the McNeil farm property along potential corridors with appropriate canopy cover and proximity to water. During a netting session, nets were opened 30 min before sunset and checked for bat captures every 10 min. Nets remained open until 2.5 hours after dark. If temperature dropped below 0°C, the nets were closed.

When a bat was captured in a net, it was carefully extracted and placed in a canvas bag. Each captured bat was identified to species and sexed. The time and temperature at which the bat was caught were also recorded. During processing, bats received a band with a unique identification number on their forearm (right forearm if male, left if female). All bats were considered adults due to the difficulty of examining the ossification of epiphyseal plates at the phalanges during the winter season. Bats had their morphometric data (i.e., forearm length and mass) collected. The presence of parasites was evaluated as well as the level of wing damage. For bats of the target species, Rafinesque's big-eared bats and Southeastern myotis, I affixed a 0.31-g LB-2X model radio-transmitter (Holohil Systems Ltd., Ontario, Canada) between their shoulder blades using surgical glue. I made sure that the receiver detected the transmitter's frequency before affixing it to the bat. The weight of the transmitter did not exceed 5% of the bat's body mass (Sikes et al. 2016). Then, I placed the bat in a bag for 3 min to ensure that the glue would dry completely and allow the bat to recover from handling stress before it was

released. Thirty-eight radio-transmitters were deployed over the two winter seasons (21 Rafinesque's big-eared bats and 17 Southeastern myotis). These transmitters were deployed in a staggered manner (~2-3 per week) throughout each season.

Tracking started the following day using 3-element and 5-element Yagi antennas (Wildlife Materials Inc., Murphysboro, IL) and TRX-1000s receiver (Wildlife Materials, Inc., Murphysboro, IL). Every day that I had access to the property, I tracked the transmittered bats to their roosting trees during the fall-winter season. In addition, roost trees previously identified during summers 2014 and 2015 (Schratz 2016) were explored because the summer roost trees might also be used for roosting during fall and winter seasons (Stevenson 2008; Rice 2009).

Once a bat was tracked to a tree, the tree was considered a confirmed roost tree. At each confirmed roost trees, I attempted to assess colony size and composition by doing emergence counts, harp-trapping, or direct counts within the cavity during the day. During emergence counts, bat behavior was observed around the tree, specifically looking for intense flight activity in circles to determine if fall swarming continues through October for these species. Swarming sex ratios tend to be male-biased, so sex ratios were noted when harp-trapping as another way to identify this activity (Cope and Humphrey 1977; van Schaik et al. 2015).

2.3 Acoustic Bat Activity

The extensive levee system around the McNeil farm property was utilized for Anabat surveys throughout the seasons. Three sites were chosen on the levee along presumed

flyways to understand nightly bat activity in the area at different temperatures and times (Fig. 2.2). Each time an Anabat was deployed on the levee, it was placed in a customized ammunition box resting on a PVC post stuck in the ground. Anabats were deployed on the levee sites as often as possible from mid-November to the first week of January each season when they were not being used to ensure random trees were vacant. Any Southeastern myotis or Rafinesque's big-eared bat calls identified were later matched with temperature and humidity data from a nearby iButton (Fig. 2.2). In 2015, the iButton was attached to the ammo box that encased the Anabat. In 2016 and 2017, data from a nearby iButton were set to collect ambient air temperature and humidity values hourly (Fig. 2.2). In both 2015 and 2016, Anabats with AA batteries failed to record under freezing temperatures. So, in 2017 12-V batteries were used exclusively.

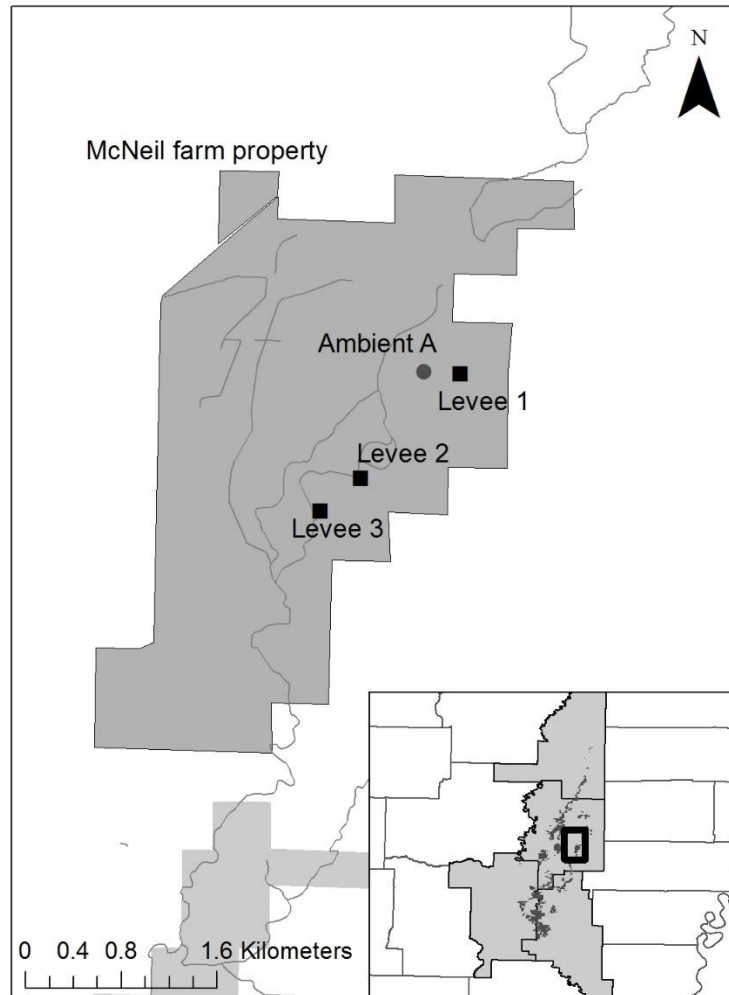


Figure 2.2. Locations of Anabats deployed on levee sites within McNeil farm property and Ambient A iButton used to collect temperature data. Inset map: Cache River National Wildlife Refuge across Jackson, Woodruff, Prairie, and Monroe counties, Arkansas.

2.4 Roost Tree Characteristics

To characterize the roost tree, data were collected at 3 scales. At the level of the roost tree plot, a modified BBird protocol (Martin et al. 1997) was used to record diameter at breast height (DBH), species, and decay status (Table 2.1) of all trees within a 5-m and

11.3-m radius of each confirmed roost tree. Basal area of each roost tree was also measured.

At the roost tree level, tree height and DBH were measured. I also recorded for each roost tree, its species, its decay status (Table 2.1), and its cavity type using the position of its cavity opening(s) (Fig. 2.3). A tree cavity needed to be within the base or buttress of the tree to be considered basal (Fig. 2.3), but did not have to be on the ground. The original 7 cavity types were later combined into 3 general categories (Basal, Upper, or Chimney; Fig. 2.3) for data analysis. Cavity type II, III, and V were not always obvious from the ground. In 2016, I used an aluminum extension pole with a pulley system and a GoPro camera at the end. The idea was to drop the GoPro down into the upper cavity and record as much of the cavity as possible to determine if cavities were connected to each other, how deep they were, and if there were bats inside. This method was only successful twice for determining the cavity type of particularly straight trees that had large upper openings with few limbs in the way. Thus, the method was abandoned in 2017 for a PCE-VE 350N Inspection camera (PCE instruments, Hochsauerland, Germany) with a two-way articulating camera head. This method was much more convenient and slightly more successful, but required drilling a 1.9-cm hole in the trunk of the tree, which did not work on trees with curved trunks.

Table 2.1. Tree Decay-Scale classification system, modified from Pyle and Brown (1998).

Stage	Description
1	Living Tree - twigs have living buds at the tips; canopy fully intact; trunk is intact and has a fresh color (Not stained by weathering)
2	Living Tree with dead or missing sections - most twigs have living buds at the tips; canopy either fully intact or with some branches missing/dead; trunk has sections that are fresh color and/or exposed wood sections that are bleached, weathered and relatively smooth
3	Recently dead tree - Most of the tree shows no signs of major decay; twigs have dead buds or no buds; most branches and twigs present; trunk has sections that are fresh color and/or exposed wood sections that are bleached, weathered and relatively smooth
4	Dead with Minor Decay - Some signs of decay; no twigs remaining; <50% of larger branches missing; exposed wood is bleached, weathered and relatively smooth
5	Dead with Major Decay - Serious signs of decay; >50% of major branches missing; top broken; exposed wood is bleached, weathered and relatively smooth

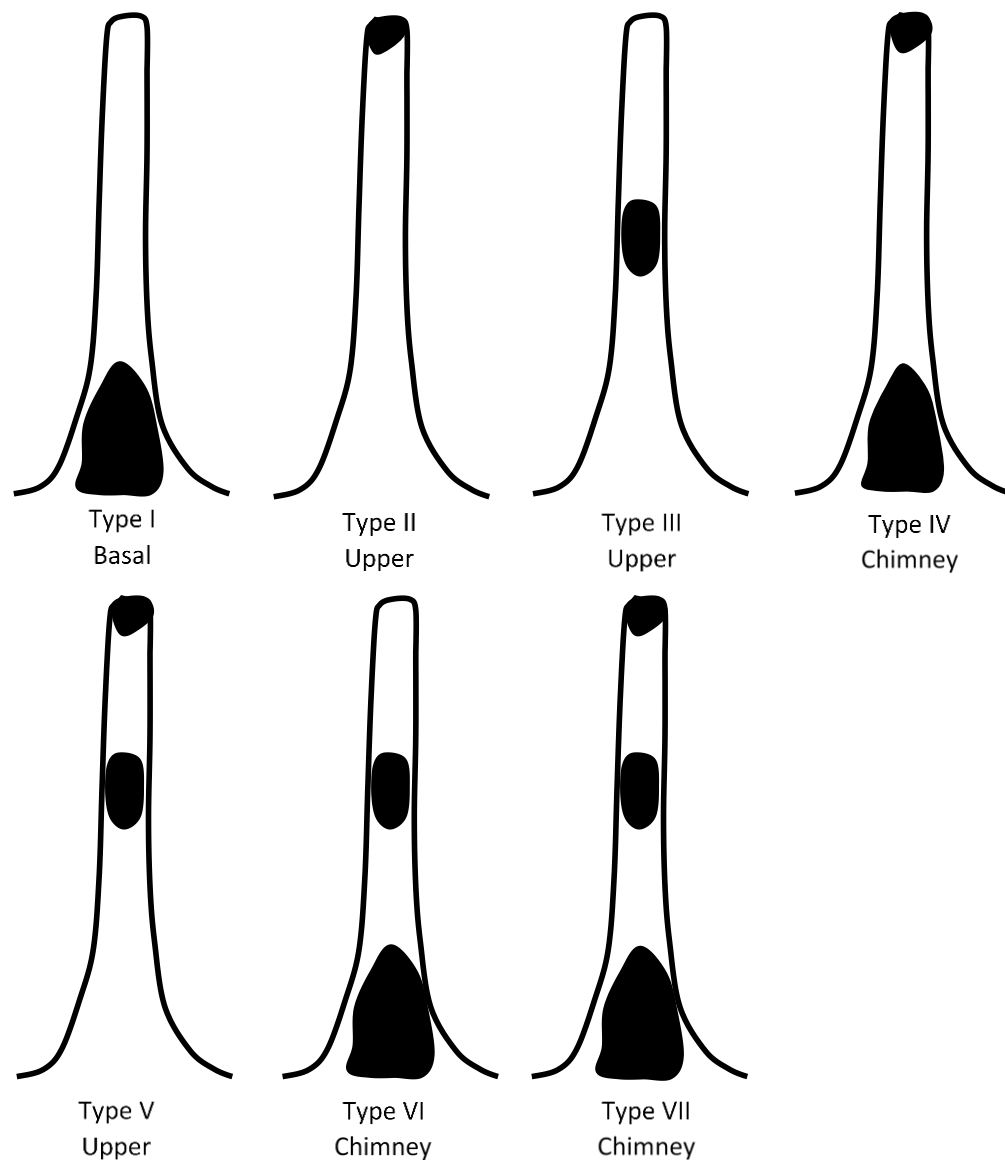


Figure 2.3. Tree cavity types. Top label represents initial numbered category and bottom label represents the combined category terms used for data analysis.

Finally, cavity-specific data collected with a Bosch GLM-20 compact laser distance measurer (Bosch, Farmington Hills, MI) and a carpenter's folding ruler were the width and height of the main opening, height from ground of the lowest point of the cavity opening, and the interior cavity width and height. The cavity opening width and height

were measured as the two largest perpendicular diameters of the cavity opening, and cavity opening area was calculated as an ellipse (Paclik and Weidinger 2007). Cavity opening orientation was recorded with a compass. If the tree was located on a slope, then the height from ground to lowest point of cavity opening was measured from the lowest point of ground at the tree base even if it was not directly beneath the cavity. If a roost tree had multiple openings, then measurements were taken for the highest and the largest openings. Any additional cavity openings that were lower or smaller were not measured because they were likely less used by bats than the biggest two openings.

For each confirmed roost tree, a random cavity tree was selected in the area for roost selection analyses and characterized using the same criteria at all three scales. Random cavity trees were selected using a random number generator. The first number, between 0 and 360, indicated the directional orientation to travel from the confirmed roost tree. The second number, between 40 and 100, indicated the distance in meters to travel from the confirmed tree. The closest cavity tree to that point was considered the random cavity tree to be compared to the confirmed roost tree. If an appropriate cavity tree could not be found within 30 m of the random point, then a second random point was selected to avoid bias searches when no cavity tree was nearby. To prove that a random cavity tree was unused, the speaker of an Anabat SD2 acoustic monitoring device (Titley, Columbia, MO) was placed inside the tree cavity for two days to record any potential bat social vocalizations. In 2016, Anabats were turned on at dusk to record calls from emergence until dawn, but this timeframe was recording foraging calls. In 2017, I turned Anabats on from 1300 until an hour after dark to record social chatter within the roost.

2.5 Roost Microclimate Monitoring

To characterize the microclimate within the confirmed roost cavities relative to random cavities, DS1923 Hygrochron iButtons (Maxim Integrated, San Jose, CA) were placed inside confirmed roost trees and their associated random cavities to record temperature and humidity. In addition, each year 3 iButtons were attached to the outside of trees in the area to collect data on ambient air temperature and humidity. Care was taken to ensure that the random locations did not receive direct sunlight, which could bias the temperature recordings. Each iButton was set to collect data every 60 min and set to start and stop at the same time. In 2016, 18 iButtons were deployed inside cavities. These iButtons were glued to the end of a dowel rod which was inserted into a 1.27-cm hole drilled through the tree at 1 m above the top of the basal opening. Unfortunately, 11 units were lost in 2016. Therefore, in 2017, the 28 iButtons that were placed in cavities and the 3 iButtons deployed to record ambient values were first secured into Ds 1990a-f5 iButton holders. The holders were screwed onto the end of a dowel rod which was then inserted into a 2.22-cm hole drilled 1 m above the top of the opening of the cavity.

2.6 Survey of Man-Made Structures

Both Rafinesque's big-eared bats and Southeastern myotis may roost under bridges, in culverts, in cisterns, and in other man-made structures during parts of the year (Jones 1977; Cochran 1999; Trousdale and Beckett 2005). In December 2015, a survey of bridges and culverts was conducted to locate man-made structures suitable for bats near

the study site. Structures within about 8 km of the McNeil farm property were considered close. Six bridge sites were found within this radius (Fig. 2.4) and each structure was surveyed twice a month during the study seasons to determine if they were being used as roost sites during the fall and winter. No other man-made structures suitable as roost sites were found throughout the entire study.

In 2017, one iButton was placed under bridge 680-West and another under bridge 680-East. Each iButton was placed in a Ds 1990a-f5 iButton holder and attached to a dowel rod with fishing line. The iButtons were then tied to hang inside the crevice of an expansion joint with the dowel rod hanging outside the expansion joint. This allowed the iButton to record temperature and humidity data from the crevice where bats roosted.

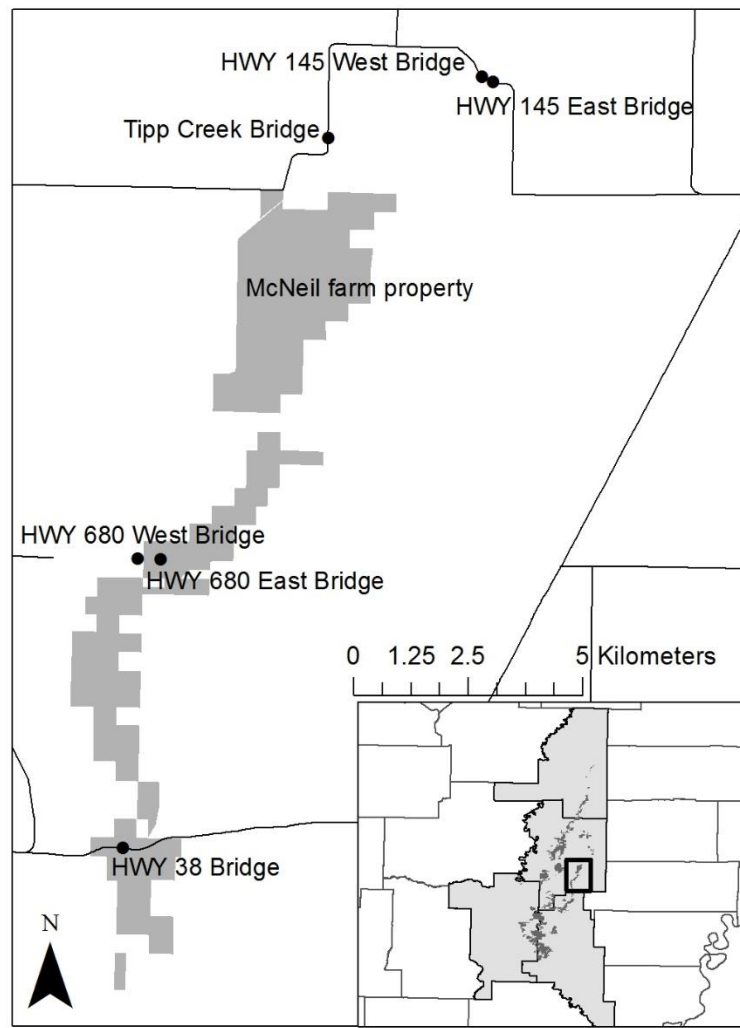


Figure 2.4. Bridges surveyed in winter 2016-17, near the McNeil farm property of Cache River National Wildlife Refuge, Arkansas.

2.7 Water Elevation Monitoring

Jason Phillips of the USFWS provided access to a pressure transducer (HOBO Water Level Data Logger, Bourne, MA) that can record up to 9.1 m of water. This device was

positioned in the main channel of the McNeil farm property (35.10225, -91.17597) from 11 July 2016 to 9 November 2017 (Fig. 2.5) and set to record water depth every hour. The data from this gauge provided information to determine the extent of seasonal flooding in the area.

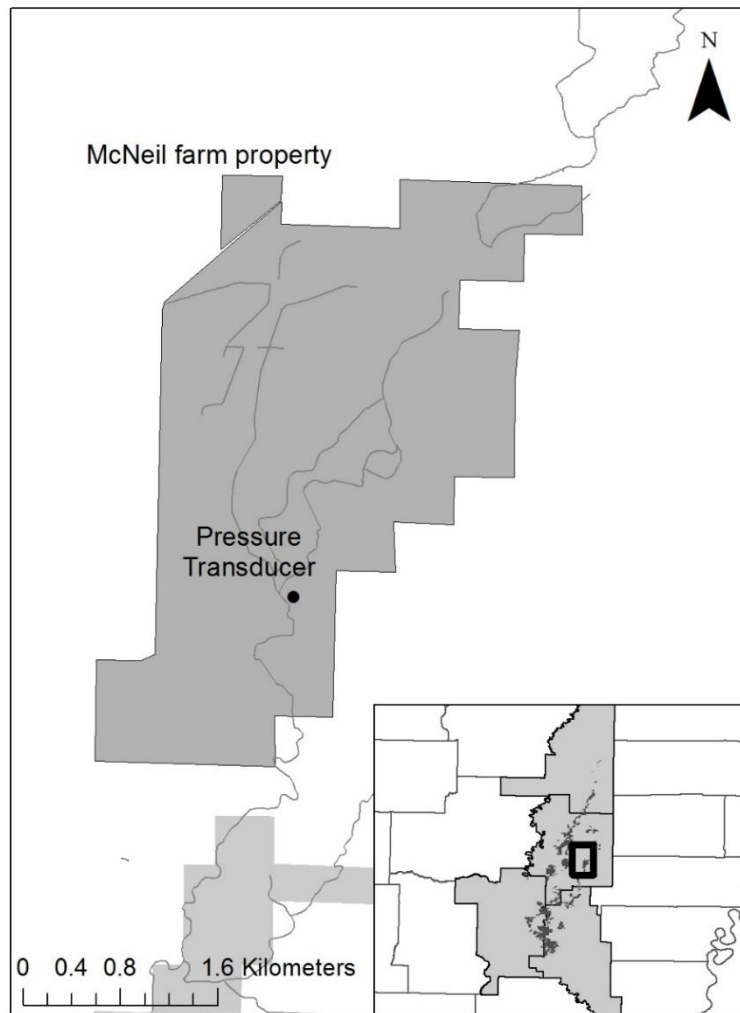


Figure 2.5. Location of pressure transducer used on McNeil farm property along Bayou de View waterway within Cache River National Wildlife Refuge, Arkansas

Since elevation varies throughout the landscape, it is difficult to determine at what stage each tree's basal opening is submerged by rising flood waters. A surveying team used a Sokkia SDL 50 digital level (Sokkia, Olathe, KS) to measure the elevation at the top of the basal cavity of 8 confirmed summer roosts, 8 confirmed winter trees, 5 random cavity trees associated with the winter roosts, and at the pressure transducer in the channel. An additional set of 3 unused trees within the study site which were not associated with confirmed roost trees were randomly selected using a random coordinate generator. These 3 unused trees were at least 30 cm in DBH and with or without a basal cavity. If a confirmed tree or a random tree did not have a basal cavity that could be measured, then the elevation was taken at the base of the tree at the lowest point. This information allows for elevation comparisons between any surveyed tree in the study and the water depth gauge.

2.8 Data Analysis

2.8.1 Bat Roost-Switching Patterns

The first freeze occurred on 20 November in 2016 and on 10 November in 2017. I used these dates as a cutoff to distinguish fall and winter seasons and I categorized a bat's movements as before or after these first freeze dates. The water level increased from 29 November through 5 December 2016 and from 20 December through 23 December 2017. Therefore, when analyzing the effect of flooding on bat activity, any roost switching that occurred between these dates was omitted from the analysis; other roost-switching events were classified as "before" if before the flood initiation date or "after" if after the flood

reached its stable level. Program R (R Core Team 2016) was used to perform all statistical analyses. To characterize fall and winter roosting habits, I analyzed the following parameters: (1) number of consecutive days spent roosting in a tree, (2) distance covered between two consecutive trees during roost switching, (3) number of roost trees used, (4) tree species used for roosting, and (5) cavity type of roost trees (Fig. 2.3).

The number of consecutive days spent roosting in a tree was analyzed for both species. For Southeastern myotis, bat ID ($sd < 0.001$) and tree ID ($sd < 0.001$) were associated with negligible standard deviation and were therefore not included as random effects. Consequently, I used generalized linear models (GLM) with year and sex as fixed effects and a Poisson error distribution. In addition, I used either Julian week to assess changes that occurred gradually throughout the season or “Freeze” to assess changes that occurred before and after the first freeze event of the season. For Rafinesque’s big-eared bats, bat ID ($sd = 0.507$) and tree ID ($sd = 0.419$) were associated with substantial variation. However, including both variables led to overparameterization and model convergence issues, maybe because of some redundancy between them. Therefore, I only included bat ID, because of its higher standard deviation, as a random effect, and I considered the same fixed effects as for Southeastern myotis. The generalized linear mixed-effect models (GLMM) for Rafinesque’s big-eared bats were built with function `glmer` in package `lme4` (Bates et al. 2015) and a Poisson error distribution. I used an information-theoretic approach to select the best model using the Akaike Information Criterion (AIC; Akaike 1973) corrected for small sample size (AICc in package `AICcmodavg`; Mazerolle 2016) for Southeastern myotis and a quasi-AICc (QAICc) for Rafinesque’s big-eared bats

to correct for overdispersion (variance inflation factor: $\hat{c} = 1.14$; Burnham and Anderson 2002).

The number of consecutive days spent roosting in a tree before and after the flooding events was analyzed separately. For Southeastern myotis bat ID ($sd < 0.001$) and tree ID ($sd < 0.001$) were associated with negligible variation so GLM models were used with a Poisson error distribution. Year, sex and Flood were fixed effects. No analysis was done on Rafinesque's big-eared bat consecutive days in a tree before and after the flooding event because there were too few data points after the flood occurred.

The distance covered between two consecutive trees was analyzed for both species. Distance traveled by bats between trees was calculated in R with function `dism` in package `geosphere` (Hijmans et al. 2017). For Southeastern myotis, bat ID ($sd = 0.663$) and tree ID ($sd = 0.878$) were associated with substantial variation, but only tree ID was included as a random effect, because of its higher standard deviation. For Rafinesque's big-eared bats, bat ID ($sd = 0.171$) was associated with substantial variation unlike tree ID ($sd < 0.001$). Therefore, only bat ID was included as a random effect. Fixed effects were year, sex, and either Julian week or Freeze. The GLMM models for both species were built with a Gamma error distribution. Issues with model convergence were avoided using an Adaptive Gauss-Hermite quadrature approximation (function `nAGQ` in package `lme4`; Bates et al. 2015). For both species, I used an information-theoretic approach to select the best model using AICc.

The distance covered between two consecutive roost trees before and after the flooding event was analyzed separately. For Southeastern myotis, bat ID ($sd = 0.68$) and tree ID ($sd = 1.13$) were associated with substantial variation, but only tree ID was included as a

random effect, because of its higher standard deviation. GLMM models with a Gamma error distribution were built with year, sex and Flood as fixed effects. Issues with model convergence were avoided using an Adaptive Gauss-Hermite quadrature approximation. I used an information-theoretic approach to select the best model using AICc. No analysis was done on Rafinesque's big-eared bat consecutive days in a tree before and after the flooding event because there were too few data points after the flood occurred.

The total number of different roost trees used by an individual bat before and after the freeze or the flood were analyzed for both species. Bat ID was not associated with variation ($sd < 0.001$) for either species so this random effect was not included. For both species, I used GLM models with a Poisson error distribution and an offset for the number of days each bat was tracked to account for unequal sampling effort per bat. Fixed effects were year and sex, as well as Freeze for both species or Flood for Southeastern myotis to assess changes that occurred before and after the first freeze or first flood event of the season. Only one Rafinesque's big-eared bat was tracked after the flood in both years, so I could not compare the number of trees used by this species before and after the flood. For both species, I used an information-theoretic approach to select the best model using AICc.

The tree species used by an individual bat were analyzed for both species. Tree species were determined in the field and sorted into categories for analysis. This was done two ways: tree species were organized either by genus or in three categories (water tupelo, bald cypress and 'Other'). I examined if the frequency of tree species used changed over time by conducting chi-square tests for all three timing definitions (before and after the freeze, before and after the flood, or by Julian week) and both tree species definitions.

The cavity types used by an individual bat were analyzed for both species. Cavity type was determined in the field and sorted into one of three categories (i.e., basal, chimney, upper) based on cavity location within the tree (Fig. 2.3). I examined if the frequency of cavity types used changed over time by conducting chi-square tests for all three timing definitions (before and after the freeze, before and after the flood, or by Julian week).

2.8.2 Acoustic Bat Activity

The program Bat Call Identification v2.7c (BCID 2015, Ryan Allen, Kansas City, Missouri) was used to identify calls recorded by the Anabats located on the levee system. All calls identified were then visually vetted. Rafinesque's big-eared bat calls were only detected twice. Therefore, I only included Southeastern myotis calls in this analysis. Because Southeastern myotis are the only myotis species active at this time of year, all myotis calls were considered Southeastern myotis calls. I did not use the number of calls to analyze bat activity or numbers because two or more calls may originate from the same individual. Instead, I used the number of nights during which at least one vetted call was recorded (hereafter called active nights). I classified each active night into one of four temperature ranges ($<0^{\circ}\text{C}$, $0\text{--}5^{\circ}\text{C}$, $5\text{--}10^{\circ}\text{C}$ and $>10^{\circ}\text{C}$) based on the lowest temperature recorded by an iButton between 1600 and 0700 to match times of bat nocturnal activity. I obtained the expected number of active nights by classifying each night that an Anabat was deployed into one of the same four temperature ranges based on the minimum nightly temperature recorded by an iButton. A Fisher's exact test was used to compare

the frequencies of observed active nights and expected active nights in each of the temperature categories.

2.8.3 Roost Tree Characteristics Analysis

Roost tree characteristics were analyzed on three scales: (1) plot (5-m and 11.3-m radius), (2) tree, and (3) cavity. For each scale the roost tree is compared with a random tree selected from the same area and characterized using the same criteria as the roost tree on all three scales. Roost and random trees are assigned a Pair ID and each pair is represented once in this analysis. For each analysis, an information-theoretic approach was used to select the best model using AIC or AICc, depending on sample size.

Plot level.—In 2016, data for trees were recorded as a list of all DBH values and counts of the number of trees in each decay class and tree species within plots. In 2017, DBH, decay class and tree species were recorded per tree within the plots. Due to the differences in how data were collected between seasons, only 2017 plots were analyzed. Some trees had overlapping plots either at the 11.3-m radius or both. For these trees, one of the overlapping plots was randomly excluded to avoid inflating the weight of characteristics appearing in two plots. If three plots overlapped, then the plot that overlapped the most was kept and the other two were excluded.

I considered 3 DBH variables, 3 decay class variables, and 3 tree species variables; all 9 variables were counts of trees within a plot. The DBH variables were counts of trees that were small (DBH < 11.05 cm), medium (DBH 11.1–31.1 cm), and large (DBH > 31.1 cm), based on the first, and third quartiles in the data. Because decay classes 3, 4 and

5 all represented dead trees (Table 2.1), decay class variables were re-defined as 1, 2, and 3+. In the 5-m radius plots, 26 tree species (17 genera) were recorded, but water tupelos (*Nyssa aquatica*) and bald cypresses (*Taxodium distichum*) were the most represented and the most likely to be used by bats (Cochran 1999; Stevenson 2008; Rice 2009). Therefore, the three tree species variables were number of water tupelos, number of bald cypresses, and number of other tree species per plot. Finally, I considered basal area as an additional predictor.

Data from 5-m and 11.3-m plots were analyzed separately for each bat species. All predictors were checked for correlation. Any correlation coefficients > 0.7 were considered strongly correlated and those variables were not included together in models (Mindrila and Balentyne 2013). For both species and both 5-m and 11.3-m radius plots, pair ID ($sd < 0.001$) and tree ID ($sd < 0.001$) were associated with negligible variation so were not used as random effects. GLM models with binomial error distribution were created to test for an effect of the plot characteristics (i.e., DBH, decay class, tree species, basal area) on the probability of a tree to be used as a roost.

Tree level.—For each bat species, the probability a tree was used as a roost was analyzed in GLM models with a binomial error distribution. No random effects were used because pair ID ($sd < 0.001$) and Year ($sd < 0.001$) were associated with negligible variation for both bat species. I used tree species, decay class, DBH, tree height, and year as fixed effects. Any predictors strongly correlated ($r > 0.7$) were not included together in models (Mindrila and Balentyne 2013). The tree species variable was organized as water tupelo, bald cypress, and “other”. First I built models with additive and interaction effects

of Year and each of the other fixed effects to determine if Year influenced the models. Then, the remainder of the fixed effects were examined.

Tree cavity type for each bat species was examined separately because no data were available for random trees with upper cavities. Therefore, the analysis focused on roost trees and random trees with basal and chimney cavities. To compare roost and random tree cavity types, GLM models were created with binomial error distribution. Pair ID and Year were excluded as random effects because they were not associated with variation.

Roost Cavity level.— For each bat species, GLM models with a binomial error distribution were created to test for an effect of cavity opening area, opening height from ground, opening orientation and internal volume on the probability of a cavity to be used as a roost. Strongly correlated predictors ($r > 0.7$) were not put together in models (Mindrila and Balentyne 2013). For both bat species, tree ID ($sd < 0.001$) and Year ($sd < 0.001$) were excluded as random effects because they were associated with negligible variation.

2.8.4 Roost Microclimate Analysis

To characterize microclimates within roosting trees in the fall and winter, I analyzed the following parameters: (1) Temperature inside roost trees before vs. after the flood, (2) Humidity inside roost trees before vs. after the flood, (3) Temperature among cavity types and location types (roost, random or ambient), and (4) Humidity among cavity types and location types (roost, random or ambient). Tree ID was included as a random

effect if it exhibited substantial variation. An information-theoretic approach was used to select the best models using AIC or AICc, depending on sample size.

Internal roost tree temperatures collected with iButtons in 2017-18 were analyzed to determine if temperature stability inside the roost trees was different before and after the flood. For this analysis, I chose eight trees (4 with chimney and 4 with upper cavity types; Fig. 2.3) that had temperatures recorded hourly during the 10 days preceding the start of the flood event and during the 10 days following the end of the flood event. For each roost tree, the coefficient of variation (CV) was calculated per day. The dataset containing the CV data for all roost trees after the flood had 10 improbable outliers (e.g., $CV = 0$) that were removed prior to analysis. I built linear mixed-effect models with the function `lmer` in package `lme4` (Bates et al. 2015). Fixed effects for the models were Flood and Cavity Type. The `emmeans` function in package `emmeans` (Lenth 2018) was used to report mean CV and 95% confidence intervals.

The internal relative humidity of roost trees was analyzed to determine if mean relative humidity inside roost trees was different before and after the flood. The same eight trees were used for this analysis as for the temperature analysis above. Each tree had internal relative humidity values recorded hourly during the 10 days prior to the start of the flood event and during the 10 days after the end of the flood event. I built GLMM models with a Gamma error distribution. The fixed effects were Flood and Cavity Type.

For temperature among cavity types and location types (roost, random or ambient), I analyzed the following three parameters: (1) Coefficient of Variation, (2) Minimum nightly temperature, (3) Mean daily temperatures. The iButtons were placed in 13 roost trees, 14 random trees, and 3 ambient locations each recording temperatures hourly from

5 January through 22 March 2018. The CV was calculated per day for each of the roost trees, random trees, and ambient locations. Similarly to the before and after flood temperature analysis, I had to remove 12 roost tree outliers and 30 random tree outliers (CV = 0).

Field challenges, such as installing an Anabat recorder at an upper cavity opening or in a flooded basal cavity, led to unequal numbers of trees with various cavity types (Table 2.2). Also, only 3 ambient iButtons were deployed and they have no cavity that can be evaluated. Consequently, for all three temperature parameters (temperature stability, mean daily temperature, minimum nightly temperature), I first compared a model with only Location type (Roost, Random or Ambient), a model with only Cavity type (Basal, Chimney or Upper), and a null model, using GLMM models with a Gamma error distribution. Then, I investigated the effect of Location type or Cavity type by pairs on available data (Table 2.2) using individual GLM models with a Gamma error distribution.

Table 2.2. Number of days for which iButton data was calculated in each category of iButton location (Ambient, Random tree, or Roost tree) and opening type, in winters 2016-17 and 2017-18 in the Cache River National Wildlife Refuge, Arkansas.

	Ambient	Basal	Chimney	Upper
Ambient	231	0	0	0
Random	0	674	374	0
Roost	0	0	536	453

To determine if internal relative humidity differed between roost trees, random trees, and the ambient environment, I used hourly relative humidity recorded at the same times as the temperature values used for the previous analysis by the same 30 iButtons.

Similarly to temperature parameters, I first compared the model with only Location type (Roost, Random or Ambient), the model with only Cavity type (Basal, Chimney or Upper), and the null model, using GLMM models with a Gamma error distribution. Then, I examined the effect of Location type or Cavity type on internal relative humidity with available data (Table 2.2) using individual GLM models with a Gamma error distribution.

2.8.5 Man-Made Structures

Survey of man-made structures did not yield substantial data. Therefore, I only report descriptive statistics of bat captures and temporal patterns in temperature and humidity recorded by iButtons in man-made structures.

2.8.6 Water Elevation

Any detailed analysis of the effect of water elevation on bat roost selection could not be completed due to a small sample size of measured trees which bats were known to use via tracking or observations. Most roost trees used in the winter were not trees measured for elevation, therefore the tree survey was only partially informative. Chapter 3 reports the temporal patterns of water levels recorded by the pressure transducer as well as qualitative results regarding tree measurements.

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CHAPTER 3: RESULTS

3.1 Bat Mist-Netting, Radio-tracking and Colony Monitoring

Bats were captured via mist-net from 24 September 2016 through 25 December 2016 and from 30 September 2017 through 21 December 2017 for a total of 29 net-nights at 10 sites. Four species were captured using mist-nets including Rafinesque's big-eared bat (*Corynorhinus rafinesquii*; $n = 53$), Southeastern myotis (*Myotis austroriparius*; $n = 29$), Evening bat (*Nycticeius humeralis*; $n = 19$), and Eastern Red bat (*Lasiurus borealis*; $n = 1$) for a total of 102 individuals. All Southeastern myotis and Rafinesque's big-eared bats caught in mist nets were banded before release.

I placed radio-transmitters on 17 Southeastern myotis (11 males, 6 females) and 21 Rafinesque's big-eared bats (9 males, 12 females); 14 Southeastern myotis and 19 Rafinesque's big-eared bats were re-located at least once, which led to identification of 42 roost trees for Southeastern myotis and 33 roost trees for Rafinesque's big-eared bats. The bats were tracked from 1–26 days depending on life of the transmitter. Southeastern myotis used 1–7 different roost trees and switched roosts every 1.69 ± 0.13 days. Rafinesque's big-eared bats used 1–5 different roost trees and switched roosts every 2.74 ± 0.29 days. There was one instance of a Southeastern myotis and a Rafinesque's big-eared bat being radio-tracked to the same tree, but it was in different years. Three trees previously identified as summer Southeastern myotis roosts by Schratz (2016) were used

by Southeastern myotis during this project. All three were water tupelos (*Nyssa aquatica*) with basal cavities, but none were used after the freeze or the flood in either year. Twelve bats were recaptured during the 2017 season, 1 at a bridge site, 2 with mist nets in flight corridors, and 9 while harp-trapping trees. The recaptures included 1 female Rafinesque's big-eared bat, 1 male Rafinesque's big-eared bat, and 10 male Southeastern myotis. Five of the male Southeastern myotis were previously banded by Schratz (2016). All other bats were previously banded during the 2017 season of this project and recaptured in the same season. Of the recaptured bats, 4 banded during this project and 1 banded by Schratz (2016) were recaptured in the same tree where they were caught initially.

In addition to capturing bats via mist nets, bats were captured via harp traps to assess colony composition within cavities. Harp traps deployed over 6 nights throughout the study resulted in 131 Southeastern myotis and 7 Rafinesque's big-eared bats successfully captured. A water tupelo tree previously identified as a summer roost tree for Southeastern myotis by Schratz (2016) was harp-trapped on 28 October 2017 resulting in the capture of 21 males and 12 females. The other 5 harp-trapping events were all at a water tupelo identified in winter 2015 as a Rafinesque's big-eared bat and Southeastern myotis roost tree (Fig. 3.1A and B). On the nights when harp traps captured Southeastern myotis, a bias toward reproductive males was revealed (Table 3.1).

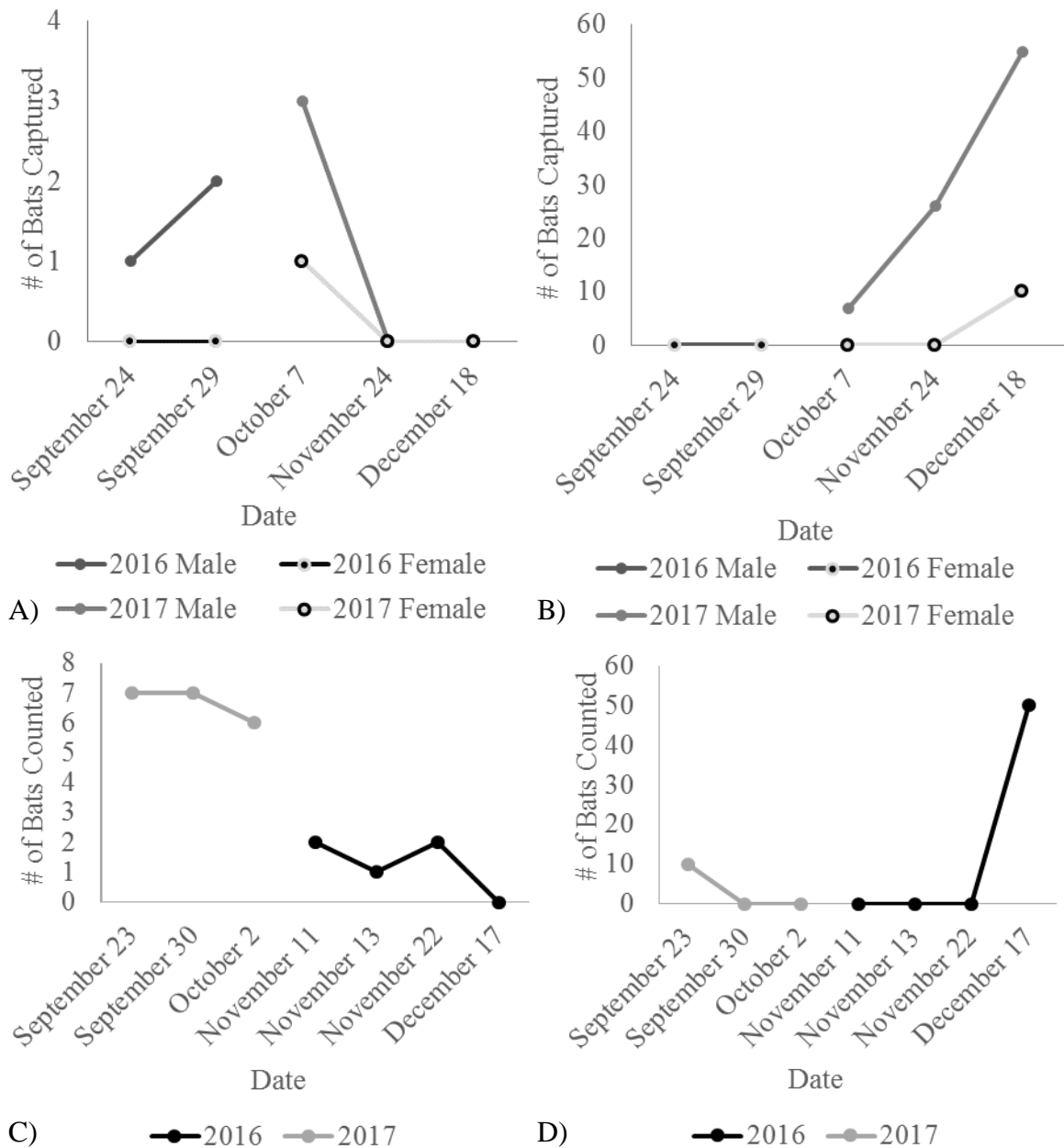


Figure 3.1 – Observations of Rafinesque's big-eared bats and Southeastern myotis at the same roost tree in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. (A) Rafinesque's big-eared bat harp-trap captures. (B) Southeastern myotis harp-trap captures. (C) Rafinesque's big-eared bat direct counts. (D) Southeastern myotis direct counts.

Table 3.1 – Number of female, reproductive male and non-reproductive male Southeastern myotis captured via harp trap in Cache River National Wildlife Refuge, Arkansas in winter 2017.

Tree ID/ Date	Number Females	Number Reproductive Males	Number Non-Reproductive Males	% Total Male
Tree 50 October 7 2017	0	5	2	100
Tree 13 October 28 2017	12	18	3	63.6
Tree 50 November 24 2017	0	26	0	100
Tree 50 December 18 2017	10	55	0	84.6

I also assessed colony size with exit counts from roost cavities. In 2016-17 this occurred on 12 nights with results ranging from 0–127 for Southeastern myotis roosts (Table 3.2). In 2017-18 this occurred on 35 nights with results ranging from 0–83 for Rafinesque’s big-eared bat roosts and 0–78 for Southeastern myotis roosts (Table 3.2). Exit counts might not be a reliable method to assess total colony size in winter since every colony member might not emerge nightly depending on weather. However, exit counts were the only way to assess colony size of bats roosting in upper cavities. I also conducted direct counts of bats within roost trees when the cavity opening allowed access. This occurred 42 times with results ranging from 0 to over 20 Rafinesque’s big-eared bats and from 0 to over 50 Southeastern myotis roosts (Table 3.2). Direct counts were also conducted 7 times at the same water tupelo identified in winter 2015 as a Rafinesque’s big-eared bat and Southeastern myotis roost tree where multiple harp traps were deployed (Fig. 3.1C and D). Counts of large numbers of these species within a tree could not be precise because these species nestle closely together making accurate counts

difficult. While conducting exit counts on Rafinesque's big-eared bat roost trees in 2016 and 2017, bats were observed circling the tree on 6 occasions and bats were observed exiting and reentering the tree on 4 occasions (Fig. 3.2). While conducting exit counts on Southeastern myotis bat roost trees in 2016 and 2017, bats were observed circling the tree on 6 occasions and bats were observed exiting and reentering the tree on 4 occasions (Fig. 3.2).

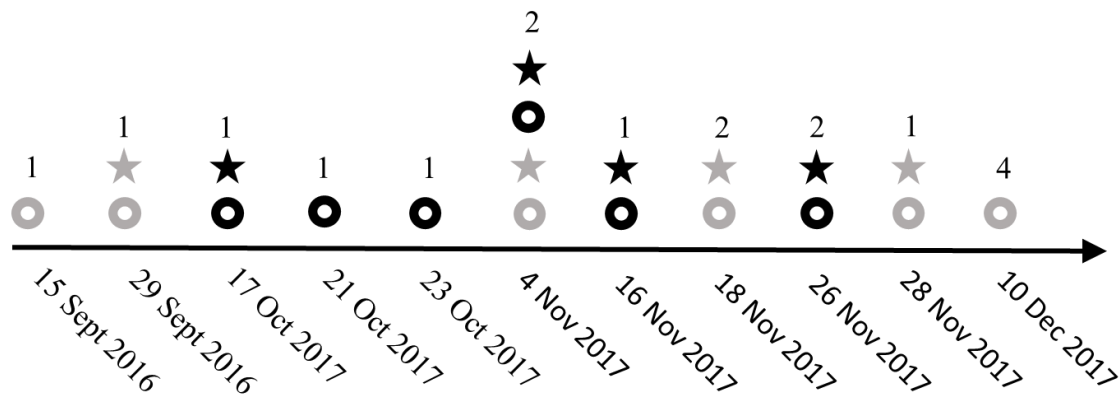


Figure 3.2 – Observations of swarming behavior of Rafinesque's big-eared bats (Black) and Southeastern myotis (Grey) during exit counts of roost trees in winters 2016 and 2017 in Cache River National Wildlife Refuge, Arkansas. Stars and circles indicate reentering and circling behavior observed, respectively. Numbers indicate the number of exit counts that were performed on that date.

Table 3.2 – Rafinesque’s big-eared bats and Southeastern myotis counted while roosting or emerging from roost trees in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. Data are presented as Early/Late in season with counts occurring before November 15 considered early and after November 15 considered late in season, and a – indicates no count was conducted. D = Direct count. E = Exit count. R = Rafinesque’s big-eared bat. S = Southeastern myotis.

Bat Counts			Bat Counts		
Tree ID	2016	2017	Tree ID	2016	2017
78	-/0-20+ ^{DS}		18		20+/- ^{DR}
90		52-55/0 ^{ER}	70		1-2/2 ^{DR}
86		55/27 ^{ER}	105		-/2 ^{DR}
85		36/1 ^{ER}	13	119-127/23 ^{ES}	61/8 ^{ES}
73		16/- ^{ER}	6		78/- ^{ES}
91		2/- ^{ER}	16	0-3/- ^{ES}	
89		18/- ^{ER}	52	0-3/- ^{ES}	
111		-/0-3 ^{ER}	87		1/- ^{ES}
110		-/19 ^{ER}	96		19/- ^{ES}
81		-/0 ^{ER}	94		0/1 ^{ES}
74	-/1 ^{DR}	-/6-83 ^{ER}	98		0-1/3 ^{ES}
59	1/- ^{DR} ; 1/- ^{DS}		95		-/2 ^{ES}
70	1/- ^{DR} ; 2/- ^{DS}		99		2 ^{DS} /36 ^{ES}
61	-/1 ^{DR}		102		-/0-2 ^{ES}
57	-/0 ^{DR}		101		-/0 ^{ES}
58	-/0 ^{DR}		116		-/1 ^{ES}
65		1/- ^{DR}	123		-/50+ ^{DS}
88		2/- ^{DR}			

3.2 Bat Roost-Switching Patterns

In 2016, the first freeze event occurred on 20 November, 2016 and the flooding event took place from 29 November–5 December, 2016. In 2017, the first freeze event occurred on 10 November, 2017 and the flooding event took place from 20–23 December 2017. These dates were used for analysis to determine if environmental events (i.e. first freeze or first flood) triggered changes in behavioral habits.

Southeastern myotis stayed in roost trees 1–9 consecutive days before switching. My model selection yielded three best models: the model with Week had the lowest AICc, the model with the Freeze factor was within 2, but the most parsimonious model was the null (Table 3.3). However, the low AICc values of both the Week (slope = -0.04 ± 0.02) and the Freeze models suggest that the number of consecutive tree days may tend to decrease as the season progresses. Before the first freeze event, individual bats switched roost trees every 2.0 ± 0.28 days ($n = 6$ bats), and after the first freeze event, the number of consecutive days spent in a roost tree reduced slightly to 1.57 ± 0.15 days ($n = 11$ bats). When Flood was analyzed, the model selection for number of consecutive days spent in a tree before switching generated 5 models with low AICc values including the Flood model, but the most parsimonious model was the null (Table 3.4).

Rafinesque's big-eared bats stayed in roost trees 1–14 consecutive days before switching. The GLMM models indicated that all models were at least 2 less than the null except for the Year and Sex models. But the best and most parsimonious model included the Freeze factor (Table 3.3). This model indicates a significant increase in the number of consecutive days each bat roosted in a tree after the first freeze event. Before the first

freeze event, individual bats switched roost trees every 1.75 ± 0.31 days, and after the first freeze event, the number of consecutive days spent in a roost tree increased to 3.99 ± 0.64 days.

Table 3.3 – Model selection for the number of consecutive days in roost trees by Southeastern myotis and Rafinesque’s big-eared bats tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. BAFreeze is a two-period factor: before vs. after the first freezing event of each winter. AICc is the Akaike Information Criterion corrected for small samples, QAICc is the quasiAICc. Differences in AICc ($\Delta AICc$) and QAICc ($\Delta QAICc$) are reported.

Predictor	Southeastern myotis		Rafinesque’s big-eared bats	
	AICc	$\Delta AICc$	QAICc	$\Delta QAICc$
Null	288.03	0.53	338.04	11.85
Week	287.50	0.00	329.41	3.22
Week * Year	289.82	2.32	333.45	7.26
Week + Year	288.84	1.34	331.45	5.26
Year	288.77	1.27	340.10	13.91
Week + Sex	289.30	1.80	331.30	5.11
Sex	289.95	2.45	340.10	13.91
Week + Year + Sex	290.78	3.28	333.44	7.25
Week * Year + Sex	291.72	4.22	335.24	9.05
BAFreeze	288.18	0.68	326.19	0.00
BAFreeze * Year	289.01	1.51	329.66	3.47
BAFreeze + Year	288.73	1.23	328.40	2.21
BAFreeze + Sex	290.16	2.66	328.27	2.08
BAFreeze + Year + Sex	290.70	3.20	330.55	4.36
BAFreeze * Year + Sex	290.79	3.29	331.57	5.38

Table 3.4 – Model selection for the number of consecutive days in roost trees by Southeastern myotis tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. BAFlood is a two-period factor: before vs. after the flooding event of each winter. AICc is the Akaike Information Criterion corrected for small samples. Differences in AICc ($\Delta AICc$) are reported.

Southeastern myotis		
	AICc	$\Delta AICc$
Null	283.03	0.00
Year	283.74	0.71
Sex	284.87	1.84
BAFlood	283.99	0.96
BAFlood * Year	284.91	1.88
BAFlood + Year	285.50	2.47
BAFlood + Sex	286.12	3.09

The distance between consecutive roosts of Southeastern myotis were 7–1,777 m with an average of 498.8 ± 67.7 m. The top 4 models included combinations of the Year and Freeze variables (Table 3.5). The most parsimonious model included only Year, indicating that the distance between consecutive roosts was greater in 2017 (324.7 ± 81.4 m) than in 2016 (137.5 ± 54.0 m). However, the low AICc of the Freeze model suggests that distances may tend to be reduced after the freeze event. Before the first freeze event, the average distance traveled between roost trees was 484.1 ± 194.6 m and afterwards decreased to 221.9 ± 53.7 m. When Flood was analyzed, the model selection for Southeastern myotis distance between roost trees showed that the best model was the Year model, but the most parsimonious was the null (Table 3.6).

The distance between consecutive roosts of Rafinesque's big-eared bats was 6–1,135 m with an average of 264.6 ± 28.7 m. There were 4 other models within 2 AICc points of the null, including Year which, similarly to Southeastern myotis results, suggests a slightly greater distance traveled in 2017 than in 2016. However, the best model was the null (Table 3.5).

Table 3.5 – Model selection for distance between roost trees used by Southeastern myotis and Rafinesque's big-eared bats tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. AICc reported for Southeastern myotis and Rafinesque's big-eared bats. BAFreeze is a two-period factor: before vs. after the first freezing event of each winter. AICc is the Akaike Information Criterion corrected for small samples. Differences in AICc (Δ AICc) are reported.

Predictor	Southeastern myotis		Rafinesque's big-eared bats	
	AICc	Δ AICc	AICc	Δ AICc
Null	121.88	2.38	65.39	0.00
Week	122.01	2.51	67.06	1.67
Week * Year	123.67	4.17	67.85	2.46
Week + Year	121.65	2.15	68.18	2.79
Year	120.38	0.88	66.10	0.71
Week + Sex	124.32	4.82	68.42	3.03
Sex	123.55	4.05	66.51	1.12
Week + Year + Sex	123.96	4.46	Failed to converge	
Week * Year + Sex	126.09	6.59	66.52	1.13
BAFreeze	121.17	1.67	67.72	2.33
BAFreeze * Year	121.46	1.96	Failed to converge	
BAFreeze + Year	119.50	0.00	68.54	3.15
BAFreeze + Sex	123.45	3.95	68.93	3.54
BAFreeze + Year + Sex	121.87	2.37	70.48	5.09
BAFreeze * Year + Sex	123.92	4.42	70.28	4.89

Table 3.6 – Model selection for distance between roost trees used by Southeastern myotis tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. AICc (Akaike Information Criterion corrected for small samples) and differences in AICc ($\Delta AICc$) are reported. BAFlood is a two-period factor: before vs. after the first flooding event of each winter.

Southeastern myotis		
Predictor	AICc	$\Delta AICc$
Null	120.75	1.57
Year	119.18	0.00
Sex	122.50	3.32
BAFlood	122.40	3.22
BAFlood * Year	122.58	3.40
BAFlood + Year	121.46	2.28
BAFlood + Sex	124.51	5.33

Southeastern myotis used 1–7 different roost trees while being tracked. The model selection for before and after the freeze showed that the best model was the null model (Table 3.7). The model selection for before and after the flood event also indicated the null model was best (Table 3.8). Overall, my models suggest no role of the Flood, Freeze, Sex or Year in the number of roost trees used by this species.

Rafinesque’s big-eared bats used 1–5 different roost trees while being tracked. The best model for Rafinesque’s big-eared bats included the Freeze variable, but the null model was the most parsimonious (Table 3.7). However, the low AICc of the Freeze model suggests that the number of roosts used may tend to decrease after the freeze event. Before the first freeze event, the average number of roost trees used was 3.0 ± 0.5 and afterwards decreased to 2.1 ± 0.4 . The number of trees used by Rafinesque’s big-

Table 3.7 – Model selection for the number of roost trees used by Southeastern myotis and Rafinesque’s big-eared bats tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. BAFreeze is a two-period factor: before vs. after the first freezing event of each winter. AICc is the Akaike Information Criterion corrected for small samples. Differences in AICc (Δ AICc) are reported.

Predictor	Southeastern myotis		Rafinesque’s big-eared bats	
	AICc	Δ AICc	AICc	Δ AICc
Null	59.49	0.00	75.66	1.30
Year	62.07	2.58	77.90	3.54
Sex	62.03	2.54	76.97	2.61
BAFreeze	62.07	2.58	74.36	0.00
BAFreeze * Year	68.09	8.60	80.19	5.83
BAFreeze + Year	65.05	5.56	77.11	2.75
BAFreeze + Sex	64.99	5.50	76.39	2.03

Table 3.8 – Model selection for the number of roost trees used by Southeastern myotis tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. BAFlood is a two-period factor: before vs. after the first flooding event of each winter. AICc is the Akaike Information Criterion corrected for small samples. Differences in AICc (Δ AICc) are reported.

Predictor	AICc	Δ AICc
Null	52.75	0.00
Year	62.47	9.72
Sex	63.00	10.25
BAFlood	62.38	9.63
BAFlood * Year	66.57	13.82
BAFlood + Year	64.91	12.16
BAFlood + Sex	65.16	12.41

eared bats before and after the flood event was not analyzed because only one bat was tracked after the flood.

Southeastern myotis used 1–5 tree species while being tracked. Tree species used changed over time for Southeastern myotis whether I tested for a difference before and after flood ($P = 0.024$) or for a weekly change ($P = 0.028$) when organized by genus. The difference before and after freeze was not significant when all tree species were organized by genus ($P = 0.162$) but was significant when organized as water tupelo, bald cypress (*Taxodium distichum*) and "other" ($P = 0.009$), suggesting that Southeastern myotis do switch tree species between fall and winter (Table 3.9A). Specifically, Southeastern myotis used water tupelos almost exclusively in the fall and used more tree species in the winter. In contrast, Rafinesque's big-eared bats did not switch tree species and used water tupelos exclusively during both seasons. No difference was detected for cavity types used over time for either species (Table 3.9B).

Table 3.9 – Results of chi-square tests for (A) Tree species and (B) Cavity types used by Southeastern myotis and Rafinesque's big-eared bats tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. BA Freeze and BA Flood are two-period factors: before vs. after the first freezing event or flooding event of each winter. P-values are reported.

A)	Southeastern myotis		
	P	df	X^2
BA Freeze (water tupelo, bald cypress, other)	0.010	2	9.24
BA Freeze (by genus)	0.162	8	11.76
BA Flood (by genus)	0.024	8	17.62
Julian Week (by genus)	0.028	88	115.09

B)	Southeastern myotis			Rafinesque's big-eared bats		
	<i>P</i>	df	X ²	<i>P</i>	df	X ²
BA Freeze	0.109	2	4.43	0.524	2	1.29
BA Flood	0.206	2	3.16	0.501	2	1.38
Julian Week	0.285	22	25.26	0.602	24	21.62

3.3 Acoustic Bat Activity

In 2015, 14 Southeastern myotis vetted calls were recorded between 14 December 2015 and 9 January 2016 with 3 calls occurring below 5°C and the lowest temperature at the time of recording being 1.5°C. In 2016, 133 Southeastern myotis calls were detected between 20 November 2016 and 25 January 2017. There were 20 calls at $\leq 5^{\circ}\text{C}$ with the lowest temperature at the time of recording being 0.5°C. A single Rafinesque's big-eared bat call was captured this season on 20 January at 13.2°C.

In 2017, there were 10 Anabat deployments on the levee for a total of 52 recording nights. There were 47 Southeastern myotis calls and one Rafinesque's big-eared bat call recorded between 29 November 2017 and 11 January 2018. The frequency of the observed Southeastern myotis calls was not significantly different ($P = 0.883$) from the frequency of the expected calls in each temperature range signifying that Southeastern myotis did not halt their activity to avoid freezing nightly temperatures (Table 3.10). The lowest temperature was -2.9°C at the time of recording.

Table 3.10 – Number of nights during which at least one Southeastern myotis call was expected based on minimum nightly temperature vs. recorded in Cache River National Wildlife Refuge, Arkansas, in winter 2017-18.

Minimum Nightly Temperature	Expected Nights with a bat call	Observed Nights with a bat call
<0°C	29	7
0-5°C	9	1
5-10°C	9	3
>10°C	5	1

3.4 Roost Tree Characteristics Analysis

Through radio-telemetry 33 confirmed roost trees were found for Rafinesque's big-eared bats. All confirmed roost trees were water tupelos. They most frequently used Upper cavity types (n = 16; 48.5%), followed by Chimney (n = 11; 33.3%) and Basal (n = 6; 18.2%). Rafinesque's big-eared bats roosted exclusively in trees that were alive, choosing either fully intact trees or trees with dead or missing sections. Roost DBH ranged from 56–130.2 cm (mean = 87.5 cm) and height ranged from 13.4–37 m (mean = 21.7 m).

Forty-one 41 confirmed roost trees were found for Southeastern myotis by using radio-telemetry. The most frequently used tree species was water tupelo (n = 24; 58.5%), followed by bald cypress (n = 4), ash spp. (*Fraxinus* spp.; n = 3), red maple (*Acer rubrum*; n = 2), hickory spp. (*Carya* spp.; n = 2), oak spp. (*Quercus* spp.; n = 2), American elm (*Ulmus americana*; n = 2), American sweetgum (*Liquidambar styraciflua*; n = 1), and honey locust (*Gleditsia triacanthos*; n = 1). Upper cavity types (n = 24; 58.5%) were most often used by Southeastern myotis, but Basal (n = 10; 24.4%) and

Chimney ($n = 7$; 17.1%) cavity types were also used. One tree used by Southeastern myotis was dead with major decay, but all other trees were either alive and fully intact or alive with dead or missing sections. Roost DBH ranged from 22.5–146.1 cm (mean = 68.3 cm) and height ranged from 10.1–41.5 m (mean = 24.8 m).

Plot-level Analysis

At the 5-m radius tree plot scale, for Southeastern myotis, four strong correlations were found: Small DBH with Other tree species ($r = 0.782$, $P < 0.001$), Medium DBH with Decay class 1 ($r = 0.727$, $P < 0.001$), and Water Tupelo with Medium DBH ($r = 0.722$; $P < 0.001$) and Decay class 1 ($r = 0.724$, $P < 0.001$). These pairs were not put together in models. The best model included Bald Cypress only (Table 3.11A). The likelihood of a roost being used by a Southeastern myotis decreased with increasing numbers of Bald Cypresses in the 5-m radius plot (slope = -0.70 ± 0.39).

At the 5-m radius tree plot scale, for Rafinesque's big-eared bats, three strong correlations were found: Small DBH with Other tree species ($r = 0.784$, $P < 0.001$), Medium DBH with Water Tupelo ($r = 0.744$, $P < 0.001$), and Medium DBH with Decay class 1 ($r = 0.731$, $P < 0.001$). These pairs were not put together in models. The best model had only Other tree species (Table 3.11B). The likelihood of a roost being used by a Rafinesque's big-eared bat decreased with increasing numbers of "other" tree species in the 5-m radius plot (slope = -0.46 ± 0.21).

Table 3.11 – Model selection for characteristics of roost and random 5-m tree plots used by Southeastern myotis (A) and Rafinesque’s big-eared bats (B) tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. Characteristics were diameter at breast height (DBH; Small, Medium and Large), decay class, tree species, and basal area. AICc (Akaike Information Criterion corrected for small samples) and differences in AICc (Δ AICc) are reported.

A) Southeastern myotis			B) Rafinesque's big-eared bat		
Predictor	AICc	Δ AICc	Predictor	AICc	Δ AICc
Null	79.64	3.31	Null	46.49	7.13
Small DBH	81.77	5.44	Small DBH	44.39	5.03
Medium DBH	81.79	5.46	Medium DBH	46.70	7.34
Large DBH	80.42	4.09	Large DBH	48.72	9.36
Decay Class 1	81.68	5.35	Decay Class 1	48.69	9.33
Decay Class 2	81.66	5.33	Decay Class 2	48.63	9.27
Decay Class 3	81.77	5.44	Decay Class 3	48.78	9.42
Water Tupelo	81.55	5.22	Water Tupelo	44.20	4.84
Bald Cypress	76.33	0.00	Bald Cypress	48.69	9.33
Other	81.05	4.72	Other	39.36	0.00
Basal Area	81.57	5.24	Basal Area	45.76	6.40
Bald Cypress + Small DBH	77.98	1.65	Other + Medium DBH	39.38	0.02
Bald Cypress + Medium DBH	78.49	2.16	Other + Large DBH	41.07	1.71
Bald Cypress + Large DBH	80.74	4.41	Other + Decay Class 1	40.53	1.17
Bald Cypress + Decay Class 1	78.45	2.12	Other + Decay Class 2	41.45	2.09
Bald Cypress + Decay Class 2	78.05	1.72	Other + Decay Class 3	41.34	1.98
Bald Cypress + Decay Class 3	78.54	2.21	Other + Water Tupelo	39.59	0.23
Bald Cypress + Water Tupelo	78.56	2.23	Other + Bald Cypress	41.66	2.30
Bald Cypress + Other	77.96	1.63	Other + Basal Area	40.74	1.38
Bald Cypress + Basal Area	79.21	2.88			

At the 11.3-m radius tree plot scale, for Southeastern myotis, four strong correlations were found: Small DBH with Other tree species ($r = 0.858$, $P < 0.001$), Medium DBH with Decay class 1 ($r = 0.893$, $P < 0.001$), Water Tupelo with Medium DBH ($r = 0.802$, $P < 0.001$), Water Tupelo with Large DBH ($r = 0.739$, $P < 0.001$), and Water Tupelo with Decay class 1 ($r = 0.811$, $P < 0.001$). These pairs were not put together in models. The best model was the additive model with Bald Cypress, Medium DBH, and Decay Class 2, but the most parsimonious model was the additive model with Bald Cypress and Medium DBH (Table 3.12A). The likelihood of a roost being used by a Southeastern myotis decreased with increasing numbers of Bald Cyresses in the 11.3-m plot (slope = -0.18 ± 0.11) and increased with trees of Medium DBH in the 11.3-m radius plot (slope = 0.05 ± 0.02).

At the 11.3-m radius tree plot scale, for Rafinesque's big-eared bats, four strong correlations were found: Small DBH with Other tree species ($r = 0.831$, $P < 0.001$), Medium DBH with Water Tupelo ($r = 0.880$, $P < 0.001$), Medium DBH with Decay Class 1 ($r = 0.760$, $P < 0.001$), and Large DBH with Water Tupelo ($r = 0.799$, $P < 0.001$). These pairs were not put together in models. The best model was additive with Other tree species and Medium DBH. However, the most parsimonious was the model with only Other tree species (Table 3.12B). The likelihood of a roost being used by a Rafinesque's big-eared bat decreased with increasing numbers of "other" tree species in the 11.3-m radius plot (slope = -0.19 ± 0.08).

Table 3.12 – Model selection for characteristics of roost and random 11.3-m tree plots used by Southeastern myotis (A) and Rafinesque’s big-eared bats (B) tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. Characteristics were diameter at breast height (DBH; Small, Medium and Large), decay class, tree species, and basal area. AICc (Akaike Information Criterion corrected for small samples) and differences in AICc (Δ AICc) are reported.

A) Southeastern myotis		
Predictor	AICc	Δ AICc
Null	79.64	3.31
Small DBH	81.67	5.34
Medium DBH	79.09	2.76
Large DBH	81.26	4.93
Decay Class 1	79.78	3.45
Decay Class 2	80.40	4.07
Decay Class 3	81.71	5.38
Water Tupelo	80.85	4.52
Bald Cypress	79.80	3.47
Other	80.51	4.18
Basal Area	81.57	5.24
Medium DBH + Small DBH	81.21	4.88
Medium DBH + Large DBH	81.33	5.00
Medium DBH + Decay Class 2	80.62	4.29
Medium DBH + Decay Class 3	81.26	4.93
Medium DBH + Bald Cypress	77.18	0.85
Medium DBH + Other	79.02	2.69
Medium DBH + Basal Area	81.32	4.99
Medium DBH + Bald Cypress + Small DBH	79.04	2.71
Medium DBH + Bald Cypress + Large DBH	79.14	2.81
Medium DBH + Bald Cypress + Decay Class 2	76.33	0.00
Medium DBH + Bald Cypress + Decay Class 3	79.46	3.13
Medium DBH + Bald Cypress + Other	77.36	1.03
Medium DBH + Bald Cypress + Basal Area	79.50	3.17
Medium DBH + Bald Cypress + Decay Class 2 + Small DBH	78.73	2.40
Medium DBH + Bald Cypress + Decay Class 2 + Large DBH	77.21	0.88
Medium DBH + Bald Cypress + Decay Class 2 + Decay Class 3	78.75	2.42
Medium DBH + Bald Cypress + Decay Class 2 + Other	78.37	2.04

Medium DBH + Bald Cypress + Decay Class 2 + Basal Area	78.57	2.24
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B) Rafinesque's big-eared bat		
Predictor	AICc	ΔAICc
Null	46.49	14.89
Small DBH	41.29	9.69
Medium DBH	42.87	11.27
Large DBH	46.82	15.22
Decay Class 1	48.42	16.82
Decay Class 2	48.59	16.99
Decay Class 3	46.91	15.31
Water Tupelo	40.71	9.11
Bald Cypress	46.57	14.97
Other	33.23	1.63
Basal Area	45.76	14.16
Other + Medium DBH	31.60	0.00
Other + Large DBH	35.49	3.89
Other + Decay Class 1	32.99	1.39
Other + Decay Class 2	35.04	3.44
Other + Decay Class 3	35.51	3.91
Other + Water Tupelo	33.20	1.60
Other + Bald Cypress	34.23	2.63
Other + Basal Area	35.67	4.07
Other + Medium DBH + Large DBH	33.33	1.73
Other + Medium DBH + Decay Class 2	33.76	2.16
Other + Medium DBH + Decay Class 3	34.21	2.61
Other + Medium DBH + Bald Cypress	33.35	1.75
Other + Medium DBH + Basal Area	33.99	2.39

Tree-level Analysis

At the roost tree scale, no variables were strongly correlated for either bat species. For Southeastern myotis initial model selection showed that AIC values for Year and Tree Species were greater than the null and could be excluded from future analysis (Table 3.13A). The next series of model selections revealed that the model with only DBH

produced the lowest AIC; all other models had $\Delta AIC < 2$ (Table 3.14A). These included combinations of the DBH, Tree height and Decay class variables, though the null model was the most parsimonious.

Table 3.13 – Model selection for characteristics of roost trees (Diameter at breast height [DBH], Decay class, Tree species, Tree height, Year) used by Southeastern myotis (A) and Rafinesque’s big-eared bats (B) tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas, and their associated random trees. AIC (Akaike Information Criterion) are presented.

A) Southeastern myotis				
	DBH	Decay Class	Species	Height
Interaction	118.59	119.17	122.30	120.29
Additive	116.61	117.23	120.50	118.44
Year	117.68	117.68	117.68	117.68
Characteristic	114.77	115.27	118.52	116.44
Null	115.68	115.68	115.68	115.68

B) Rafinesque's big-eared bat				
	DBH	Decay Class	Species	Height
Interaction	88.70	79.29	78.27	97.80
Additive	90.80	79.78	76.27	97.45
Year	95.50	95.50	95.50	95.50
Characteristic	88.81	78.51	74.27	95.45
Null	93.50	93.50	93.50	93.50

Initial model selection for Rafinesque’s big-eared bats showed that Year was greater than the null and could be excluded from future analysis (Table 3.13B). The next series of model selections revealed that best model included Tree species and DBH (Table

3.14B). The wider roost trees were more likely to be occupied (slope = 0.04 ± 0.02) and more likely to be water tupelos than random trees in the area (Fig. 3.3).

Table 3.14 – Model selection for characteristics of roost and random trees (Diameter at breast height [DBH], Decay class, Tree height) used by Southeastern myotis (A) and Rafinesque’s big-eared bats (B) tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas, and their associated random trees. AIC is the Akaike Information Criterion. Differences in AIC (Δ AIC) are reported.

A) Southeastern myotis		
Predictor	AIC	Δ AIC
Null	115.68	0.91
DBH	114.77	0.00
Decay class	115.27	0.50
Tree height	116.44	1.67
Tree height + Decay class	115.07	0.30
DBH + Decay class	115.62	0.85
DBH + Tree height	116.16	1.39

B) Rafinesque's big-eared bats		
Predictor	AIC	Δ AIC
Null	93.50	26.21
DBH	88.81	21.52
Tree Species	74.27	6.98
Decay class	78.51	11.22
Tree height	95.45	28.16
Tree height + Decay class	80.27	12.98
Tree height + DBH	90.14	22.85
Decay class + DBH	76.90	9.61
Tree Species + Tree Height	73.31	6.02
Tree Species + Decay class	72.16	4.87
Tree Species + DBH	67.29	0.00
Tree Species + DBH + Tree Height	68.76	1.47
Tree Species + DBH + Decay Class	68.30	1.01

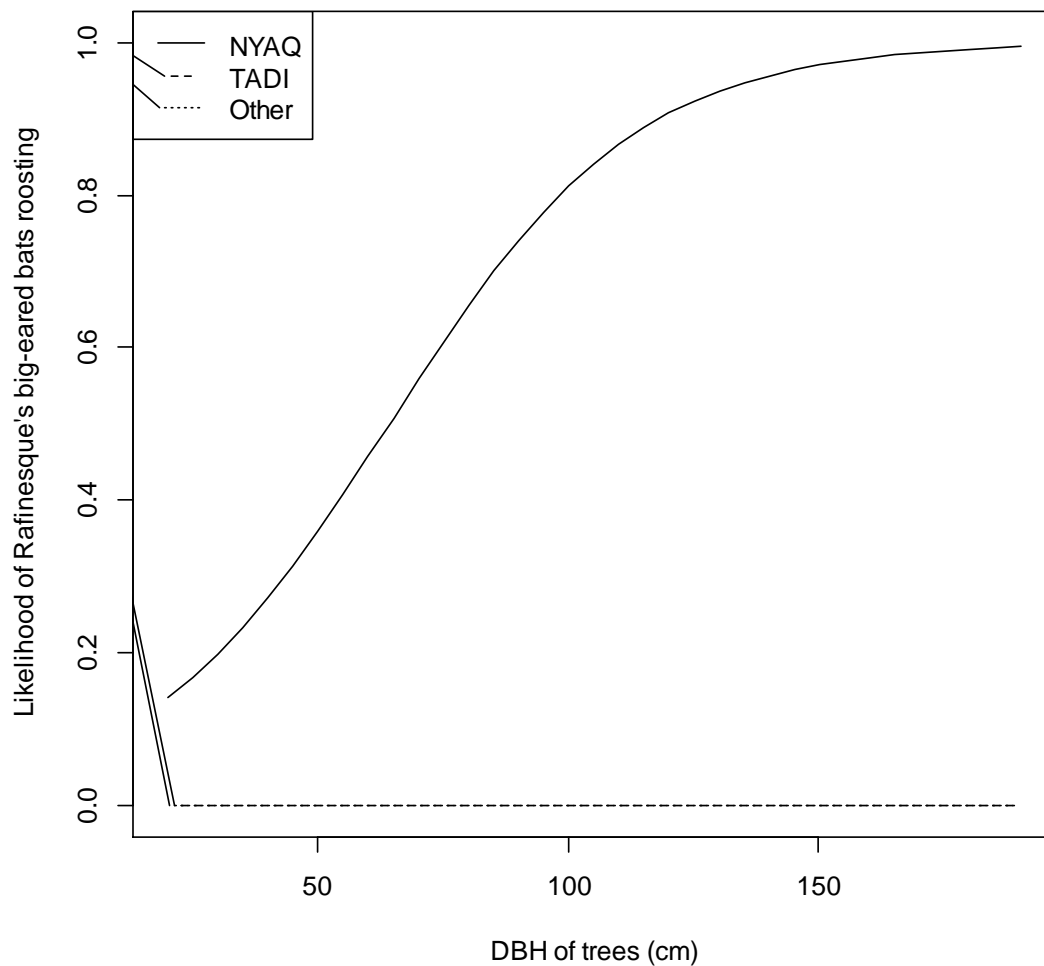


Figure 3.3 – Probability of a tree to be occupied by Rafinesque’s big-eared bats as a function of diameter at breast height (DBH) and tree species in Cache River National Wildlife Refuge, Arkansas in winters 2016-2017 and 2017-2018. Probabilities were estimated from my best model. Tree species were Water tupelo (*Nyssa aquatica*; NYAQ), Bald Cypress (*Taxodium distichum*; TADI), and other tree species. The probability of a tree to be occupied was null for TADI and other trees, regardless of DBH.

The model selection for cavity types used by Southeastern myotis revealed that the null was the best (Table 3.15), indicating that there was no difference between the cavity types of roost trees and random trees. Conversely, the model selection for Rafinesque's big-eared bats showed that the cavity model was the best model (Table 3.15). The chance that a basal tree was occupied by a Rafinesque's big-eared bat was lower ($30 \pm 10.2\%$) than for a chimney tree ($80 \pm 10.3\%$).

Table 3.15 – Model selection for cavity types used by Southeastern myotis and Rafinesque's big-eared bats tracked in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas, and their associated random trees. AICc is the Akaike Information Criterion corrected for small sample size. Differences in AICc ($\Delta AICc$) are reported.

	Southeastern myotis		Rafinesque's big-eared bat	
	AICc	$\Delta AICc$	AICc	$\Delta AICc$
Null	50.61	0.00	50.61	6.79
Combined Cavities	50.89	0.28	43.82	0.00

Cavity-level Analysis

At the cavity scale, for both species, no variables were strongly correlated. The best model for Southeastern myotis was the additive model of internal volume and cavity height from ground (Table 3.16). The likelihood of a roost being used by a Southeastern myotis increased with internal volume (slope = 0.27 ± 0.09) and with cavity height from ground (slope = 2.50 ± 1.05). The best model for Rafinesque's big-eared bats included only internal volume (Table 3.16). Similarly to Southeastern myotis, the likelihood of a

roost being used by a Rafinesque's big-eared bat increased with internal volume (slope = 0.07 ± 0.03).

Table 3.16 – Model selection for characteristics of roost and random cavities (Area of cavity opening, Height of opening from ground, Orientation of cavity opening, and Internal Volume) used by Southeastern myotis and Rafinesque's big-eared bats in winters 2016-17 and 2017-18 in Cache River National Wildlife Refuge, Arkansas. AICc is the Akaike Information Criterion corrected for small samples. Differences in AICc ($\Delta AICc$) are reported.

Predictor	Southeastern myotis		Rafinesque's big-eared bat	
	AICc	$\Delta AICc$	AICc	$\Delta AICc$
Null	74.65	27.31	65.37	12.06
Area	76.76	29.42	65.35	12.04
Volume	51.18	3.84	53.31	0.00
Height	73.03	25.69	65.19	11.88
Orientation	76.79	29.45	64.84	11.53
Volume + Height	47.34	0.00	55.36	2.05
Volume + Orientation	53.33	5.99	53.39	0.08
Volume + Area	51.91	4.57	53.54	0.23
Volume + Height + Area	49.49	2.15		
Volume + Height + Orientation	49.61	2.27		

3.5 Roost Microclimate Analysis

Of the iButtons deployed in the 14 roost trees, 14 random trees, and at 3 ambient locations, all iButtons were retrieved and downloaded except one from a roost tree that was lost. These iButtons were deployed between 5 November 2017 and 4 January 2018 and all were recovered 23 March 2018.

The model selection used to determine if the flood had an influence on the stability of the internal temperature of the roost trees indicated that the best model included cavity type (Table 3.17). Trees with chimney cavities (Fig. 2.3) had a less stable internal temperature (CV = 29.4%; CI₉₅ = 24.1%, 36.0%) than trees with upper cavities (CV = 19.9%; CI₉₅ = 16.3%, 24.3%). However, neither cavity type experienced a difference in temperature stability before and after the flood event.

The model selection used to determine if the flood had an influence on the internal relative humidity of the roost trees indicated that the best model included the additive effect of Flood and cavity type, but the most parsimonious model only included the Flood variable (Table 3.17). Internal relative humidity before the flood was $92.38 \pm 1.88\%$ and increased to $99.50 \pm 2.02\%$ after the flood. Trees with upper cavities (Fig. 2.3) were more humid than trees with chimney cavities before and after the flood (Table 3.18). Each cavity type increased internal humidity after the flood (Table 3.18).

Table 3.17 – Model selection used to determine if the flood influenced the stability of the internal temperature or internal relative humidity of roost trees in the winter of 2017-18 in Cache River National Wildlife Refuge, Arkansas. BAFlood is a two-period factor: before vs. after the first freezing event of each winter. AICc is the Akaike Information Criterion corrected for small samples. Differences in AICc (Δ AICc) are reported.

Predictor	Relative Humidity		CV Temperature	
	AICc	Δ AICc	AICc	Δ AICc
Null	27658.6	674.6	340.3	3.0
Cavity Type	27657.9	673.9	337.3	0.0
Flood	26984.7	0.7	342.2	4.9
Flood + Cavity Type	26984.0	0.0	339.1	1.8

Table 3.18 – Mean (\pm SE) internal relative humidity (%) recorded before and after the first flood event of 2017-18 in upper and chimney cavity type roost trees in Cache River National Wildlife Refuge, Arkansas.

	Upper	Chimney	Overall
Before	97.3 \pm 0.14	87.9 \pm 0.29	92.6 \pm 0.25
After	100.0 \pm 0.0	98.9 \pm 0.05	99.5 \pm 0.04

Overall temperatures and variability within roost cavities were similar to random cavities but higher and more stable than ambient (Table 3.19). Initially, for internal temperature coefficient of variation (CV), the Location type model and the Cavity type model were both better than the null model, indicating a difference between Ambient and trees (roost or random) in temperature variability. The individual model selections without the ambient temperature data indicated no difference (Table 3.20): (A) Among chimney cavities, the null model was the best model showing no difference in CV between roost and random trees. (B) Among random trees, the null model was the best model indicating no difference in CV between chimney and basal cavities. (C) Among roost trees, the cavity type model had the lowest AIC, but the null model was the most parsimonious model. However, the low value of the cavity type model may indicate a tendency for upper cavities ($CV = 0.37 \pm 0.12$) to be more stable than chimney cavities ($CV = 0.78 \pm 0.23$) throughout the season.

Table 3.19 – Overall internal tree temperature characteristics and mean internal humidity at Cache River National Wildlife Refuge, Arkansas, between 5 January and 22 March 2018.

	Roost	Random	Ambient
Mean \pm SD ($^{\circ}$ C)	8.1 \pm 5.7	8.0 \pm 5.6	8.1 \pm 7.5
Coefficient of variation	69%	69%	93%
Relative Humidity (%)	97.4 \pm 0.05	92.0 \pm 0.12	86.5 \pm 0.15

Table 3.20 – Model selections for effects of (A) Location type with chimney cavities, (B) Cavity type of random trees, and (C) Cavity type of roost trees on internal temperature coefficient of variation. All temperature data collected in cavities of roost and random trees within Cache River National Wildlife Refuge, Arkansas, from 5 January through 22 March 2018. AIC (Akaike Information Criterion) and differences in AIC (Δ AIC) are reported.

Predictor	AIC	Δ AIC
A) Null	860.37	0.00
Location Type	861.20	0.83
B) Null	-31.38	0.00
Random Cavity Type	-30.57	0.81
C) Null	729.23	0.53
Roost Cavity Type	728.70	0.00

Initially, for internal minimum nightly temperature, the Location type model and the Cavity type model were both better than the null model, indicating a difference between ambient air and tree cavities (roost or random). The individual model selections without the ambient temperature data indicated no difference (Table 3.21): (A) Among chimney

cavities, the null model was the best model, indicating no difference between the minimum nightly temperature for roost and random trees. (B) Among random trees, the Cavity type model was the lowest; however, the null model was the most parsimonious. The low value for the Cavity type model may show a tendency for the minimum temperature for basal cavities to be higher ($7.38 \pm 0.4^{\circ}\text{C}$) than chimney cavities ($6.2 \pm 0.5^{\circ}\text{C}$). (C) Among roost trees, the null model was the best model indicating no difference between the minimum nightly temperature for basal and chimney trees.

Table 3.21 – Model selections for effects of (A) Location type with chimney cavities, (B) Cavity type of random trees, and (C) Cavity type of roost trees on internal minimum nightly temperature. All temperature data collected in cavities of roost and random trees within Cache River National Wildlife Refuge, Arkansas, from 5 January through 22 March 2018. AIC (Akaike Information Criterion) and differences in AIC (ΔAIC) are reported.

Predictor	AIC	ΔAIC
A) Null	6900.54	0.00
Location Type	6901.04	0.50
B) Null	7964.46	0.82
Random Cavity Type	7963.64	0.00
C) Null	7328.68	0.00
Roost Cavity Type	7329.90	1.22

For internal mean daily temperature, the null model was better than the Location or Type models, indicating no difference between mean daily temperatures of roost trees, random trees, and ambient environment.

Roost trees had a higher mean relative humidity (range: 84.3–100%) than random trees (range: 63.9–100%; Table 3.22). Overall, upper cavity types had a higher mean internal relative humidity than other cavity types and all cavity types were more humid than the ambient environment (Table 3.22). The model selection used to determine if there was a difference in internal relative humidity between Location types (Roost, Random or Ambient) indicated that the most parsimonious model was the null model (Table 3.23A). Similarly, the individual model selection without the ambient temperature data indicated the Location type model was best, but the most parsimonious model was the null (Table 3.23B). However, the low AIC value of the Location type model in both steps suggests that the internal relative humidity of roost trees ($97.76 \pm 1.89\%$) may tend to be slightly higher than that of random trees ($91.51 \pm 2.09\%$).

Table 3.22 – Mean internal relative humidity (%) of upper, chimney and basal cavity types and ambient environment from 5 January through 22 March 2018 in Cache River National Wildlife Refuge, Arkansas.

	Upper	Chimney	Basal	Ambient
Overall	97.0 ± 0.04	95.2 ± 0.04	92.2 ± 0.09	86.5 ± 0.05
Roost Trees	97.0 ± 0.06	97.8 ± 0.03		
Random Trees		91.6 ± 0.06	92.2 ± 0.13	

Table 3.23 – Model selections for effects of (A) Location type or Cavity type of trees, and (B) Location type of chimney trees on the internal relative humidity. All humidity data collected in cavities of roost and random trees within Cache River National Wildlife Refuge, Arkansas, from 5 January through 22 March 2018. AIC (Akaike Information Criterion) and differences in AIC (Δ AIC) are reported.

Predictor	AIC	Δ AIC
A) Null	537595.5	0.70
Location Type	537594.8	0.00
Opening Type	537598.2	3.40
B) Null	152532.6	1.70
Location Type	152530.9	0.00

3.6 Man-Made Structures

Only reproductive male Southeastern myotis ($n = 3$) were observed during the bridge surveys and the HWY 680 bridges were the only bridges where they were observed. Only one bat was observed during bridge surveys in 2016. It was captured and banded from the 680-East bridge. On October 8 2017, two bats were observed at the 680-West bridge. Only one of these was successfully captured and banded. This bat was recaptured at the 680-East bridge two months later on 2 December 2017. No other bats were detected at any other bridge for the duration of the seasons.

Both iButtons were retrieved from the 680-West and 680-East bridges. Unfortunately, the iButton from the 680-West bridge failed to record data. The iButton from 680-East successfully recorded temperature and humidity data from within the expansion joint every hour from 2 December 2017 through 23 March 2018. Temperatures in the bridge

appear more stable than the ambient environment (Fig. 3.4). The mean temperature in the bridge was similar to that of tree roosts and the ambient environment but the coefficient of variation was higher than roost trees and closer to the ambient environment (Table 3.24). Relative humidity in the bridge was lower than roost trees and the ambient environment, but the standard deviation was much larger (Table 3.24).

Table 3.24 – Temperature characteristics and mean internal humidity of 680-East bridge, roost trees and ambient environment at Cache River National Wildlife Refuge, Arkansas, between 2 December 2017 and 23 March 2018.

	Bridge	Roost	Ambient
Mean \pm SD ($^{\circ}$ C)	7.22 \pm 6.4	8.1 \pm 5.7	8.1 \pm 7.5
Coefficient of variation	89%	69%	93%
Relative Humidity (%)	79.2 \pm 17.6	97.4 \pm 0.05	86.5 \pm 0.15

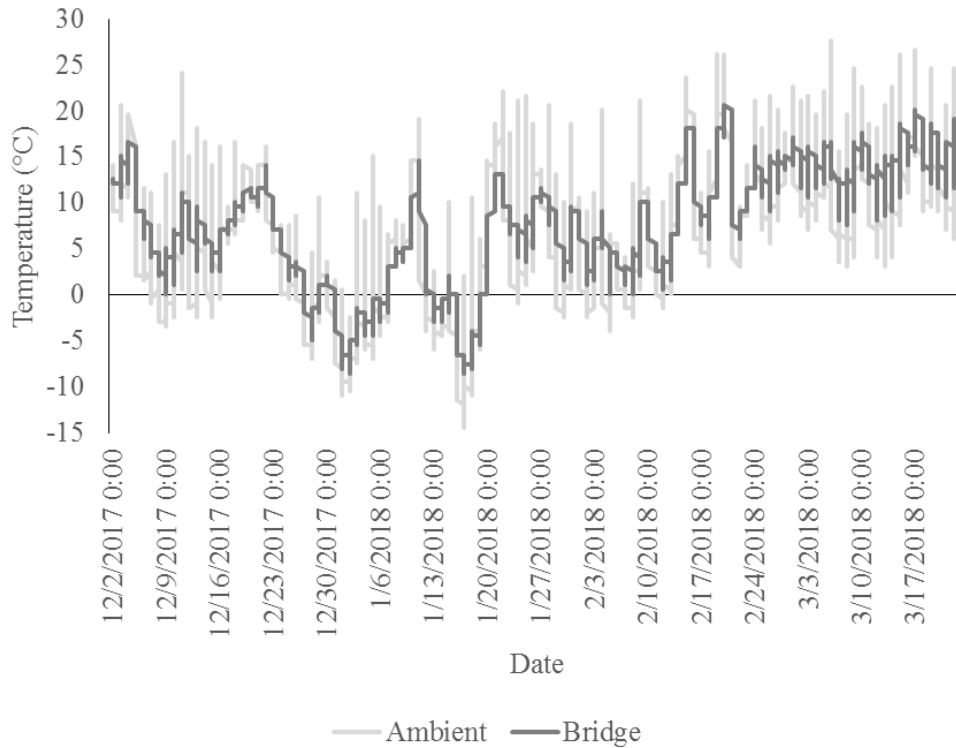


Figure 3.4 – Temperatures of 680-East bridge and Ambient environment at Cache River National Wildlife Refuge, Arkansas, between 2 December 2017 and 23 March 2018.

3.7 Water Elevation

The only trees used while water was in the cavity were Tree 79 and Tree 81, which are both chimney trees with an additional opening allowing bats access despite flooding (Table 3.25). However, Tree 79 was not as connected with the channel as anticipated, being in a neighboring slough. Due to this location, on all four days that the measurements show Tree 79 to be flooded, it was actually dry. On the contrary, on the day the measurements show that Tree 81 was flooded, the bottom cavity was fully inundated with water, showing this method can be accurate if the tree location is fully

connected with the channel. Tree 81 was a water tupelo with a chimney cavity type used by a male Southeastern myotis on 27 December 2016. The bottom cavity of Tree 81 began accumulating water with the initial flood event but did not become fully inundated until 25 December 2016, 2 days before it was used. It remained fully inundated for 13 total days until waters began to drop 7 January 2017.

Table 3.25 – Number of days Southeastern myotis and Rafinesque’s big-eared bats were tracked to measured trees while the tree was inundated with water or dry (based on measurements) in Cache River National Wildlife Refuge, Arkansas in winter 2016-17.

	No Water	Fully Inundated	Cavity Type
Tree 13	1	0	Basal
Tree 72	1	0	Upper
Tree 73	3	0	Upper
Tree 74	1	0	Upper
Tree 79	0	4	Chimney
Tree 80	2	0	Upper
Tree 81	0	1	Chimney
Tree 50*	4	0	Basal

*Observed bats opportunistically

The pressure transducer in the main channel of the McNeil farm property successfully recorded hourly water level changes from 1 October 2016 through 1 January 2017 through the entire season. The water level varied by up to 1.47 m throughout the study season with the highest water level on 31 December 2016 (Fig. 3.5). For most of the season, the water levels were gradually decreasing on the landscape with the pressure transducer recording an average hourly change of -0.1 ± 0.1 mm between 1 October and 29 November 2016 (Fig. 3.5). However, over a period of 5 days (30 November through 4 December 2016), the water levels increased 1.1 m total or an average of 8.8 ± 0.9 mm

hourly (Fig. 3.5), with the most rapid water level increase being on 1 December when the average change per hour was 23 ± 0.8 mm.

The pressure transducer recorded additional data before and after the study season. These recordings occurred from 11 July 2016 through 27 May 2017 and show that the study season from October to December is one of the driest times of the year (Fig. 3.5). The water level varied by up to 2.1 m throughout the study property with the highest water level occurring on 4 May 2017 (Fig. 3.5). However, data from the USGS gauge 07077730 upstream from Cache River National Wildlife Refuge shows that gradual decreases in fall water levels are common annually. Additionally, the USGS gauge shows that the high water level in May 2017 is not a common yearly trend, yet the peak water level is generally in the spring (Fig. 3.6).

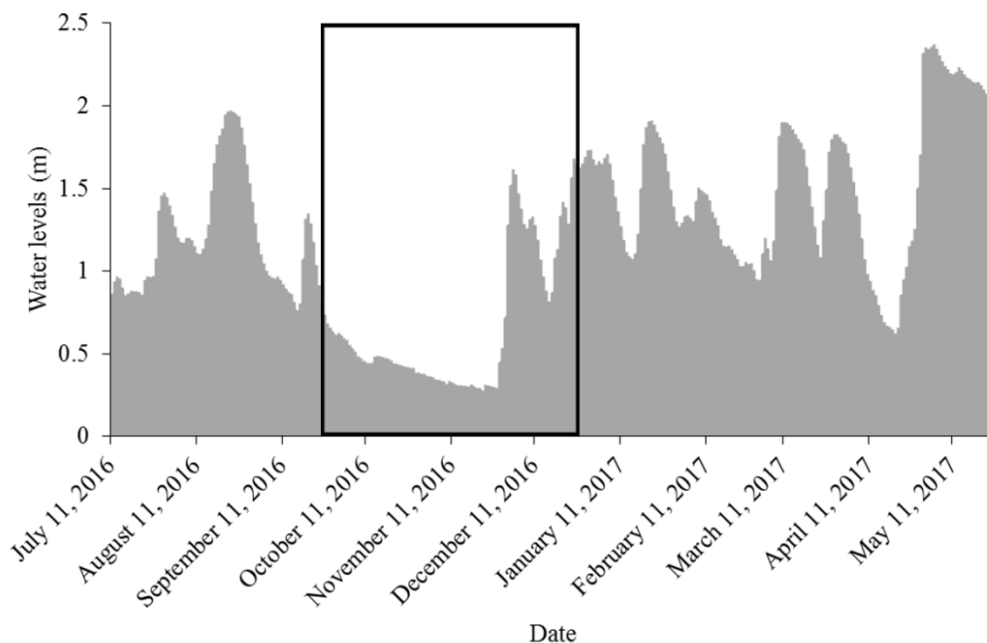


Figure 3.5 – Water levels measured by Pressure Transducer in Cache River National Wildlife Refuge, Arkansas from 11 July 2016 through 27 May 2017. Black box indicates first winter field season 1 October 2016 through 4 January 2017.

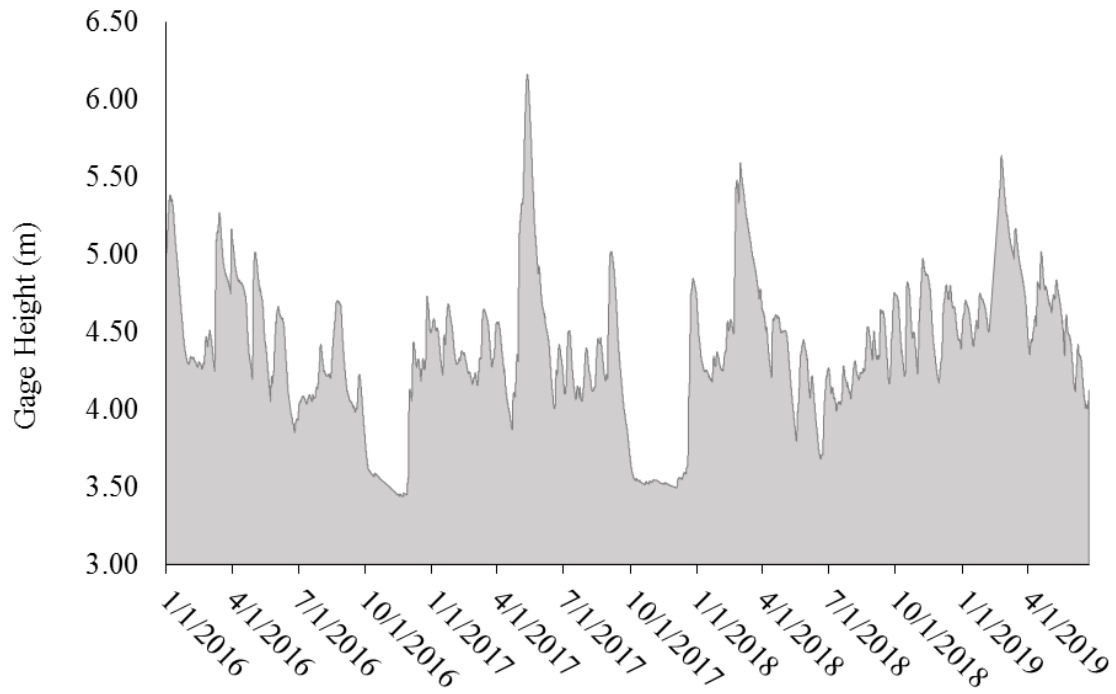


Figure 3.6 – Water levels measured by USGS gauge 07077730 in Bayou De View near Brinkley, Arkansas, from 1 January 2016 through 24 June 2019.

CHAPTER 4: DISCUSSION

This study addressed research needs to better understand the winter roosting ecology of Southeastern myotis and Rafinesque's big-eared bats in the southern part of their range where they are more active in the winter, cave and karst features are scarce, and seasonal flooding makes roost surveys difficult (Clement and Castleberry 2013a). Although this is not the first winter study on these species (e.g., Trousdale and Beckett 2004; Sasse et al. 2011; Johnson et al. 2012b; Clement and Castleberry 2013a; Fleming et al. 2013), it is unique in that I relied on radio-telemetry to locate bat roosts, eliminating a bias towards basal cavities by transect or opportunistic searches. My study highlights activity patterns and roost selection from fall to winter as flood levels rise and temperatures drop.

4.1 Seasonal Colony Composition

Although Southeastern myotis and Rafinesque's big-eared bats were only tracked to the same tree on one occasion, throughout the fall they were observed in trees together and captured in harp traps together exiting the same tree. However, they were not observed together in the winter season. Observing both bat species together in fall but not winter is partial support for my prediction that both species would share some of the same roosts in the fall and winter and corroborates findings from southwest Arkansas (Sasse et al. 2011) and Mississippi (Stevenson 2008).

Seven Rafinesque's big-eared bat roost trees and 3 Southeastern myotis roost trees were used by both males and females, which confirms my prediction that colonies would be composed of males and females for both species. These findings were previously observed in winter in Florida (Rice 1957) and Arkansas (Sasse et al. 2011). However, the sex-ratio was not balanced in these mixed groups.

Van Schaik et al. (2015) reported 7 *Myotis* species in Poland during the swarming period with a male-bias of 67% or more. In Kentucky, Indiana bats (*Myotis sodalis*) were reported with a male-bias throughout the swarming season which lasted from mid-August to late October (Cope and Humphrey 1977). All the *Myotis* species reported by van Schaik et al. (2015) and Cope and Humphrey (1977) were observed at underground sites and none were Southeastern myotis. Here, although I do not have behavioral data before October, I report a similarly high male bias from October to December for Southeastern myotis at tree roosts. A male bias at this time of year is supportive of my prediction regarding swarming behavior, as potential swarming behavior was recorded in October, but continued into December. Zinn and Humphrey (1981) noted Southeastern myotis behavior suggestive of swarming at the entrance of a culvert in February in Florida showing this behavior may continue throughout winter in some parts of their range. Thus, the difference in timing between myotis species may be because tree-roosting bats, such as Southeastern myotis, behave differently from cave-roosting bats by continuing to swarm later into the year if they remain active, do not hibernate, and are more scattered across the landscape. Rafinesque's big-eared bat winter sex ratios cannot be addressed since none of their winter roost trees were harp trapped. Interestingly, both species were observed exiting and reentering roosts or circling roosts during exit counts. Although this

occurred on several occasions, only a small number of individuals (<6) were involved each time. Cave bats are reported to swarm in numbers of over 100 (Schowalter 1980; Lowe 2012), but tree bats may exhibit this behavior in smaller numbers, simply because of their lower density in a given area (Harvey et al. 2011).

Many of my exit counts focused on roost trees with upper openings because it was the only way to safely assess colony size. This also made it impossible to know if all bats had exited the colony at a given time. However, since transmitted bats moved often but not daily, it can be assumed that exit counts reflect a low estimate of colony size at each tree. Some trees had large numbers of bats and other trees had only the transmitted bat, implying that some trees are used as social hubs for interaction (Trousdale et al. 2008). Additionally, some bat pairs were tracked to the same roost, roosted separately, then roosted together again. The fission-fusion social structure of these bat species may play a role in roost selection (Kerth and König 1999; Willis and Brigham 2004).

Results from harp-trapping and direct-counting Tree 50 several times throughout the seasons seem to show that Southeastern myotis were rarely using this tree in September and October, but that it became an important hub tree for the colony's social network later in the season. The 18 December harp-trapping event captured 65 individuals, 2 of which were recaptures from the harp-trapping event on 24 November. Interestingly, an individual was banded at this tree on 7 October, was absent on 24 November, but was recaptured on 18 December. Of the 12 total bats recaptured in 2017, 5 were recaptured in the same tree where they were caught initially from 6 days to 25 months after initial capture, showing the long-term fidelity they have to roost sites (Lewis 1995; Trousdale et al. 2008). However, the low number of recaptures from one month to the next at Tree 50

implies that this colony contains a large number of individuals that have not yet been captured and that more roost trees on the landscape have not yet been located. The fluidity of the movements of individuals into and out of Tree 50 are representative of the fission-fusion model of social behavior (Kerth and König 1999, Willis and Brigham 2004; Trousdale et al. 2008). Further evidence of this tree being an important hub tree is that while the average distance traveled by Southeastern myotis between roosts was 498.8 ± 67.7 m, 3 males and 1 female traveled over 1,101 m to arrive at this tree (with the longest distance traveled being 1,777 m), showing the social interaction anticipated at hub trees is important enough to travel longer distances than average. The social network and the tree network of both bat species should be further investigated in all seasons (Willis and Brigham 2004; Johnson et al. 2012a). Attention could be focused on what characteristics, if any, make a hub tree compared with other less frequently used roost trees on the landscape.

Examining bridges twice a month for the duration of the study was a successful method of identifying those bridges used as roost sites, and is consistent with Loeb's (2017) recommendation of surveying potential roosts multiple times to document use. Since only 3 Southeastern myotis and no Rafinesque's big-eared bats were found in bridges near the site, this is evidence that bridges were used but were not important roosting sites or social hubs in this area during the winter season (Kerth and König 1999; Johnson et al. 2012a). No bats were tracked to any other bridges or manmade structure during the course of the project. Although others have reported Southeastern myotis in bridges and culverts in the winter (Rice 1957; Katzenmeyer 2016), no one has reported the large numbers that are often found in the summer (Keeley and Tuttle 1999).

Similarly, Trousdale and Beckett (2004) report finding Rafinesque's big-eared bats in Mississippi bridges in colonies of up to 25 in the summer and only solitary individuals in the winter. Therefore, the minimal use of bridges as roost sites in the winter combined with the number of roost trees identified (Southeastern myotis $n = 41$; Rafinesque's big-eared bat $n = 33$), the small proportion of recaptures, and the high degree of movements suggest that the Cache River National Wildlife Refuge (NWR) likely provides enough suitable roost trees to sustain populations of Southeastern myotis and Rafinesque's big-eared bats. Even the 5 bats that traveled to 8 roost trees beyond the Cache River NWR border, remained in Bayou de View, on neighboring private property within the same forested area. Daily colony composition and breeding opportunities are in fluctuation daily based on the weather and the roost switching patterns of individuals within the colony.

4.2 Seasonal Bat Activity and Roost-Switching Patterns

Seasonal activity and roosting habits must provide benefits to fitness or survival to Southeastern myotis and Rafinesque's big-eared bats to be worthwhile and likely involve benefits of thermoregulation, foraging prospects, and breeding opportunities (Clement and Castleberry 2013a). The analysis of acoustic data showed that Southeastern myotis can still be active near freezing temperatures, indicating that temperature is not the main and only factor in Southeastern myotis choosing to be active on any particular night. Other factors involved might include predation risk, pursuit of breeding opportunity, or social interaction. Agosta et al. (2005) found that capture times or capture success were

not explained by nightly ambient temperatures while looking at multiple species in North America. Southeastern myotis activity was reported by Reed (2004) at a mine in Southwest Arkansas at -2.2°C . Likewise, the lowest temperature at which I recorded their call was -2.9°C . Therefore, though activity of the population may be reduced, Southeastern myotis individuals can still be active at all temperatures including temperatures below freezing, which confirms my prediction that winter flight activity would be reduced but not be eliminated as temperatures decrease.

As anticipated, only 1 Rafinesque's big-eared bat acoustic call was recorded because their echolocation calls have a low amplitude, which makes them difficult for audio equipment to record (Clement and Castleberry 2011; Lacki and Bayless 2013). Therefore, we must rely on temperature data from nights during which Rafinesque's big-eared bats were known to have switched roosts. On 6 occasions a Rafinesque's big-eared bat was confirmed to have switched roosts on a night when overnight temperatures were $6.7 - 7.8^{\circ}\text{C}$, with 1 individual switching roosts when overnight temperatures were $1.7 - -7.8^{\circ}\text{C}$. Therefore, like Southeastern myotis, Rafinesque's big-eared bats also continue activity as temperatures drop, supporting my prediction that winter flight activity would not be eliminated as temperatures decrease. Overall, my results confirm that both bat species are shallow hibernators; arousing regularly to forage, perform activities to maintain homeostatic balance (drink, urinate, etc.), and exhibit roost-switching behaviors (Jones 1977; Jones and Manning 1989; Sealander and Heidt 1990; Foley et al. 2011).

Bats were also captured by mist-net at Cache River NWR in the summers of 2014 and 2015. Of 6 species captured, the most common summer species were Rafinesque's big-eared bats (33.9%), Eastern red bats (*Lasiurus borealis*) (22.6%), and Southeastern

myotis (19.7%; Schratz et al. 2017). Conversely, during my winter study, 4 species were captured by mist-net with the most captured species being Rafinesque's big-eared bats (52%), Southeastern myotis (28.4%) and evening bats (*Nycticeius humeralis*; 18.6%), and a single Eastern red bat. (1%) Eastern red bats have been reported to be frequently active during the winter in parts of their range, including West Virginia, Indiana, Illinois, Missouri, Virginia and North Carolina (Davis and Lidicker 1956, Whitaker et al. 1997, Dunbar and Tomasi 2006), but this was not the case at the Cache River National Wildlife Refuge, Arkansas. These results highlight that comparatively, if Eastern red bats are considered active in the winter, then Rafinesque's big-eared bats and Southeastern myotis at Cache River NWR are exceptionally active in the winter.

Southeastern myotis used previously identified summer roost trees during this study. Two females and 1 male used 3 summer trees on 9 different nights. However, these trees were all used early in the season and not again after the first freeze event or flood in either year. Additionally, most Southeastern myotis roost trees (71%) identified before the freeze event were not used later in the season. Roosts used in summer and fall seasons only are likely unsuitable for winter conditions. For example, roosts with basal openings would have been fully inundated with water after the flood. The seasonal difference shows that roosting habits of Southeastern myotis changed between summer-fall and winter.

Previous studies reported Rafinesque's big-eared bats using some of the same roost sites throughout the year and showing strong fidelity to these sites (Stevenson 2008; Rice 2009). Eight Rafinesque's big-eared bat summer roosts were identified on the McNeil farm property, and 12 on the surrounding property of Cache River NWR (Schratz 2016).

However, no bats were tracked to these trees during the fall or winter and it is unlikely that all summer roosts were identified, so seasonal fidelity cannot be accurately assessed for Rafinesque's big-eared bats. It would be expected for them to also show seasonal fidelity to select trees on my site.

Individual Southeastern myotis used 1–7 different roost trees and Rafinesque's big-eared bats used 1–5 different roost trees throughout the fall and winter, which supports my prediction that individuals of both species would use multiple roost trees.

Additionally, these bats using so many individual trees for roosting throughout the year is further evidence that Cache River NWR maintains enough suitable trees for large colonies of each species to be successful.

After the first freeze event, Rafinesque's big-eared bats increased the number of consecutive days they spent at roost trees before switching roosts. They also showed a tendency to decrease the number of tree roosts used after the first freeze event. When combined, these switches may reflect an overall decrease in activity for Rafinesque's big-eared bats after the freeze event. However, the number of consecutive days at a roost was measured with daily tracking data, so it is unclear if there was no movement on those nights or if bats exited the tree and returned to the same tree without switching. In the first case it may reflect a decrease in activity, but in the second it shows that trees suitable for winter conditions are being frequently selected for and relied upon. No difference was found for Rafinesque's big-eared bats in distance travelled or cavity type used as the season changed. These bats traveled an average of 572.8 ± 640 m between tree roosts in the summer in southern Mississippi (Trousdale et al. 2008) and $2,535 \pm 437$ m between Kentucky cave roosts in the winter (Johnson et al. 2012b). By contrast, Rafinesque's big-

eared bats in Cache River NWR traveled much shorter winter distances between tree roosts (averages of 264.6 ± 28.7 m). The differences may be explained by dissimilarities in the study sites and how concentrated suitable tree roosts are on each landscape compared to caves.

Rafinesque's big-eared bats have been reported to use many different tree species as roosts (Clark 1990; Stevenson 2008; Fleming et al. 2013). They were reported to use water tupelo, bald cypress and American hornbeam in Cache River NWR in the summer (Schratz 2016), however, in my fall and winter study, they exclusively used water tupelo (*Nyssa aquatica*) trees. This may indicate a switch in their selection from summer to fall and winter, but more likely shows that roost selection depends on tree species availability at each site location or that not all roost trees were identified at my site in the summer (n = 8 summer roost trees) and more data collection is needed in this season.

Unlike Rafinesque's big-eared bats, Southeastern myotis changed the tree species they were using as roosts as the season progressed. The change was revealed when analyzed by freeze event, flood event and weekly changes. Similarly to a population in Georgia, Southeastern myotis on my site switched from using mainly water tupelo in the summer and fall to include varied hardwood species located in adjacent floodplain forests (Clement and Castleberry 2013a). The switch from cypress-gum swamps to hardwood floodplain forests will require managers to provide different summer and winter habitats for this species.

Although I found a tendency for Southeastern myotis to travel shorter distances and spend less time at a given roost after the first freeze event, none of the parameters evaluated (e.g., number of roosts, cavity types) differed significantly as the season

progressed. The lack of differences suggests that this species remains active throughout the season, although it changes tree species, maybe in anticipation of the flooding. Interestingly, if significant changes in behavior were detected with a larger sample size, my results would suggest that Southeastern myotis and Rafinesque's big-eared bats switch their habits in different and generally opposite ways; Southeastern myotis spent less time in a given roost but traveled shorter distances between roost trees, whereas Rafinesque's big-eared bats spent more time in fewer roost trees. Also, while Rafinesque's big-eared bats used 1 tree species exclusively, Southeastern myotis increased the number of tree species used as the season progressed.

Overall, this study shows little effect of flooding on roost selection or activity of either bat species. In contrast, Clement and Castleberry (2013a) reported Rafinesque's big-eared bats and Southeastern myotis exhibiting different roosting habits from summer to winter in Georgia and concluded the switch was in relation to flooding. The difference in conclusions is possibly because (1) the first flood event at my site occurred late in both seasons, resulting in limited data for the post-flooding period, (2) seasonal roost selection changes are affected by local tree availability at each site, (3) the Georgia study did not take other environmental factors into account (i.e., freezing temperatures), or (4) these species were not affected by flooding because of changes they made before the first freeze event. More data should be collected at my site and across the ranges of both bat species to see how roost selection is affected by these weather events if flooding comes before the freeze. However, my study does show that both bat species altered their behavior after the first freeze event (Southeastern bats increased number of tree species used and Rafinesque's big-eared bats spent more time in roost trees), which occurred

before the flood in both seasons, and as the season progressed. Both species exhibiting seasonal switches in roosting habits during the winter supports Clement and Castleberry's (2013a) findings that a switch occurred in winter, but these studies conflict in what triggered the switch.

During the course of this study, a bat was never found inside or tracked to a tree that was fully or partly inundated with water unless that tree had an alternate opening that was unaffected by the water levels, allowing bats to safely enter and exit. Lack of detecting or tracking a bat to a flooded cavity is evidence to support the prediction that bats would select winter tree roosts with cavity openings that are high enough to remain open during flood events. Furthermore, Clement and Castleberry (2013a) reported that both species switched to higher cavities in the winter which would be protected from flood waters and promote activity.

Both species showed strong preferences for tree roosts that were living trees. A dead tree was used on only 1 occasion by a single Southeastern myotis for only 1 night, which is consistent with findings in Georgia where both bat species used mostly living trees with the only exception being a Southeastern myotis that used a snag (Clement and Castleberry 2013a). Also, since there is no way to externally determine if an individual is a young-of-the-year or an adult during this season, I assumed all bats to be adults. However, there could be differences between the roost selection of adults and immature bats, particularly during inclement weather events due to the experience of the adults and the inexperience of the immature bats. For both species, sex was never found associated with roost selection during any season, showing that both sexes made similar choices during this study. Roost selection and activity patterns are likely to be influenced by

many factors including weather, pursuit of breeding or social opportunities, disturbance, or roost tree characteristics

4.3 Roost Tree Characteristics

At the plot scale, an increased number of bald cypresses decreased the chances a Southeastern myotis would use a roost at both the 5-m and 11.3-m scales. This coincides with the switch Southeastern myotis made to hardwood tree species which were not in close proximity to the hydrophilic bald cypress (see section 4.2). I also found that increased medium DBH (11.1–31.1 cm) trees present in the 11.3-m plot increased the chances of Southeastern myotis use. This characteristic may reflect a preference for mature tree stands lacking saplings or may be a result of a low concentration of large trees on the landscape since the site logged was 30 years ago (E. Johnson USFWS, personal communication). Increased abundance of ‘Other’ tree species decreased the chance Rafinesque’s big-eared bats would use a roost at the 5-m and 11.3-m plot scales. Rafinesque’s big-eared bats were reported to select roosts at lower elevations in the winter in Mississippi (Fleming et al. 2013). Decreased abundance of ‘Other’ trees at lower elevations is in agreement with Rafinesque’s big-eared bats exclusive selection of water tupelo trees as roosts, because this tree species tends to occupy swamps and waterways, thereby excluding other, not as hydrophilic, tree species from their proximity.

At the tree scale, Rafinesque’s big-eared bats were more likely to occupy wider trees that were water tupelo with chimney cavities than random trees in the area. In Georgia (Clement and Castleberry 2013a) and Louisiana (Rice 2009), Rafinesque’s big-eared bats

also selected for water tupelos without basal openings in the winter. None of the parameters evaluated (DBH, decay class, tree height, tree species) differed significantly for Southeastern myotis. However, in Mississippi both bat species were reported to select roost trees with larger DBH than random trees in the winter (Fleming et al. 2013). Trees with volcano openings were not included in my analyses due to a lack of random volcano trees to compare them to. This bias occurred due to the difficulties in identifying a volcano tree from the ground and proving it was unoccupied with my acoustic equipment. I recommend future studies to include upper trees in their selection of random trees by using a system to mount (i.e., ladders, PVC poles, tree climbing techniques) acoustic devices into upper openings for stronger and more comprehensive results.

At the cavity scale, the likelihood of a tree being used as a roost by either Southeastern myotis or Rafinesque's big-eared bats increased with increased internal volume. This supports the prediction that winter roosts would have larger cavities than random trees in the area and corresponds with findings at 3 sites in South Carolina for Rafinesque's big-eared bats (Loeb 2017) and in Mississippi for Southeastern myotis (Fleming et al 2013). The likelihood of a tree being used as a roost also increased with increased height from ground for Southeastern myotis, which shows that, during the winter season, this bat species selected cavities higher off the ground and less likely to be inundated with flood waters than surrounding random cavity trees. Similarly, Rafinesque's big-eared bats in Georgia also selected roosts with higher cavity openings than surrounding random trees in the winter (Clement and Castleberry 2013a). However, I did not detect this pattern at my site for this species, possibly due to differences in tree availability between the two landscapes.

Rafinesque's big-eared bats used mainly water tupelo and bald cypress at my site in the summer (Schratz 2016) but used only water tupelo during my study in the fall and winter. Bald cypress and water tupelo trees are commonly the largest on the landscape (Appendix A) and their use for roosting may be more a factor of their preference for wide DBH trees and large internal cavity volumes than for these specific tree species. Additionally, low numbers of bald cypress used at my site across seasons may be a result of a lower concentration of bald cypresses than water tupelos due to previous logging activity on the site (E. Johnson USFWS, personal communication) and not a true preference for water tupelos over bald cypresses.

Overall, the analysis of winter roost tree characteristics supports my hypothesis that both bat species selected roost trees with properties that were different from random trees in the area. In the winter, Rafinesque's big-eared bats selected roosts with large internal volume and large DBH, findings that match only trees with large DBH could also have a large internal volume. These bats selected wide water tupelos with large internal cavities and chimney openings within plots that were predominantly water tupelo and bald cypress. Southeastern myotis selected roost trees with high cavity openings and large internal volumes within plots that include trees of medium DBH and lack bald cypresses. Although both species selected larger trees than others in the area, Rafinesque's big-eared bats used water tupelo exclusively and Southeastern myotis used trees of different species, including some small trees or trees that would be considered atypical. Therefore, any conservation action or habitat management practice taken on the property should consider maintaining a diverse assortment of tree species and ages in cypress-gum swamps and adjacent hardwood floodplain forests for bats to use as roosts throughout the

year. Other roost tree characteristics that may play a role in roost selection include proximity to desirable or undesirable locations, number of conspecifics present to offer opportunity for social or breeding interaction, or internal microclimate.

4.4 Roost Microclimate

Based on the before and after flood analysis, cavity type had a significant influence on the stability of the internal temperature of roost trees, but the flood did not. Trees with upper cavities had temperatures 9.5% more stable than trees with chimney cavities, likely because upper cavities are not as subject to air flow through the inside of the chamber as the chimney trees. Similarly, trees with upper cavities tended to have more stable internal temperatures than chimney cavities when looking at CV of temperature among roost cavity types. These results likely explain why upper cavities were the most used cavity type by both bat species. For Southeastern myotis, 58.5% of identified roosts were upper cavities. Similarly, 48.5% of Rafinesque's big-eared bat roosts were upper cavity trees. Rice (2009) in Louisiana also found that upper cavities were more stable than other cavities. The fact flooded tree cavities were not more stable than dry tree cavities is likely due to the floodwaters moving rapidly through the system so retained water within tree cavities was not stagnant to aid in heat retention. However, flooding did have a significant influence on the internal relative humidity (RH) of the roost trees, with RH increasing from 92.4% to 99.5% after the flood, likely because many of the roost trees were dry before but inundated with water after the flood event.

Roost and random trees had internal temperatures similar to each other, but different from the ambient environment. Internal temperatures for both tree types were significantly more stable, and internal minimum nightly temperatures were significantly higher than the ambient environment throughout the season. This aligns with my prediction that temperatures inside winter roost tree cavities would be higher and more stable than the ambient environment since trees provide a buffering effect from outside weather, allowing bats to thermoregulate without using extra energy (Coombs et al. 2010; Clement and Castleberry 2013b). Similarly, I found a tendency for trees (roost or random) to have a higher internal RH than the ambient environment (Table 3.22A). However, the similarities between roost and random tree microclimates show that while bats may be benefitting from warm, stable internal tree microclimates as a way to reduce energy expenditure, they are not choosing trees based on this quality and instead selecting trees based on other structures (i.e., cavity type, tree size or species). Furthermore, internal tree microclimate, while more stable, is still influenced by weather fluctuations, such that greater changes in ambient temperature will cause less stable tree temperatures, making cavity microclimate difficult to predict based on external tree structures (i.e., cavity type, tree size or species; Paclik and Weidinger 2007; Clement and Castleberry 2013b). Additionally, this internal fluctuation based on ambient temperatures may allow tree bats to monitor and exploit warmer nights (Boyles et al. 2006). My analysis had to be divided into smaller segments due to a lack of random volcano trees (same bias as discussed in section 4.3). Fixing the sampling bias should be a priority since it negatively affects several analysis and combining these temperature analyses would make these analyses stronger.

Average internal roost tree temperatures (8.1 ± 5.7 °C) were similar to those found in caves in Indiana, Illinois, and Arkansas where Southeastern myotis were found at 4.4–10°C (Rice 1957), and in Kentucky caves where Rafinesque’s big-eared bats were found at 5–11°C (Johnson et al. 2012b; Table 3.19). Southeastern myotis and Rafinesque’s big-eared bats seem to select similar roost temperatures in winter whether in caves or trees. These temperatures are also comparable to the temperatures at which white-nose syndrome (WNS) produces optimal growth (7–15°C) (Blenhert et al. 2009; Bernard et al. 2015; Langwig et al. 2015). White-nose syndrome causes rapid population decline as the RH within the hibernacula increases; declines start near 90% RH (Langwig et al. 2012), which is similar to the average RH ($97.4 \pm 0.05\%$) I recorded in roost trees. However, neither bat species population has been strongly affected by the fungus. The sustained activity and shallow torpor bouts of both bat species during winter months may help them resist progression of the disease if they encounter it (Johnson et al. 2012b). Also even though my average internal tree temperatures in the winter were within the range of those found in caves exposed to WNS, interior cave temperatures are likely more stable (Poulson and White 1969) than those within trees and average internal tree temperatures in the summer are not within this range. Both of these factors would negatively affect growth of *P.destructans* (the fungus causing WNS) making its survival unlikely within trees year-round.

4.5 Water Elevation

My method of assessing water elevation and its effects on bat roost selection was not successful due to a small sample size and problems with the overall connectivity of the system due to slight elevation differences between the channel and adjacent sloughs. However, I was successful in collecting data on 1 tree that showed a bat that used a chimney tree while the bottom cavity was fully inundated with water, showing that water within the cavity is not a deterrent to roosting. The pressure transducer was 1,141 m away from this tree and both were within the same channel. Conversely, a second chimney tree was a similar distance from the pressure transducer (1,140 m) but was located in a different slough and consequently disconnected from the channel. At this location, the tree was dry when the measurements indicated that it should be fully inundated. This tree was therefore not useful to assess the effect of flooding on tree use by bats. If this surveying method is attempted again, care should be taken to measure only roost trees that are within the same channel as the pressure transducer since proximity is less important. Additionally, I began measuring the elevation of known summer trees and random trees before they were used by bats in the winter, which were all wasted efforts. This method would be more effective and efficient if only identified roost trees (and their associated random) were measured and pressure transducer results compared retroactively.

The sharp increase in water level over 5 days beginning on 30 November 2016 could potentially be problematic for bats if they were using trees within the floodplain with only basal openings, but my results show that bats avoid this hazard in the winter.

Anthropogenic alterations to waterways, such as levees, that are present along much of Bayou de View may alter the flow of the system making fluctuations in water level more extreme than would have naturally occurred. The ability of these bat species to adjust to sudden changes in water level (both natural and anthropogenic) are positive signs that they will also be able to adjust to extreme weather events (e.g., flash flooding) associated with climate change (Allen et al. 2018). Although no evidence was found that bats became trapped within roost trees as water levels change seasonally, any anthropogenic use of the waterways (i.e. discarding agricultural waters into Bayou de View) in any season that could cause a rapid or severe change in water levels should be discouraged.

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CHAPTER 5: CONCLUSION

5.1 Summary

Fall and winter are critical times of year for Southeastern myotis and Rafinesque's big-eared bats with seasonal adverse changes in weather and water levels, reduction in prey abundance, new priorities of breeding activity, and new challenges for young-of-the-year animals. The goal of this project was to increase knowledge of winter habitat use and requirements of these bottomland forest bat species. Mist-netting, acoustics, and radio-tracking techniques were used to characterize the fall and winter activity of these bats and to locate their roost trees. I addressed the research needs during these seasons that were outlined in the Arkansas Wildlife Action Plan (Fowler 2015), identified throughout their respective ranges (Lacki and Bayless 2013) and listed under mammals as Priority #2 in the 2015 Arkansas State Wildlife Grant Request for Proposal. All objectives of my study were accomplished (Table 5.1) and the increased understanding of fall and winter roosting requirements will be important to managing habitats to ensure these species thrive during all seasons.

Using acoustic devices and radio-telemetry, I determined that both species continue activities in fall and winter even as temperatures fall below freezing, although they alter their roosting choices (e.g., tree species, amount of time spent in roosts) to prepare for seasonal winter weather and water level changes. Though activity did not

cease, Rafinesque's big-eared bats exhibited a general reduction of some activities as the season progressed notably the significant increase in time spent in each roost after the freeze. Southeastern myotis changed the tree species used for roosts as the season progressed, choosing to leave water tupelos in channels and sloughs for species in adjacent hardwood floodplains, possibly as a flood-avoidance tactic. Additionally, Southeastern myotis showed seasonal differences in roost selection by selecting against summer-fall trees in the winter, likely because these trees are unsuitable for winter conditions. Continued roost switching at this time of year allows for communication and social interaction while also likely increasing breeding success. Although Southeastern myotis colonies were composed of males and females, there was a strong male bias, which was likely a function of breeding activity. However, no difference was found in roost selection between sexes.

Both bat species preferred large living trees for roosting with properties that were different from random trees. Rafinesque's big-eared bats roosted in wide water tupelos with chimney cavities within plots that were predominantly water tupelo and bald cypress. Southeastern myotis selected roost trees with high cavity openings and large internal volumes within plots that included trees of medium DBH and lacked bald cypress. Although Rafinesque's big-eared bats used water tupelo exclusively, Southeastern myotis mixed their selection with some smaller trees of different species or trees that would be considered atypical. Therefore, any habitat management for Southeastern myotis should include a diverse assortment of tree species and ages in both cypress-gum swamps and adjacent hardwood floodplain forests to provide enough suitable roosts for year-round survival. Rafinesque's big-eared bats would also benefit

from this strategy. Interestingly, the roost selection changes made by these species caused them to also switch from occasionally roosting together in the fall to segregating themselves into different but neighboring habitats on the landscape where they were no longer found to roost together in the winter.

Internal tree roost temperatures were more stable and warmer than the ambient environment, creating roosting conditions that allow for cheaper thermoregulation. Internal relative humidity (RH) of tree roosts was significantly higher after the flood. However, there was no significant difference in microclimate between roost and random trees indicating that bats are benefiting from the internal microclimate of tree roosts, but are selecting roosts based on other factors (i.e., tree species, age or cavity type).

5.2 Implications and Recommendations

Protection of tree roosts is critically important to sustain populations of Southeastern myotis and Rafinesque's big-eared bats including in fall and winter when they continue roost-switching activities. The continued activity of both species allows for frequent social interaction throughout the fall and winter, improves breeding success and allows them to benefit from potential foraging opportunities on warm nights. However, this study has shown that they need a variety of tree species, ages and cavity types to choose from year-round to support their life cycle, demands for thermoregulation and social network (Clement and Castleberry 2013). Therefore, focusing management on summer roost requirements only would be a mistake, requirements for all seasons should be considered and provided for.

Cache River National Wildlife Refuge (NWR) seems to provide suitable habitat in the summer (Schratz et al. 2017) as well as in fall and winter (this study) for both species. Therefore, this would be an ideal place to continue research as many individuals are previously banded and many seasonal roosts have been identified. More data from this site could bolster results from small-sample analyses (roost-switching), determine if Rafinesque's big-eared bats use summer roosts in the winter, estimate site fidelity, and increase season length to capture more flood data (which would improve roost-switching and microclimate analysis). Additionally, future studies on this site and across their range on fall and winter insect availability, and on the energetic costs and benefits of winter activity including foraging could be valuable for managers to maintain a suitable insect community for both species. Although no difference was found in roost selection between sexes, there may be differences in selection between young-of-the-year and adults in the fall and winter, which could be determined at this site.

Cache River NWR is clearly a suitable habitat to support large colonies of both species of bats throughout the year. The refuge is maintained and managed for winter waterfowl hunting, but is susceptible to increased encroachment by farms and other anthropogenic uses. Continued management for winter duck habitat will benefit both species of bats by reforesting areas adjacent to the refuge to increase suitable habitat. No evidence was found that bats get trapped within roost trees as water levels change seasonally. However, bats used many tree roosts that later became inundated during floods and there is evidence that flooding may happen rapidly therefore any anthropogenic use of the waterways (i.e. discarding agricultural waters into Bayou de View) in any season that could cause a rapid or severe change in water levels should be discouraged.

Even though roost and random trees exhibited average internal temperature and RH values similar to those preferred by *P. destructans*, the fungus that causes white-nose syndrome (WNS), neither bat population has been strongly affected by the disease due to their continued winter activity and shallow torpor bouts (Blenhert et al. 2009; Johnson et al. 2012b; Bernard et al. 2015; Langwig et al. 2012). Their preference for the same microclimate increases their risk of encountering the disease in the winter; however, *P. destructans* is unlikely to survive in trees year-round because summer temperatures are too high and the roost temperature is less stable in a tree cavity than in cave systems. These factors would negatively affect the growth of *P. destructans* making it unlikely to survive within trees year-round and improbable for these bat species to become significant vectors to spread WNS throughout the landscape.

More research is needed to shed light on the breeding and swarming activity and duration in other parts of their range. Fall is a critical time of year for bat species, since they reproduce slowly and loss of breeding opportunities can decrease fitness. Also, the social network and the tree network of both bat species should be further investigated in all seasons and across their range (Willis and Brigham 2004; Johnson et al. 2012a). Understanding more about their social network could lead to better understanding of social organization, including length of parental involvement, family units, harems and breeding. Attention could also be focused on what characteristics, if any, create a suitable hub tree compared to other less popular roost trees on the landscape, which could lead to a better forest management practices for these species.

Table 5.1 Summary of Objective-specific results. Results that do not completely agree with the predictions are italicized.

Objectives	
Predictions	Results
O1. Document fall and winter activity (e.g., emergence patterns, colony size) of Southeastern myotis and Rafinesque's big-eared bat.	
P.1.A. Both species will share some of the same roosts in the fall and winter.	Yes, they were caught in harp traps exiting the same tree and observed together in the fall. <i>They were not observed together in the winter.</i>
P.1.B. Colonies will be composed of male and female bats.	Yes, for both species, both sexes were tracked to the same tree multiple times.
P.1.C. Swarming activity will occur through October.	<i>Some evidence that swarming might occur later in the season with tree bats (into December), but more research is needed.</i>
P.1.D. Winter flight activity will be reduced but not eliminated as temperatures decrease.	Yes, acoustic data showed Southeastern myotis were similarly active across temperatures and nightly temperatures on roost switch nights showed Rafinesque's big-eared bats are also active across temperatures.
O2. Characterize roost trees (e.g., cavity size, orientation) used by Southeastern myotis and Rafinesque's big-eared bats during fall and winter compared to randomly selected potential roost trees.	
P.2.A. Winter roosts will have higher DBH and larger cavities than surrounding unused cavity trees.	Yes, increased internal cavity volume was a significant factor in roost selection for both species. <i>DBH was significant for Rafinesque's big-eared bats, but was not significant for Southeastern myotis.</i>
P.2.B. Some winter roost trees will be the same trees as those used in the summer.	Southeastern myotis used 3 summer trees in the fall but not in the winter. <i>Rafinesque's big-eared bats used no known summer trees.</i>

O3. Characterize habitat (e.g., basal area) surrounding confirmed roost trees compared to randomly selected locations.	
P.3.A. Individual bats will use multiple roost trees.	Yes, in both species, individuals were tracked to multiple roost trees (up to 7 in 21 days of tracking).
P.3.B. Bats will not use bridges and other manmade structures in areas <8 km from the study site.	Yes, bridges were used by 3 Southeastern myotis but were not important social hubs or roosting sites for the colony. Rafinesque's big-eared bats did not use bridges.
O4. Characterize the microclimate (i.e., temperature and humidity) of confirmed fall and winter roost trees and potential roost trees.	
P.4.A. Temperatures inside winter roost tree cavities will be higher and more stable than ambient.	Yes, internal tree temperatures were more stable and minimum nightly temperatures were higher than ambient.
O5. Determine if and how seasonal flooding and freezing affect Southeastern myotis and Rafinesque's big-eared bats.	
P.5.A. Bats in the study area will select winter tree roosts with cavity openings that are high enough to remain open during flood events.	Yes, a bat was never tracked or observed inside a flooded tree unless that tree had an alternate opening unaffected by the water levels.
CONCLUSION:	Southeastern myotis and Rafinesque's big-eared bats have effective tree roosting strategies for surviving flooding and freezing events, including switching roosting habits seasonally.

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APPENDIX A

MEASUREMENTS OF TREES AT CACHE RIVER NATIONAL WILDLIFE REFUGE, ARKANSAS

The following figures show differences in size measurements of tree species on the landscape in Cache River National Wildlife Refuge, Arkansas. For these figures, roost and random trees measured for both bat species are combined to show average sizes of available tree species on the landscape. Tree species include water tupelo (*Nyssa aquatica*; NYAQ), bald cypress (*Taxodium distichum*; TADI), and all other tree species ($n = 16$). Water tupelos have a larger diameter at breast height (DBH; Fig. A-1) and a larger internal volume (Fig. A-2) than other tree species but these measurements do not differ between water tupelos and bald cypresses.

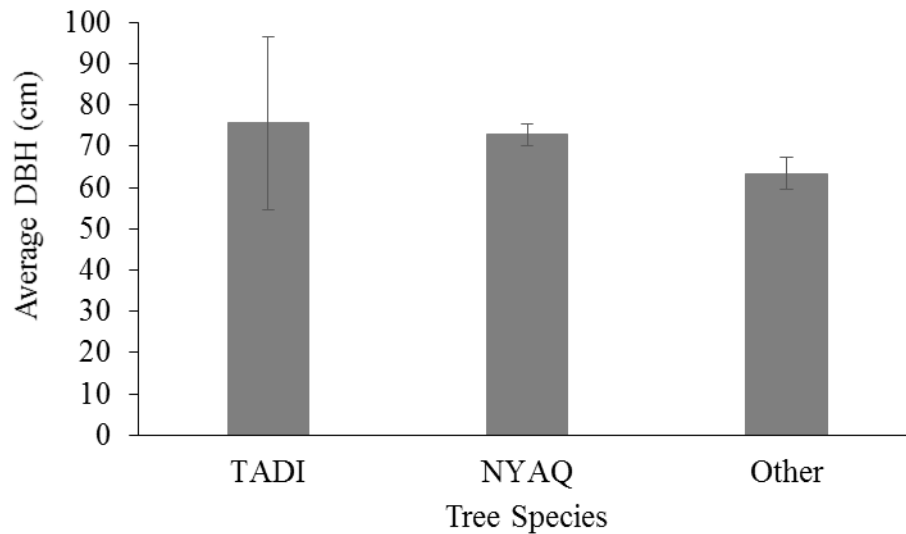


Figure A.1 – Average (± 1 SE) Diameter at breast height (DBH) of bald cypress (TADI), water tupelo (NYAQ) and other tree species measured in Cache River National Wildlife Refuge, Arkansas.

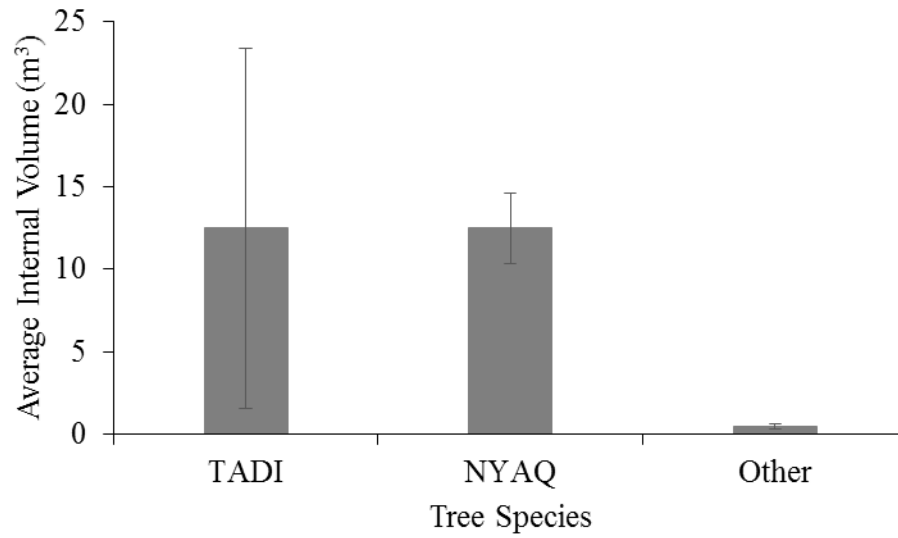


Figure A.2 – Average (\pm 1SE) internal volume of bald cypress (TADI), water tupelo (NYAQ) and other tree species measured in Cache River National Wildlife Refuge, Arkansas.