2017

HABITAT USAGE OF TRI-COLORED BATS (PERIMYOTIS SUBFLAVUS) IN WESTERN KENTUCKY AND TENNESSEE POST-WHITE NOSE SYNDROME

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HABITAT USAGE OF TRI-COLORED BATS (*PERIMYOTIS SUBFLAVUS*) IN WESTERN KENTUCKY AND TENNESSEE POST-WHITE NOSE SYNDROME

A Thesis

Presented to

the Faculty of the Department of Biology

Murray State University

Murray, Kentucky

In Partial Fulfillment

of the Requirements for the Degree

of Master of Science in Biology

Katherine Schaefer

December 2016
HABITAT USAGE OF TRI-COLORED BATS (*PERIMYOTIS SUBFLAVUS*) IN WESTERN KENTUCKY AND TENNESSEE POST-WHITE NOSE SYNDROME.

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ACKNOWLEDGEMENTS

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ABSTRACT

The tri-colored bat (Perimyotis subflavus) has declined severely across its range since 2006 due to white nose syndrome, a fungal disease causing massive bat mortality in North America. My objective was to determine distinguishing characteristics of roost trees and habitat used by tri-colored bats so that the species’ needs can be considered in management plans. I mist-netted for tri-colored bats in western Kentucky and Tennessee during the summers of 2015 and 2016, and attached a radio transmitter to each captured adult bat. I tracked 15 bats to their day roosts and collected habitat data at 38 roost areas and at 74 randomly selected trees in the area of the capture radius of each bat for comparison. Tri-colored bats used roost trees within a relatively small area. The average distance between roosts was 86 m and bats roosted within 2.5 km of their original capture site. All roosting bats were located in the foliage of live trees. Tri-colored bats’ roost tree selection was nonrandom. Bats were observed roosting in nine different species of tree, with the most commonly selected species being *Carya tomentosa* and *Quercus alba* (46% and 23% of roost trees, respectively). The most abundant species among the randomly selected trees was *Q. alba*, which was selected roughly in proportion to its abundance, and *Acer saccharum*, which was never selected as a roost tree species. A generalized linear model on all variables measured showed that increasing tree crown depth, distance from roads, and basal area of trees were correlated with roost tree selection. Management needs of tri-colored bats differ from those of several other declining bat species which prefer trees in mid-decay stages. Tri-colored bats in my study typically used mature live deciduous trees that were further than average from roads, had a greater than average crown depth, and were in a location with higher than average basal area of trees. It is
likely necessary to conserve large parcels of heterogenous forest, with high numbers of mature trees to adequately protect habitat for remnant populations of tri-colored bat that persist on the landscape.
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CHAPTER 1

LITERATURE REVIEW
Introduction

Bat populations are mere remnants of their previous numbers (Alves et al. 2014). Habitat destruction, colony disturbance, and deliberate extermination have historically been some of the largest threats to bats in the United States causing drastic population reductions (Lacki et al. 2007). There are currently numerous devastating threats to tri-colored bat populations including habitat loss due to agricultural and residential development, logging, natural gas development, coal mining, wind energy, and mine closures (Grindal and Brigham 1999; The Center for Biological Diversity and Defenders of Wildlife 2016). The most immediate and largest threat to the tri-colored bat, however, is disease (Government of Canada 2011; Frick et al. 2016). Starting in 2006, white-nose syndrome (WNS) hit North American bat populations with individuals found dying at cave and mine entrances in unprecedented numbers. In just six short years WNS wiped out countless bat colonies and killed more than 5.5 million bats (Coleman 2014). All species known to be affected by WNS in North America feed nearly exclusively on insects, acting as an important biological control agent against insect pests (Kunz and Parsons 2009). With a predicted economic cost of at least four billion dollars a year due to the loss of bats from WNS, the need for effective conservation efforts is dire (Boyles et al. 2011). Despite all of the challenges, remnant populations of some WNS-susceptible bat species are surviving in areas where the main populations were decimated (Frick et al. 2015).

An active area of research exists for quantifying the effects of WNS as it moves across the country, including the conditions it leaves in its wake. Great effort has focused on pathogen growth, spread, and ways to directly combat the effects of WNS on bat
populations (Frick et al. 2010; Reeder and Moore 2013; Hoyt et al. 2014; Langwig et al. 2015). Less research has focused on the needs of species to survive and rebound post-WNS (Baker et al. 2011; Martin 2015). To make constructive decisions, knowledge of the ecology and habitat requirements of susceptible species is critical. Quantification of summer and fall habitat used by most forest-dwelling species, namely their requirements for roosting and foraging, is an active area of research for many imperiled species (Silvis et al. 2014). Many aspects of bat habitat selection are just beginning to be understood and many questions about comprehensive management plans are still unanswered.

The tri-colored bat (*Perimyotis subflavus*) is a species suffering some of the highest mortality from WNS and a species for which we have limited knowledge of its habitat needs (Veilleux et al. 2003). The tri-colored bat has historically been one of the most common and widely-distributed bat species in the eastern United States, yet there are few data on its roosting and foraging requirements (Briggler and Prather 2003; Veilleux et al. 2003; Perry and Thill 2007; O’Keefe et al. 2009). Making constructive management decisions for remnant populations across the country, pre- and post-WNS, is imperative. In order to make these decisions, more information about the habitat requirements of the tri-colored bat is urgently needed.

*Population Decline*

It is difficult to study bats and obtain accurate population estimates due to their life history and behavior. Bats are exceptional among mammals for their capability for true flight. All 45 species inhabiting the United States are part of the Suborder Yangochiroptera, distinguished by their highly sophisticated modifications for echolocation (Foresman 2012). The capability of echolocation and numerous associated
anatomical and physiological adaptations sets yangochiropterans apart from other orders and sheds light onto their critical and unique roles in North American ecosystems as night flying insect consumers and pollinators (Harvey et al. 1999). However, their ability to fly and to use habitat that is difficult for humans to access, in combination with their small size and nocturnal habits, make the majority of bats very difficult to capture. Despite these difficulties, capture, along with population counts during winter hibernation, is necessary to determine bat population trends and demographics, and to set conservation goals.

White-nose syndrome

White-nose syndrome was first discovered in North America in 2006 in Howes Caverns in eastern New York. It has since spread through the United States and Canada (Figure 1). Washington was documented as officially having WNS occurrence in March of 2016. This occurrence on the western coast of the United States represents a significant jump in the disease’s range (Washington Department of Fish and Wildlife 2016), potentially expediting spread of WNS through the western United States. Thirty states are currently confirmed to be affected. In March of 2017, WNS deaths were confirmed for the first time in Nebraska, and the causative agent for WNS was detected on an additional two bat species for the first time in Texas (TPWD 2017; USFWS 2017).

The disease, WNS, is caused by the fungus *Psuedogymnoascus destructans* (*Pd*, formerly *Geomycetes*). The hyphae of *Pd* often grows on the affected bats’ muzzles and gives the bats’ noses a powdery white appearance, which is responsible for the name of the disease (Blehert 2012). Infected bats have a mortality rate of up to 99% and there is no effective practical treatment or method to halt the spread of WNS or fatalities at this
time. Temperatures between 3-15°C and greater than 90% relative humidity are ideal for fungal growth of *Pd* (Hoyt et al. 2014). These environmental conditions are similar to those found in North American bat hibernacula and in bats themselves. Bats often have condensed moisture on their pelage and decreased body temperature reflecting their surroundings during hibernation (Foley et al. 2011). The hibernation preferences of the tri-colored bat for warm cave areas and higher humidity compared with various other North American bat species is thought to increase their contraction of the disease as these reflect ideal conditions for the fungus (Fujita and Kunz 1984; Briggler and Prather 2003; Quinn and Broders 2007; The Center for Biological Diversity and Defenders of Wildlife 2016).

![Map showing the spread of WNS in the Eastern United Stated from 2006-2017. (Source: www.whitenosesyndrome.org/resources/map).](image)

The *Pd* hyphae grow on the exposed skin of bats such as the nose, ear, and wing membranes, causing many physiological changes. The complex physiological effects and
ultimate causes of mortality from WNS are active areas of research. The progression of WNS starts with the infection of the bat by *Pd*, and moves on to invasion of the epidermis, erosion and ulceration of epidermis and dermis, invasion of connective tissue, and disruption of wing functions. The concurrent physiological effects include increased metabolic rate and decreased carbon dioxide excretion through wing membranes, leading to a buildup of carbon dioxide that causes increased arousal from hibernation and hyperventilation, increased water loss, increased electrolyte loss, doubled winter energy use, and often times mortality (Verant et al. 2014).

Recent molecular comparisons support the hypothesis that the source population of the fungus is European or Asian, and that *Pd* was introduced through anthropogenic activities (Leopardi et al. 2015; Hoyt et al. 2016a; b; Zukal et al. 2016). Many hypotheses exist for the difference in the effect of *Pd* on bat populations in the palearctic and North America, as there are no reports of mass mortality in Europe despite extensive population monitoring. These hypotheses include that bat populations in Europe have developed resistance to the fungus. Alternatively, the significantly less dense populations of bats throughout Europe compared with those in North America pre-WNS may be due to the historic occurrence of *Pd* (Frick et al. 2015).

The species currently known to be affected by WNS in North America are the big brown bat (*Eptesicus fuscus*), eastern small-footed bat (*Myotis leibii*), tri-colored bat, little brown bat (*M. lucifugus*), the endangered gray bat (*M. grisescens*) and Indiana bat (*M. sodalis*), as well as the northern long-eared bat (*M. septentrionalis*) which was listed as threatened under the endangered species act May 4th, 2015 (Blehert et al. 2009; USFWS 2015). Other species such as the eastern red bat, southeastern bat (*M.*
austroriparius), silver-haired bat (Lasionycteris noctivagans), Rafinesque's big-eared bat (Corynorhinus rafinesquii), cave bat (M. velifer), Townsend’s big-eared bat (C. townsendii) and the Virginia Big-Eared Bat (C. townsendii virginianus) have tested positive for Pd without showing signs of having WNS (Bernard et al. 2015). All of the species that are infected by WNS, and all but two species known to carry Pd, occur within Kentucky.

Summer Habitat

The tri-colored bat occurs generally in the eastern and midwestern portions of the United States along with areas of eastern Central America and southern Canada (IUCN 2008). The tri-colored bat, like most bat species, relies on a diverse combination of habitat types for roosting and foraging including various types of caves, human structures, forests, clearings, and riparian areas. Tri-colored bats are thought to prefer areas near water and riparian zones, more so than other sympatric bat species (Fujita and Kunz 1984; Owen et al. 2004; Ford et al. 2005; Menzel et al. 2005). As many populations across species are being decimated due to WNS, the need to conserve habitat that remnant populations rely on is heightened. For many species undergoing severe decline due to WNS there is patchy knowledge of their summer roosting and foraging needs, specifically post-WNS. This lack of information is particularly critical for the tri-colored bat.

The summer habitat use of the tri-colored bat is rarely documented, and roost use has only been observed systematically in rather limited portions of its range. To date, summer studies focused on tri-colored bats roosting in buildings (Humphrey et al. 1976; Veilleux 2001). Incidental observations of roosts in human structures and caves made up
some of the earliest summer records for the species, and often became sites for further study (Barbour and Davis 1969; Jones and Suttkus 1973). However the tri-colored bat was also observed in tree roosts, using Spanish moss or foliage on occasion, and were suspected to roost commonly in tree substrates (Findley 1954; Jennings 1958; Davis and Mumford 1962). More recently, some summer telemetry studies have proven that tri-colored bats use tree roosts commonly over the summer period. Thus, the tri-colored bat is currently considered a foliage roosting species of bat (IUCN 2008).

There are roost records for tri-colored bats from telemetry surveys in six U.S. states and one Canadian province. There were 42 individual bats tracked in two studies in Indiana, 28 in Arkansas, 53 in two studies in Nova Scotia, seven in North Carolina, five in two studies in South Carolina, and one bat in Michigan for a total of 137 bats tracked (Krishon et al. 1997; Carter et al. 1999; Kurta et al. 1999; Menzel et al. 1999; Veilleux 2001; Veilleux et al. 2003, 2004; Leput 2004; Veilleux and Veilleux 2004; Poissant et al. 2010; Perry and Thill 2007; Quinn and Broders 2007; O’Keefe et al. 2009; Whitaker Jr et al. 2014). The sites were throughout eastern North America, yet there are still many areas for which there is little to no available information (Figure 2). At this time there are no published records available for Kentucky for summer roosts used by tri-colored bats.

There are also no published comprehensive roost studies on the tri-colored bat in any area after WNS was documented and the susceptible bat populations in that area decreased. Indirect effects of WNS alter community structure and niche partitioning in bat species, thereby affecting the continuing viability of populations (Jachowski et al. 2014). Data gathered on summer habitat use of the tri-colored bat post-WNS is valuable as a starting point to understanding the current resource needs of tri-colored bat populations.
Figure 2. Site of prior summer telemetry studies on *P. subflavus*. Yellow stars denote a study on multiple individuals, blue stars denote a study on one individual, and the blue border denotes the species range (Fujita and Kunz 1984).

Tree Roost Characteristics

Although tri-colored bats are known to roost in human-made structures as well as other non-tree structures during their active season, it appears certain that tree-based day roosts are an integral part of their summer habitat (Veilleux 2001; Perry and Thill 2007; Quinn and Broders 2007; O’Keefe et al. 2009; Poissant et al. 2010; Whitaker Jr et al. 2014). The time spent roosting by bats outweighs the time bats spend on any other activity (Lacki et al. 2007). There are both similarities and differences in summer habitat use across the range of the tri-colored bat. Using the total 137 tri-colored bats (79% female and 21% male) tracked to 313 day roosts, I examined reported information and trends in summer habitat use (Krishon et al. 1997; Carter et al. 1999; Kurta et al. 1999;
Menzel et al. 1999; Veilleux 2001; Leput 2004; Perry and Thill 2007; Quinn and Broders 2007; O’Keefe 2009; Poissant et al. 2010). Tri-colored bats use different materials to hang from or hide within (e.g., leaves or lichen) as roost “substrate” at separate sites where the bats were monitored (Jennings 1958; Krishon et al. 1997; Veilleux and Veilleux 2004; Perry and Thill 2007; Quinn and Broders 2007). Similarities among tri-colored bats’ preferred habitats also appeared across different study sites (Figure 2) in terms of selection for certain species of tree roosts and extent of their movements being generally a few kilometers or less (Perry and Thill 2007; O’Keefe et al. 2009). The majority of the roost substrates documented for tri-colored bats fall under the category of foliage roosts. Analyses of foliage roosting bat studies across many foliage roosting species, including the tri-colored bat, showed a preference by bats for roost trees with a larger diameter at breast height (DBH), greater height, and more closed canopy compared with random trees (Kalcounis-Rüppell et al. 2005). There were many physical variables recorded across several studies (Table 1), and the averages of these quantities begins to form a picture of how the tri-colored bat uses its habitat.
Table 1. Summary of roost use variables measured repeatedly in previous studies

<table>
<thead>
<tr>
<th>Variable</th>
<th>Average</th>
<th>Range</th>
<th>Sources*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Distance moved from capture site to roost (m)</td>
<td>790</td>
<td>300-5000</td>
<td>3, 5, 7</td>
</tr>
<tr>
<td>Days per roost</td>
<td>2.7</td>
<td>0.85-4.31</td>
<td>1, 2, 3, 4, 5, 7, 9, 10</td>
</tr>
<tr>
<td>Roost tree height (m)</td>
<td>19.6</td>
<td>9.5-26.5</td>
<td>1, 3, 4, 5, 7, 8, 9, 10</td>
</tr>
<tr>
<td>Diameter at breast height (cm)</td>
<td>30.1</td>
<td>10.2-42.6</td>
<td>3, 5, 7, 8, 9, 10</td>
</tr>
<tr>
<td>Roost height (m)</td>
<td>13.4</td>
<td>4.9-17.1</td>
<td>1, 2, 3, 4, 5, 7, 8, 9, 10</td>
</tr>
<tr>
<td>Crown closure (%)</td>
<td>54</td>
<td>31-86</td>
<td>1, 2, 3, 4, 7, 8, 9, 10</td>
</tr>
<tr>
<td>Basal Area (m²/ha)</td>
<td>17</td>
<td>1.7-27.2</td>
<td>1, 2, 5, 7, 9</td>
</tr>
<tr>
<td>Distance from roost to nearest overstory tree (m)</td>
<td>5.3</td>
<td>1.5-9.6</td>
<td>3, 4, 5, 6, 9</td>
</tr>
<tr>
<td>Distance from roost to edge (m)</td>
<td>71.5</td>
<td>25-186</td>
<td>3, 4, 5, 6, 9</td>
</tr>
<tr>
<td>Distance to water (m)</td>
<td>126</td>
<td>34-212</td>
<td>3, 4, 5, 10</td>
</tr>
<tr>
<td>Distance to next roost (m)</td>
<td>87.5</td>
<td>19-1500</td>
<td>1, 3</td>
</tr>
<tr>
<td>Roost area with 3+ roosts (ha)</td>
<td>0.53</td>
<td>0.02-1.1</td>
<td>1, 2, 3, 4, 5, 9, 11</td>
</tr>
<tr>
<td>Individuals observed returning to a roost used earlier (%)</td>
<td>35</td>
<td>28-44</td>
<td>3, 5, 9</td>
</tr>
</tbody>
</table>

Differences in roost substrates used throughout the monitored locations within the tri-colored bats’ range may be associated with differences in resource availability or regional environmental conditions (Lacki et al. 2007). Tri-colored bats from the northern portion of the bats’ documented range in Nova Scotia, Canada strongly preferred beard lichen (*Usnea trichodea*) growing in tree canopies for roosting, a lichen which is not reported as a predominant forest species in the majority of the tri-colored bats range (Halonen et al. 1998; Quinn and Broders 2007; Poissant et al. 2010). In the studies done by Quinn and Broders (2007) and Poissant et al. (2010), at least 159 roosts were in beard lichen. In the southern coastal United States, at least 12 roosts were documented in Spanish moss (*Tillandsia usneoides*) hanging in trees (Jennings 1958; Krishon et al. 1997; Menzel et al. 1999). However, it appears that for the majority of the eastern United States, documented in studies from South Carolina to Michigan, tri-colored bats roost directly in live or dead tree foliage (Kurta et al. 1999; Veilleux et al. 2003; Leput 2004; Perry and Thill 2007). Of the studies where foliage was used for roosting and the specific roost substrate was described, 69% were documented as dead leaves or dead leaf clusters while the remaining 31% were living leaves or leaf clusters. The majority of these 143 roosts were in the leaves of deciduous trees; however, at least three of these clusters were in clumps of pine needles that were used as maternity colonies (Perry and Thill 2007).

In the majority of the tri-colored bats’ range where tree foliage was used as the roost substrate, the species of tree had a significant effect on tree use by bats (Veilleux 2001; Perry and Thill 2007; O’Keefe 2009). A wide variety of trees were used by the tri-colored bat for a leaf-based day roost. The species included: box elder (*Acer negundo*).
silver maple (A. saccharinum), sugar maple (A. saccharum), bitternut hickory (Carya cordiformis), black hickory (C. texana), common hackberry (Celtis occidentalis), white ash (Fraxinus americana), American holly (Ilex opaca), black walnut (Juglans nigra), American sweetgum (Liquidambar styraciflua), tulip poplar (Liriodendron tulipifera), Fraser magnolia (Magnolia fraseri), black tupelo (Nyssa sylvatica), American hophornbeam (Ostryra virginiana), shortleaf pine (Pinus echinata), eastern white pine (P. strobus), eastern cottonwood (Populus deltoids), white oak (Quercus alba), laurel oak (Q. laurifolia), bur oak (Q. macrocarpa), swamp chestnut oak (Q. michauxii), pin oak (Q. palustris), chestnut oak (Q. prinus), northern red oak (Q. rubra), post oak (Q. stellata), black oak (Q. velutina), and the American elm (Ulmus americana). Using only roost trees that were determined to species, two species of Quercus made up more than a third of roosts, with the white oak comprising 20.5% and the northern red oak making up 17.9% of all roosts. Box elder, tulip poplar, and black oak each comprised 5-7% of all roosts with all other species making up less than 5% of the total. Looking at the genera used for leaf roosting by tri-colored bats (Table 2), only Quercus, Acer, and Carya make up more than 5% of the documented roost tree species.
Table 2. Genera of trees in which *P. subflavus* used leaf roosts, with two or more records

<table>
<thead>
<tr>
<th>Tree Genera</th>
<th>Percentage of genera (%)</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Quercus</td>
<td>53.7</td>
<td>65*</td>
</tr>
<tr>
<td>Acer</td>
<td>18.2</td>
<td>22*</td>
</tr>
<tr>
<td>Carya</td>
<td>7.4</td>
<td>9*</td>
</tr>
<tr>
<td>Populus</td>
<td>5.0</td>
<td>6*</td>
</tr>
<tr>
<td>Liriodendron</td>
<td>5.0</td>
<td>6</td>
</tr>
<tr>
<td>Ulmus</td>
<td>3.3</td>
<td>4*</td>
</tr>
<tr>
<td>Juglans</td>
<td>2.5</td>
<td>3*</td>
</tr>
<tr>
<td>Liquidambar</td>
<td>2.5</td>
<td>3</td>
</tr>
<tr>
<td>Pinus</td>
<td>2.5</td>
<td>3</td>
</tr>
</tbody>
</table>

*Denotes minimum count when exact counts were not given for a genera or species within genera by the author(s).

In several of the studies on roost choices, a preference for one or more habitat resources was reported. These included a preference for upland and riparian areas for female tri-colored bats in Indiana, as well as a preference for *Quercus* species by reproductive tri-colored females and a preference for *Acer* species by non-reproductive females (Veilleux 2001; Veilleux et al. 2004). In Arkansas, tri-colored females preferred larger DBH roost trees compared with random trees, and roosted higher from the ground compared with males. Males preferred mature (more than 50 years old) roost trees and trees in unharvested stands (Perry and Thill 2007). In North Carolina tri-colored bats used roosts that were closer than random to non-linear openings, closer to edges, at lower
elevations, closer to intermittent streams, and in trees taller than the nearest tree (O’Keefe 2009).

**Study Site**

The 69,000 ha Land Between the Lakes National Recreation Area (LBL), is a human-made peninsula roughly 65 km north to south and 12-16 km east to west with large expanses of forested lands. The forests are comprised of different stand ages that contain many riparian areas and linear corridors (Franklin et al. 1993; Schulte 2012). The forests are predominantly hardwood but include some pine plantings. The impoundment of the Tennessee River into “Kentucky Lake” and the impoundment of the Cumberland River into “Lake Barkley” formed LBL. The peninsula is located in Lyon and Trigg counties in Kentucky and Stewart county in Tennessee. The area was under the management of the Tennessee Valley Authority from 1964 to 1999, and since 1999 has been managed by the United States Department of Agriculture Forest Service (Fleming et al. 2002). Land Between the Lakes is a valuable resource supporting a diverse assemblage of bat species in the Western highland rim ecoregion of Kentucky/Tennessee and is included in the Western Mesophytic Forest Section of the Eastern Deciduous Forest Biome (Fralish 2002). The vegetation communities within LBL include prairie, oak-hickory forest, swamp forest, and mixed mesophytic (Fralish 2002). The major tree species include white oak, black oak, southern red oak (*Q. falcata*), scarlet oak (*Q. coccinea*), blackjack oak (*Q. marilandica*), post oak, sugar maple, and American beech (*Fagus grandifolia*; Fralish 2002). Of remnant bottomland hardwood areas studied within LBL by Fralish (2002), dominant overstory was composed of American sweetgum, cherrybark oak (*Q. pagoda*), black tupelo, red maple, sugar maple, shagbark hickory
(Carya ovata), tulip poplar, and winged elm (Ulmus alata). Although large areas of LBL were stripped of forest when land was used by the iron industry (1843-1912), today over 80% of LBL is forested (Smith 2015). Most bat species known to occur in the southeastern United States were found within LBL during pre-WNS bat surveys. After initial habitat surveys in 1992, summer mist-net and/or acoustic surveys were conducted at selected sites within LBL at roughly five-year intervals with surveys occurring seven times between 1993-2010 (Gardner 1992; Moyer et al. 1993; Rebar and Hendricks 1994; Harvey and Britzke 2000; Palmer Engineering 2003; Derting 2011). The tri-colored bat was captured at 69% of sites surveyed pre-WNS, with an average of one tri-colored bat captured per 1.3 net-nights between 1995 and 2005 (Figure 3). An average of 35 tri-colored bats were caught during each of these summers. Historical records from bat surveys (1993-2008), in conjunction with the abundance of bat species present makes LBL an ideal area for studying habitat use by bat species impacted by WNS.

![Figure 3. Tri-colored bat captures during bat surveys in LBL pre-WNS occurrence.](image-url)
Research Objectives

My objectives were to identify key features of roosting habitat that need to be conserved in order to facilitate the survival of remnant tri-colored bat populations post-WNS. I characterized roosting habitat used, and provided new knowledge by gathering descriptive data for bat numbers, movements, and habits. I tested three hypotheses that stemmed from my research questions.

Research Questions: Do roost trees used by tri-colored bats differ from a random sample of trees? Do roost trees used by different reproductive classes of tri-colored bat differ from each other? Does the habitat used by the tri-colored bat differ from the habitat protected for the endangered Indiana bat or the listed northern long-eared bat?

\( H_0 \): The roost trees and sites used by tri-colored bats do not differ significantly from a random sample of trees.

\( H_a \): The roost trees used by tri-colored bats differ significantly from a random sample of trees as determined by measured tree and site variables.

\( H_{0r} \): The roost trees and sites used by male, pregnant or lactating female, and non-reproductive female tri-colored bats do not differ significantly from each other.

\( H_{ar} \): The roost trees and sites used by male, pregnant or lactating female, and non-reproductive female tri-colored bats do differ significantly from each other.
$H_{0n}$: The roost trees and sites used by tri-colored bats do not differ significantly from those protected under the federal guidelines for the endangered Indiana bat or the federal listing for the northern long-eared bat (USFWS 2014, 2015).

$H_{an}$: The roost trees and sites used by tri-colored bats differ significantly from those protected for under the federal guidelines for the endangered Indiana bat or the federal listing for the northern long-eared bat (USFWS 2014, 2015).
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CHAPTER 2
MANUSCRIPT
Roost selection of tri-colored bats post-white nose syndrome in western Kentucky and Tennessee

Written by Katherine Schaefer

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Roost selection of tri-colored bats post-white nose syndrome in western Kentucky and Tennessee

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The tri-colored bat (Perimyotis subflavus) has declined severely across its range since 2006 due to white nose syndrome. My objective was to determine distinguishing characteristics of roost trees and habitat used by tri-colored bats so that the species’ needs can be considered in management plans. I mist-netted for tri-colored bats in western Kentucky and Tennessee during the summers of 2015 and 2016, and attached a radio transmitter to each captured adult bat. I tracked each of 15 bats to its day roost for 1-12 days and collected habitat data at 38 roost areas and at 74 randomly-selected trees in the area of the capture of each bat for comparison. Tri-colored bats used roost trees within relatively small geographic areas. The greatest distance moved between successive roosts by a bat was 482 m; average distance between roosts was 86 m. Bats remained within 2.5 km of their original capture site. Tri-colored bats’ roost tree selection was nonrandom. All roosting bats were located in the foliage of live trees. Bats were observed roosting in nine different species of tree, with the most commonly-selected species being Carya tomentosa and Quercus alba (46% and 23% of roost trees, respectively). The most abundant species among the randomly-selected trees were Q. alba, which was selected as a roost roughly in proportion to its abundance, and Acer saccharum which was never selected as a roost tree species. A generalized linear model showed that increasing tree crown depth, basal area, and distance from roads were correlated with roost tree
selection. Management needs of tri-colored bats differ from those of other protected bat species, which prefer trees in mid-decay stages. Tri-colored bats in this study typically used mature live deciduous trees that were further than average from roads, had a greater than average crown depth, and occurred in a location with higher than average basal area. Understanding the habitat selection of tri-colored bats is important to aid in minimizing their population decline. It is likely necessary to conserve large parcels of heterogenous forest, with high numbers of mature trees, to adequately protect habitat for the remnant populations of tri-colored bat that persist on the landscape.

Key words: bat, *Carya*, Chiroptera, Kentucky, *Perimyotis*, roost, tri-colored, white-nose syndrome, Tennessee, habitat

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The tri-colored bat is a species which has been frequently documented and was once considered common across much of its range in eastern North America, from southern Canada to Central America (Davis and Mumford 1962; Fujita and Kunz 1984). However, research and population monitoring on tri-colored bats has been infrequently performed. Keeping track of the total population of tri-colored bats is difficult for many reasons. The regular monitoring of hibernating populations of wintering bats by State and Federal resource agencies includes tri-colored bats, however surveys were primarily focused on species listed at the federal or state level as endangered, threatened, or species of concern. Rarely were more than a subset of tri-colored bats present within a site counted due to the commonness of the species, as well as the dispersed roosting behavior of the species within the hibernacula (Armstrong 2017). Therefore, pre-white nose syndrome (WNS) estimates are rarely accurate enough to be useful as a regional baseline (Armstrong 2017). The extensive range of the species makes population documentation across regions difficult. The true range of the species is not well-defined, and is potentially shifting or incomplete. There are now regular but infrequent records occurring in northern areas where tri-colored bats were originally thought to be absent as well as possible expansion in western regions (The Center for Biological Diversity and Defenders of Wildlife 2016). The comprehensive populations trends, therefore, are not definitively known. Across the northeastern United States, however, the population decline since the documentation of WNS has been significant (Langwig et al. 2012; NatureServe 2015).

There are many features of the tri-colored bats’ natural history thought to make the species highly vulnerable to WNS, shown by high mortality rates reported for WNS
positive hibernacula (Frick et al. 2015; Powers et al. 2015). Tri-colored bats seem to have one of the three highest mortality rates from WNS in bat species, along with the little brown bat (*Myotis lucifugus*) and northern long-eared bat (*M. septentrionalis*; Glaeser et al. 2016). Glaeser et al. 2016). In four eastern states tri-colored bat population trajectories were modeled based on winter hibernacula counts, with population declines observed in pre-WNS counts (Ingersoll et al. 2013). Early sensitivity to WNS or other contributing threats to survival of the tri-colored bat were indicated to be of concern at the population level (Ingersoll et al. 2013, 2016). Hibernacula counts in states such as Maryland, West Virginia, Pennsylvania, Ohio, and Virginia indicate population declines from 77-99% (Herzog and Reynolds 2013; Feller and Nagel 2015; The Center for Biological Diversity and Defenders of Wildlife 2016). Several states have added the tri-colored bat to the state threatened or endangered list in recent years with 14 states listing the species as at least state vulnerable and five states describing it as either unrankable or not ranked (NatureServe 2015). It is too early to fully know the effect of WNS on tri-colored bat populations, however, one study indicated that the tri-colored bat stabilized at a decreased population post-WNS, while a separate study indicated that the time until greatest mortality occurred up to two years later than in other heavily affected species (Langwig et al. 2012; The Center for Biological Diversity and Defenders of Wildlife 2016). Considering the multi-faceted decline and uncertainty about long-term population trends, the mortality suffered by tri-colored bats due to WNS is potentially devastating (Government of Canada 2011; The Center for Biological Diversity and Defenders of Wildlife 2016).
Information on the ecology of declining and understudied bat species is necessary for effective conservation. Without understanding the resources required for individuals to survive and maintain adequate body condition in absence of a specific challenge, it is difficult to aid populations facing extreme challenges like WNS. As with many cave hibernating bats, there is information available about the winter needs of the tri-colored bat (Harvey et al. 2011). Surprisingly little is known, however, about its summer day roost needs. Research such as that done by Veilleux (2003) in Indiana and Perry in Arkansas (2007) provided important insights on the tri-colored bat’s habitat through radio-telemetry summer roost studies in eastern deciduous forest habitats. Roost selection varied by location for tri-colored bats. For example, tri-colored bats roosted among dead deciduous leaf clusters, pine needles, or lichens in different proportions in distinct sites within their range (Veilleux et al. 2003; Perry and Thill 2007; Poissant et al. 2010). Thus, as populations of tri-colored bats shrink due to WNS mortality, understanding more about habitat resource use in the altered post-WNS ecosystem is needed for habitat conservation efforts.

Land Between the Lakes National Recreation Area (LBL), is a human-made peninsula with large expanses of forested lands. The resources at LBL support a diverse assemblage of bat species in the Western highland rim ecoregion of Kentucky/Tennessee (Fralish 2002). Most bat species known to occur in the southeastern United States were found within LBL during pre-WNS bat surveys. The tri-colored bat was captured at 69% of sites surveyed pre-WNS, with an average of one tri-colored bat captured per 1.3 net-nights in four surveys conducted between 1995 and 2005. An average of 35 tri-colored bats was caught during each of these summer surveys. White-nose syndrome was first
confirmed in Kentucky in 2011 in Trigg county, one of the three counties containing LBL (Carr 2011). There appear to be notable effects to the bat populations in the years post-WNS within LBL. For example, pre-WNS the northern long-eared bat (*Myotis septentrionalis*) accounted for 52 out of 300 bat captures during summer mist-net captures in LBL (1993-2008). In surveys four and five years post-WNS (2015-2016), northern long-eared bats accounted for only one of 285 captures, thus highlighting the aftermath of the disease.

I examined key features of the roosting habitat of the tri-colored bat in the diverse oak-hickory landscape within LBL. My goal was to characterize the post-WNS day roost sites and movements and to identify resources that need to be conserved in order to facilitate the survival of remnant tri-colored bat populations post-WNS in western Kentucky and Tennessee. My specific objectives were to determine resources selected for by the tri-colored bat at the roost tree and site level, and to describe the characteristics of the roosts that they occupied and their daily movements. I also examined differences in habitat occupied by the tri-colored bat and habitat currently protected for the endangered Indiana bat and the threatened northern long-eared bat.

**MATERIALS AND METHODS**

*Study area.—* The study was conducted in the 69,000 ha Land Between the Lakes National Recreation Area (LBL). The forest communities within LBL include oak-hickory, swamp, and mixed mesophytic forest of different stand ages that contain many riparian areas and linear corridors (Fralish 2002; Schulte 2012). The peninsula is located in Lyon and Trigg counties in Kentucky and Stewart county in Tennessee. The major tree species include white oak (*Quercus alba*), black oak (*Q. velutina*), southern red oak
(Q. falcata), scarlet oak (Q. coccinea), blackjack oak (Q. marilandica), post oak (Q. stellata), sugar maple (Acer saccharum), and American beech (Fagus grandifolia (Fralish 2002). Of the remnant bottomland hardwood areas studied within LBL by Fralish (2002), dominant overstory was composed of sweetgum (Liquidambar styraciflua), cherrybark oak (Q. pagoda), blackgum (Nyssa sylvatica), red maple (A. rubrum), sugar maple, shagbark hickory (Carya ovata), tulip poplar (Liriodendron tulipifera), and winged elm (Ulmus alata). Although large areas of LBL were stripped of forest when land was used by the iron industry (1843-1912), today roughly 89% of LBL is forested with only 7% being open lands and 4% being infrastructure such as roads and right-of-way’s (USFS 2004). Mean minimum and maximum daily temperatures from May 1st to August 31st were 18.5°C and 28.2°C in 2015 and 18.9°C and 28.9°C in 2016. Precipitation during May through August was 42.2 cm and 68.5 cm in 2015 and 2016, respectively. Temperature and precipitation estimates were remotely sensed near Golden Pond, KY using an Ambient Weather WS-2090 centrally located within my study area by a quality controlled personal weather station owner. 

Bat capture, handling, and marking.—During May through August 2015 and 2016, I followed standard protocols for bat mist-netting and USFWS Indiana bat survey guidelines (Kunz and Parsons 2009; USFWS 2017). Each mist net set-up consisted of at least two 6-12 m wide by 2.6 m tall nets (Avinet, Inc., 75 dernier/2-ply, 38mm mesh, 4 shelves) stacked vertically on fixed extension poles. Nets were placed before dusk in areas that likely served as bat travel or foraging corridors such as streams and trails, when temperatures were greater than 10°C, there was no sustained precipitation (>30 min bouts or continuously intermittent), and sustained wind speeds were less than 4 m/sec. Each
net was checked every 10 minutes and light and sound disturbances around the sites were minimized. For each captured bat I measured species, sex, age (adult/juvenile), body mass (g), forearm length (mm), wing-damage index for bats affected by WNS (Reichard 2008), and reproductive status. I attached a Kentucky Department of Fish and Wildlife Resources lipped aluminum alloy wing band to each captured bat using a 2.9 mm band for most species but the smaller 2.4 mm bands for the tri-colored bat. Age-class was determined as juvenile or adult by the degree of epiphyseal-diaphyseal ossification (Wilkinson 2009). Reproductive condition was determined by examining the abdomen and mammmae for evidence of embryos or lactation in females or descent of testes in males (Kunz and Parsons 2009).

During the mist-netting surveys, a radio transmitter (0.27 g, Holohil Systems Ltd., LB-2X) was attached to each captured adult tri-colored bat for whom the transmitter mass was no more than 5% of the individual’s total body weight. A small amount of hair was trimmed on the dorsal surface between the scapulae and the transmitter was attached using skin bonding latex adhesive (Osto-Bond, Montreal Ostomy Products; Carter et al. 2009). All capture, handling, banding, and marking of the bats followed the guidelines of the American Society of Mammalogists (Sikes et al. 2016) and were approved by Murray State University’s Animal Care and Use Committee (Project Protocol No. 2015-028).

**Roost-site documentation.**— I tracked bats to their day roosts until the transmitter dropped or the battery died (~12 days). I used a telemetry receiver (TRX 1000S, Wildlife Materials Inc.) with a 5-element car-mounted Yagi directional antenna to initially locate the individual. Once the general location of a tagged bat was determined, individuals were tracked by foot to a roost using a 3-element folding Yagi directional antenna and the
roost site was located (Wilson et al. 1996). Roost site locations were recorded using a handheld GPS unit (Garmin GPSMAP® 62sc, ± 3 m). The roost was initially pinpointed as closely as possible using a 6 m coaxial cable without an antenna attached, that was raised on an extendable pole, and surveying leaf clusters within the presumed roost tree and any adjacent trees with overlapping canopy. Tree branches and leaf clusters were examined by binocular (10x or greater) for either a visible bat or transmitter antenna (Wilson et al. 1996; Perry and Thill 2007).

Frequently, I was unable to see a bat or its antennae at the presumed roost tree. Therefore, I used emergence counts and observations to confirm the roost location to a tree and usually a specific leaf cluster. I conducted an evening emergence count every night when a potential roost location was known, with priority given to roosts not yet observed. I started observations for an emergence count one-half hour before sunset and continued until one hour after sunset or longer if bats were still observed to be emerging. Surveyors positioned themselves so that bats emerging were silhouetted against the sky, near to but not directly under the roost. Surveyors were equipped with telemetry equipment to verify when the tagged individual emerged. Data collected during emergence counts included number of individuals sighted, time of emergence, and location of emergence on the tree (Kunz and Parsons 2009; USFWS 2016).

**Habitat measurements.**—For each roost tree used by a bat I established two randomly-selected trees for comparative measurements. These two randomly-selected trees were located within a circular buffer centered around the point of capture and release for each bat. The radius of the buffer was equal to the distance that the bat moved from its capture point to its initial roost location (Kalcounis-Rüppell et al. 2005; O’Keefe
and Loeb 2017). The randomized points used to locate the randomly-selected trees were formed with the Create Random Points tool in ArcGIS 10.3.1 (Environmental Systems Research Institute, Redlands, California). Any point that fell within unusable habitat (specifically water, infrastructure, or open lands) was discarded and replaced. The randomly-selected tree itself was determined by navigating to the point and locating the nearest overstory tree to the exact location, with overstory trees defined as being ≥ 10 cm diameter at breast height (DBH).

I measured habitat variables on the roost and randomly-selected trees, and in a 0.04 ha plot centered on each tree. For the roost or randomly selected tree I took a GPS location and determined species, height (m), DBH (cm), crown depth (m), decay class (0-5; USFWS 2016), slope position (bottomland, ridge, or mid-slope), aspect of slope (°), crown exposure of tree (none, some, or all), and canopy cover (%) at the four cardinal directions. For roost trees, I additionally recorded the roost height (m), roost aspect (°), canopy closure 2 m above the roost (%), and roost composition (i.e., live leaves or dead leaf cluster). Within each 0.04 ha plot I recorded an understory tree count (number < 10 cm DBH), understory vegetation density (low, medium, high), height of tallest understory tree (m), dominant vegetation species, basal area of plot, percentage of plot within 5 m radius of roost occupied by shrubs (%), height of nearest overstory tree to roost (m), and average DBH of overstory trees in plot (cm). For every tree in a plot with a DBH ≥ 10 cm, species, DBH, and decay class was recorded.

I also documented macrohabitat variables that were of potential importance in roost tree selection using ArcMap 10.3.1. The variables used were distance from tree to
the closest water feature, distance to nearest road, and elevation of the tree. Distance to other features were determined using the Near tool in ArcMap 10.3.1. The water feature layers used were from the National Hydrography Dataset surface water drainage system maps at 1:24,000 scale, the features contained in this layer are designated as a stream, river, or lake. The roads layers used were the Kentucky Transportation Cabinet road centerline map and the Land Between the Lakes Motorized Vehicle Use Map layers. Elevation values were extracted from a 10-m DEM (Digital Elevation Model) of the region to each randomly-selected tree using the Extract Values to Points tool. For variables extractable through GIS (distance to the closest water feature, distance to the nearest road, aspect, and elevation), an additional set of randomized comparison points was used to further examine differences between roost trees known to be used and landscape level habitat. A larger circular buffer surrounding the areas used by the bats was created. This buffer was equal to the longest recorded flight by any bat within the study period (5 km), and was the area in which the randomized points were formed with the Create Random Points tool in ArcMap.
Table 3.— Characteristics of roost sites of tri-colored bats and randomly-selected tree
sites in Land Between the Lakes National Recreation Area, KY and TN, during summer

<table>
<thead>
<tr>
<th>Category</th>
<th>Parameter</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tree</td>
<td>HGT</td>
<td>Tree height (m)</td>
</tr>
<tr>
<td></td>
<td>DPTH</td>
<td>Tree crown depth (m)</td>
</tr>
<tr>
<td></td>
<td>TDBH</td>
<td>Tree DBH (cm)</td>
</tr>
<tr>
<td></td>
<td>NCAN</td>
<td>North canopy cover (%)</td>
</tr>
<tr>
<td></td>
<td>ECAN</td>
<td>East canopy cover (%)</td>
</tr>
<tr>
<td></td>
<td>SCAN</td>
<td>South canopy cover (%)</td>
</tr>
<tr>
<td></td>
<td>WCAN</td>
<td>West canopy cover (%)</td>
</tr>
<tr>
<td></td>
<td>EXPO</td>
<td>Exposure potential of tree crown (none, some, all)</td>
</tr>
<tr>
<td>Plot (0.04 ha)</td>
<td>BA</td>
<td>Basal area (m2/ha)</td>
</tr>
<tr>
<td></td>
<td>PDBH</td>
<td>Average plot DBH (cm)</td>
</tr>
<tr>
<td></td>
<td>HGTU</td>
<td>Height of tallest understory (m)</td>
</tr>
<tr>
<td></td>
<td>DOV</td>
<td>Distance to nearest overstory tree (m)</td>
</tr>
<tr>
<td></td>
<td>SRB</td>
<td>Shrub cover 5 m radius of center (%)</td>
</tr>
<tr>
<td></td>
<td>ELEV</td>
<td>Elevation (m)</td>
</tr>
<tr>
<td></td>
<td>DROAD</td>
<td>Distance to nearest road (m)</td>
</tr>
<tr>
<td></td>
<td>DWATER</td>
<td>Distance to nearest perennial water source (m)</td>
</tr>
<tr>
<td></td>
<td>SLPO</td>
<td>Slope position (bottomland, mid-slope, or ridge)</td>
</tr>
<tr>
<td></td>
<td>UDEN</td>
<td>Understory vegetation density (low, medium, or high)</td>
</tr>
<tr>
<td></td>
<td>UNDC</td>
<td>Count of understory stems (&lt;10 cm DBH)</td>
</tr>
</tbody>
</table>
Nightly movements between successive roost trees were characterized by calculating the distance between roost trees using the Near function in ArcGIS. I also created a minimum convex polygon of the roosting range for each bat that had three or more roost locations using the Minimum Bounding Geometry tool in ArcGIS, with the convex hull shape selected.

Data analysis and modeling.— All tests were completed using R software (R Core Team 2016). There was no significant effect of year as a response when used in a generalized linear model of all variables. Therefore, I pooled my data from 2015 and 2016 to compare the roost trees with unused randomly-selected trees. There were no significant differences between sexes and reproductive classes using a one-way multivariate analysis of variance (MANOVA) with all variables, an alpha level of 0.05 was used. Therefore, I pooled my data for all individuals due to low sample size. Because my data contained multiple observations for most individual bats, not all data points were independent of each other. Dependence was tested for using a one-way analysis of variance (ANOVA) as a function of bat individual (Snider et al. 2013). Dependence was not significant for any individual.

Numerical habitat variables were examined for significant differences in means between trees used for roosting and randomly-selected trees. Symmetry of numeric data was considered by examining the spread of data on box-plots and normality was examined using the Shapiro-Wilk hypothesis test. If data were non-normal then the data were transformed by either logarithmic or square root transformations, and retested for normality. Since normality or approximate normality was achieved, means were then
compared using a one-way ANOVA. Circular variables (aspects in degrees) were tested for directional trends using Rao’s spacing test of uniformity. A chi-squared test or Fisher’s Exact test was used to determine differences between trees used as roosts and randomly-selected trees. To test differences between plots centered on trees used for roosting and plots centered on unused randomly-selected trees a Welch’s ANOVA test for unequal variance was used for numerical variables with a significance level of 0.05.

For data at the tree and plot levels I used a generalized linear model, logistic regression of occupancy (roost tree occupied by a tri-colored bat versus randomly-selected tree assumed not occupied by a tri-colored bat) with binomial variance and a logit link to determine the variables that were associated significantly with tri-colored bat summer roost occupancy (Table 3). I also used a generalized linear model of occupancy for the large scale plots within a 5 km buffer of roosts to determine macrohabitat features that were associated significantly with bat summer roosting areas. I based model selection on the lowest AICc. I reported generalized linear models with a small (< 4 units) difference from AIC$_{\text{min}}$ ($\Delta_i$) using Akaike information criterion modified for small samples (AIC$_c$). Along with the AIC$_c$ value, $\Delta_i$ value, model weights ($w_i$) and generalized $R^2$ are reported. All data are reported as mean ± 1 S.E.

**RESULTS**

Across the summers of 2015 and 2016, I captured 21 tri-colored bats during 216 net-nights, 65% of which were female and 35% were male. Of the 21 tri-colored bats captured, 15 were of sufficient weight to carry a radio transmitter (transmitter weight <5% body weight). The tri-colored bat accounted for 4.2% of all bat captures. Six of the radio-tagged females were pregnant, two were lactating, and one was post-lactating.
Among the six radio-tagged males, two had testes descended. I tracked these 15 bats to 38 confirmed roost sites with 22 confirmed leaf roosts documented within the trees, and probable leaf roosts documented for the 16 remaining roost sites. Three roosts were considered to be maternity colonies as $\geq 2$ adult bats exited a leaf cluster occupied by pregnant or lactating females (Perry and Thill 2007).

**Roost tree characteristics.**— All confirmed leaf roosts were located in live deciduous trees (Table 4). Most of the females (62%) and males (70%) roosted in dead leaf clusters hanging in the live trees, with the remaining bats roosting in live leaf clusters. All roosts were within the top third of their respective roost tree, with the average roosting height ($20.4 \pm 2.1$ m) being 2.2 m less than the average roost tree height ($22.6 \pm 2.0$ m). The aspects of the slopes roosts were located on were not significantly different from a uniform distribution across aspects (Rao’s test statistic = 148, critical value = 158). The average percent canopy closure two m above the roosts ($89.9 \pm 1.6 \%$) was similar to the overall average percent canopy closure ($88.9 \pm 0.7 \%$) measured.
Table 4.— Roosting habitat variables of tri-colored bats with comparisons between roost trees and unused randomly-selected trees in Land Between the Lakes National Recreation Area, KY and TN, during summer 2015–2016.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Abbreviation</th>
<th>Day roosts (n = 22)</th>
<th></th>
<th></th>
<th></th>
<th>Randomly-selected trees (n = 76)</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>$X$</td>
<td>SE</td>
<td>95% CI</td>
<td></td>
<td>$X$</td>
<td>SE</td>
<td>95% CI</td>
<td>F</td>
</tr>
<tr>
<td><strong>Tree</strong></td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tree height (m)</td>
<td>HGT</td>
<td>22.6</td>
<td>2.0</td>
<td>20.7–24.6</td>
<td></td>
<td>19.9</td>
<td>1.1</td>
<td>18.7–21</td>
<td>2.10</td>
</tr>
<tr>
<td>Crown depth (m)</td>
<td>DPTH</td>
<td>16.8</td>
<td>1.6</td>
<td>15.2–18.4</td>
<td></td>
<td>13.1</td>
<td>0.8</td>
<td>12.2–13.9</td>
<td>6.06</td>
</tr>
<tr>
<td>Tree DBH (cm)</td>
<td>TDBH</td>
<td>34.9</td>
<td>3.7</td>
<td>31.2–38.6</td>
<td></td>
<td>31.2</td>
<td>2.0</td>
<td>29.2–33.2</td>
<td>1.07</td>
</tr>
<tr>
<td>North canopy cover (%)</td>
<td>NCAN</td>
<td>88.1</td>
<td>1.8</td>
<td>86.3–90</td>
<td></td>
<td>88.7</td>
<td>0.8</td>
<td>87.8–89.4</td>
<td>0.10</td>
</tr>
<tr>
<td>East canopy cover (%)</td>
<td>ECAN</td>
<td>88.9</td>
<td>1.3</td>
<td>87.5–90.2</td>
<td></td>
<td>88.7</td>
<td>0.7</td>
<td>87.9–89.3</td>
<td>0.27</td>
</tr>
<tr>
<td>South canopy cover (%)</td>
<td>SCAN</td>
<td>88.6</td>
<td>1.4</td>
<td>87.2–90.0</td>
<td></td>
<td>90.3</td>
<td>0.7</td>
<td>89.6–91.2</td>
<td>2.02</td>
</tr>
<tr>
<td>West canopy cover (%)</td>
<td>WCAN</td>
<td>88.2</td>
<td>1.5</td>
<td>86.7–89.7</td>
<td></td>
<td>89.9</td>
<td>0.5</td>
<td>89.3–90.4</td>
<td>1.90</td>
</tr>
<tr>
<td><strong>Plot</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basal area (m$^2$/ha)</td>
<td>BA</td>
<td>8.0</td>
<td>0.6</td>
<td>7.4–8.5</td>
<td></td>
<td>6.1</td>
<td>0.2</td>
<td>5.9–6.3</td>
<td>8.99</td>
</tr>
</tbody>
</table>

*Denotes significance at α = 0.05
<table>
<thead>
<tr>
<th>Measure</th>
<th>Mean</th>
<th>SD</th>
<th>Range</th>
<th>Mean</th>
<th>SD</th>
<th>Range</th>
<th>Mean</th>
<th>SD</th>
<th>Range</th>
<th>n</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Average plot DBH (cm)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PDBH</td>
<td>26.7</td>
<td>1.3</td>
<td>25.4–28</td>
<td>27.7</td>
<td>1.1</td>
<td>26.5–28.8</td>
<td>0.34</td>
<td>0.56</td>
<td>1, 86</td>
<td></td>
<td></td>
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<tr>
<td>Height of tallest understory (m)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HGTU</td>
<td>10.5</td>
<td>0.7</td>
<td>9.8–11.2</td>
<td>11.2</td>
<td>0.6</td>
<td>10.5–11.7</td>
<td>0.53</td>
<td>0.47</td>
<td>1, 82</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Distance to nearest overstory tree (m)</td>
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<td></td>
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<td></td>
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</tr>
<tr>
<td>DOV</td>
<td>6.6</td>
<td>0.5</td>
<td>6.1–7.2</td>
<td>6.1</td>
<td>0.4</td>
<td>5.7–6.5</td>
<td>0.26</td>
<td>0.61</td>
<td>1, 32</td>
<td></td>
<td></td>
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<tr>
<td>Shrub cover 5 m radius (%)</td>
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<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>SRB</td>
<td>38.1</td>
<td>4.5</td>
<td>33.6–42.6</td>
<td>45.1</td>
<td>3.2</td>
<td>41.9–48.2</td>
<td>1.58</td>
<td>0.21</td>
<td>1, 71</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Elevation (m)</td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ELEV</td>
<td>131.3</td>
<td>3.0</td>
<td>128.2–134.3</td>
<td>126.8</td>
<td>1.9</td>
<td>124.9–128.7</td>
<td>1.55</td>
<td>0.21</td>
<td>1, 64</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Distance to nearest road (m)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DROAD</td>
<td>380.1</td>
<td>32.1</td>
<td>348–412.2</td>
<td>257.5</td>
<td>30.5</td>
<td>226.9–287.9</td>
<td>7.71</td>
<td>0.007*</td>
<td>1, 93</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Distance to nearest perennial water source (m)</td>
<td>107.3</td>
<td>16.1</td>
<td>91.2–123.4</td>
<td>116.1</td>
<td>11.8</td>
<td>104.3–127.8</td>
<td>0.19</td>
<td>0.662</td>
<td>1, 74</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Asterisks denote statistical significance (Welch’s ANOVA test, P = 0.05*
Roosts were located in nine different tree species. Only two tree species, white oak and mockernut hickory, had greater than one documented roost. The roost tree species used were white oak (5 roosts), mockernut hickory (10 roosts), and one roost each in black tupelo, Southern hackberry (*Celtis laevigata*), American elm (*Ulmus americana*), post oak, tulip tree, Northern red oak (*Quercus rubra*), and black oak. The composition of tree genera (trees ≥ 5 cm DBH) in the 0.04 ha plots centered on randomly-selected trees was 31% oak, 17% hickory, 14% maple, 8% liquidambar, 6% elm, 2% pine, and < 5% of any of 16 other recorded hardwood genera. However, over half of the random trees (51%) consisted of four individual species; specifically, white oak (23%), mockernut hickory (10%), sugar maple (10%), and American sweetgum (8%).

The roost tree species used by tri-colored bats differed significantly from what was expected based on random tree species occurrence (Fisher’s Exact test, $P = 0.02$). Bats roosted more frequently than expected in mockernut hickory trees, less frequently than expected in maple species, and did not differ significantly from expected in other tree species including white oak (Figure 4). Although there was much variation, a typical roost occurred in a mockernut hickory of tree average height (22.9 ± 2.1 m) with a relatively deep crown (17.7 ± 1.4 m), in a locale with high basal area (9.2 ± 1.4 m²/ha), and several hundred meters (476 ± 43 m) from the nearest road. The bats typically roosted within a dead leaf cluster about 2 m (+/- 1.2 m) below the tree top.
I determined the best fit generalized linear model that explained the difference between roost sites and random sites for tri-colored bats (Table 5). Three models had similar low AICc values and each included basal area, crown depth, and distance to nearest road as model parameters. The best fit model, as determined by the lowest AICc value, also contained slope position and understory vegetation density as significant variables. The probability that it was the best of the models considered for the data collected was 56% (Table 5). The odds ratio indicated that when compared to random sites, bats were more likely (Table 6) to roost in trees with a greater than average crown

Figure 4. Proportions of tri-colored bat roost tree species used, and proportions of common (>2% of total) random tree species surveyed.
depth that occurred at sites with a higher than average basal area and that were further than average away from road openings, preferring a moderate level of understory vegetation and bottomland locations compared with relative hill topography. The odds are 20% higher that a tree is used as a roost for every 10 m²/ha increase in basal area, and over seven times more likely to be used if the surrounding understory vegetation was moderate in density. For every 50 m distance from a road, the odds were 13% higher that a tree was used as a roost, and for every 10 m increase in crown depth, the odds were 86% higher that a tree was used as a roost.

Table 5.—Model parameters that explained the difference between roost tree sites and random tree sites for tri-colored bats in LBL, KY and TN, USA, 2015-2016. Along with the AICc value, difference from AICmin (Δi), model weights (wi) and generalized R² is given for each model. Model parameters are defined in Table 3.

<table>
<thead>
<tr>
<th>Model</th>
<th>AICc</th>
<th>Δi</th>
<th>wi</th>
<th>R²</th>
</tr>
</thead>
<tbody>
<tr>
<td>BA+DPTH+DROAD-SLPO+UDEN</td>
<td>124.67</td>
<td>0.00</td>
<td>0.56</td>
<td>0.35</td>
</tr>
<tr>
<td>BA+DPTH+DROAD</td>
<td>126.07</td>
<td>1.40</td>
<td>0.27</td>
<td>0.26</td>
</tr>
<tr>
<td>BA+DPTH+DROAD-SLPO+UDEN+ELEV-HGTU</td>
<td>127.15</td>
<td>2.48</td>
<td>0.16</td>
<td>0.38</td>
</tr>
</tbody>
</table>
Table 6.— Coefficient estimates, standard errors, and odds ratios for parameters from the model (Table 5) that best predicted roost habitat selection of tri-colored bats in Land Between the Lakes National Recreation Area, KY and TN, during summer 2015 and 2016. Model parameters are defined in Table 3.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
<th>SE</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intercept</td>
<td>-5.1918</td>
<td>1.3888</td>
<td></td>
</tr>
<tr>
<td>BA</td>
<td>0.0182</td>
<td>0.0079</td>
<td>1.018</td>
</tr>
<tr>
<td>DPTH</td>
<td>0.06209</td>
<td>0.0316</td>
<td>1.064</td>
</tr>
<tr>
<td>DROAD</td>
<td>0.00247</td>
<td>0.00117</td>
<td>1.003</td>
</tr>
<tr>
<td>SLPO[Mid]</td>
<td>-1.2284</td>
<td>0.58992</td>
<td>0.893</td>
</tr>
<tr>
<td>UDEN[Med]</td>
<td>2.05835</td>
<td>1.08264</td>
<td>7.833</td>
</tr>
</tbody>
</table>

I also determined the best fit generalized linear model that explained the difference between roost sites and the large scale sites within a 5-km buffer of roosts used by tri-colored bats ($R^2 = 0.21$; Table 7). The model only contained elevation of the roost tree or randomly-selected point and distance to the nearest road. The probability that it was the best model of the models tested for the data collected was 49% (Table 7). The odds ratio indicated that when compared to random large scale sites, bats were more likely to roost in sites further away from roads and lower in elevation. The odds were 11% higher (Table 6) that a site was used for roosting with every 50 m increase in distance from a road.
Table 7.— Model parameters that explained the difference between roost sites and the larger scale random sites from the 5-km buffer for tri-colored bats in LBL, KY and TN, USA, 2015-2016. Along with the AICc value, Δi value, model weights (wi) and generalized R2 is given for each model. Model parameters are defined in Table 3.

<table>
<thead>
<tr>
<th>Model</th>
<th>AICc</th>
<th>Δi</th>
<th>wi</th>
<th>R²</th>
</tr>
</thead>
<tbody>
<tr>
<td>ELEV + DROAD</td>
<td>2</td>
<td>128.53</td>
<td>0.49</td>
<td>0.21</td>
</tr>
<tr>
<td>ELEV + DROAD + DWATER</td>
<td>3</td>
<td>129.24</td>
<td>0.34</td>
<td>0.22</td>
</tr>
<tr>
<td>ELEV</td>
<td>1</td>
<td>130.63</td>
<td>0.17</td>
<td>0.16</td>
</tr>
</tbody>
</table>

Table 8.— Coefficient estimates, standard errors, and odds ratios for parameters from the model (Table 7) that best predicted roost habitat selection of tri-colored bats in Land Between the Lakes National Recreation Area, KY and TN, during summer 2015–2016. Model parameters defined in Table 3.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
<th>SE</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intercept</td>
<td>4.495</td>
<td>1.736</td>
<td></td>
</tr>
<tr>
<td>ELEV</td>
<td>-0.4215</td>
<td>0.01269</td>
<td>0.656</td>
</tr>
<tr>
<td>DROAD</td>
<td>0.001997</td>
<td>0.0009906</td>
<td>1.002</td>
</tr>
</tbody>
</table>

Movement characteristics.— Bats moved on average 1120 ± 190 m from their point of capture to their first roost (range 165–2290 m). The average distance moved between roosts was 86 m ± 19 m (range 5–482 m). The number of roosts per bat was 1–6 for males and females. Bats were tracked for an average of 6.0 ± 0.9 days (range 1–12).
The mean number of roosts for females (2.6 ± 0.6) was not significantly different than for males (3.0 ± 0.8; \( t_{13} = 0.41, P = 0.69 \)). The number of days per roost per bat varied from 1–11 days for females and 1–4.5 for males with the mean number of days per roost for females (3.2 ± 1.1) not significantly different than for males (2.2 ± 0.6; \( t_{13} = 0.69, P = 0.50 \)). Of individual bats studied 27% of them returned to at least one previously used roost cluster.

Only bats with three or more locations were used to complete the roosting range estimation (\( n = 7 \)). The roosting ranges had a large span with the smallest range recorded using only 0.002 ha and the largest using 4.4 ha. The average range for all bats was 0.79 ± 0.6 ha with the average for males (0.20 ± 0.19 ha) not being significantly different (\( t_{13} = 0.73, P = 0.48 \)) than for females (1.2 ± 1.1 ha).

**DISCUSSION**

Bats spend over half their life roosting (Kunz and Parsons 2009). Summer roosts provide places to rear young, appropriate conditions for daily rest, shelter from predators, and shelter from adverse weather. Bats’ selection of roosts are likely influenced by food resource distribution, roost availability, predation risks, and energetics associated with roost conditions and commutes (Kunz and Fenton 2006). Twelve of the 16 bat species known to inhabit Kentucky and Tennessee occur within LBL. The abundance of species implies that there are adequate roosting and foraging resources within the landscape at LBL to support a diverse community of bat species. Considering the large amount of forested lands with various tree species and foliage types, as well as a number of human-made structures, it follows that roost sites are not likely to be a limiting factor for tri-colored bat populations in LBL.
Tri-colored bats are known to roost in a variety of structures and trees, and use different types of foliage and lichen as roosts (Lacki et al. 2007; Poissant et al. 2010). Since tracking small bat species through telemetry has become more feasible due to decreasing radio transmitter sizes, it has been discovered through summer day roost telemetry of the tri-colored bat that dead leaf clusters in mature trees are a common roost choice throughout much of their range (Veilleux et al. 2003; Perry and Thill 2007; O’Keefe 2009; Whitaker Jr et al. 2014). Selection for clusters of dead leaves was obvious in my research, and the roosts selected by tri-colored bats were exclusively within the leaves of live deciduous trees. Oaks were often noted as used or selected tree species for roosts by tri-colored bats (Veilleux et al. 2003; Leput 2004; Perry and Thill 2007; Whitaker Jr. et al. 2014).

At LBL, tri-colored bats selected for mockernut hickory trees and used white oak trees in accordance with their abundance as roost trees. These two frequently-used tree species have several similarities. They grow together in oak-hickory forests in temperate regions and have similar maximum heights of around 30 m which often places mature trees into or near the canopy (Tirmenstein 1991; Coladonato 1992). Oaks are known for having leaves which persist on the trees longer than most other vegetation, often retaining dead leaf clusters (Tirmenstein 1991). Tri-colored bats use these persistent dead leaf clusters to roost, as they seem to provide weather and visual protection from above and the sides (Perry and Thill 2007; Whitaker Jr et al. 2014). Visual crypsis is a common tactic for foliage roosting bats. The tri-colored bat has characteristic multicolored brown, yellow, and black hairs which resemble the coloration of browned foliage (Kunz and Fenton 2006; IUCN 2008). The mockernut hickory is known for having a broader and
rounder crown than other hickories, as well as having the heaviest nut within the hickory genus. Common to all hickory species, the mockernut hickory has pinnately compound leaves. Features of the leaves include 7–9 leaflets per leaf with each leaflet being about 5–15 cm long and 2-5 cm across (Coladonato 1992). Based on the leaf size and complexity, it is plausible that dense areas of leaves and dead or snapped branches that retain leaves of this type would provide substantial shelter both from visual predators and from inclement weather. Also, with the retention of the large nuts and nut husks it is possible that foliage roosting bats resemble these items. I documented use of dead and live hickory leaf clusters as roosts.

Although oaks were a selected roost in some of the most thorough studies of tri-colored bat roost selection, hickories were recorded as the most frequent roost tree used by tri-colored bats in North Carolina and for sympatric foliage-roosting eastern red bats in Kentucky (Hutchinson and Lacki 2000; O’Keefe 2009). Tri-colored bats and red bats share some common physical characteristics such as wing patterning of rouge forearms and black wing membranes which creates a disruptive pattern, as well as multicolored hairs that aid in camouflage (Wacker et al. 2016). Tri-colored and red bats may, therefore, share similar requirements for crypsis and preferences for roosts within leaf clusters as they have both been recorded roosting in dead leaf clusters in hickory trees in Kentucky (Hutchinson and Lacki 2000).

Regionally, there may be temperature, humidity and precipitation trends that drive selection for certain roosts. I did not collect data to compare roost sites based on microclimates. However, the average roost height for tri-colored bats was very near to the canopy suggesting that solar radiation and wind conditions, which change near the
canopy level (Kunz and Fenton 2006), may be important in roost site selection. Lower susceptibility to terrestrial predators is also thought to be a driver of high roosting heights and may be important in areas with a high density or diversity of predators (Hutchinson and Lacki 2000; Kunz and Fenton 2006; O’Keefe 2009).

I did not find any statistically significant differences in roost selection between sexes or reproductive classes in the tri-colored bat when I tested for each roost tree and plot habitat variable measured. Although differences have been reported elsewhere (Veilleux et al. 2004; Perry and Thill 2007), the lack of differences may be attributable to my small sample size for each reproductive class and lack of data on factors such as roost microclimate, that are likely to differ between reproductive classes due to the energetic costs inherent to each class (Crichton and Krutzsch 2000; Veilleux et al. 2004).

Most of the roosting habitat variables that I measured did not differ significantly between day roosts and random areas (Roost tree characteristics.— All confirmed leaf roosts were located in live deciduous trees (Table 4). Most of the females (62%) and males (70%) roosted in dead leaf clusters hanging in the live trees, with the remaining bats roosting in live leaf clusters. All roosts were within the top third of their respective roost tree, with the average roosting height (20.4 ± 2.1 m) being 2.2 m less than the average roost tree height (22.6 ± 2.0 m). The aspects of the slopes roosts were located on were not significantly different from a uniform distribution across aspects (Rao’s test statistic = 148, critical value = 158). The average percent canopy closure two m above the roosts (89.9 ± 1.6 %) was similar to the overall average percent canopy closure (88.9 ± 0.7 %) measured.
Table). Lack of significant differences in roosting habitat signify either a true absence of differences in selection for these habitat factors or an inability to observe differences due to other factors. One of these potential other factors was a lack of local variation. There appeared to be a high level of homogeneity in factors such as canopy cover and tree DBH within LBL when looking at the standard errors of the random plots.

Roost tree characteristics.— All confirmed leaf roosts were located in live deciduous trees (Table 4). Most of the females (62%) and males (70%) roosted in dead leaf clusters hanging in the live trees, with the remaining bats roosting in live leaf clusters. All roosts were within the top third of their respective roost tree, with the average roosting height (20.4 ± 2.1 m) being 2.2 m less than the average roost tree height (22.6 ± 2.0 m). The aspects of the slopes roosts were located on were not significantly different from a uniform distribution across aspects (Rao’s test statistic = 148, critical value = 158). The average percent canopy closure two m above the roosts (89.9 ± 1.6 %) was similar to the overall average percent canopy closure (88.9 ± 0.7 %) measured.
Table). There was less variation in elevation (<300 m), stream density, and forest age within LBL compared to that in the tri-colored bats’ extensive range.

The three tree factors that were of statistical significance (i.e., greater roost tree crown depth, greater roost plot basal area, and greater distance to nearest road) and the preference for medium understory vegetation density (Table 3) signified that there was some selection for tree and plot factors by tri-colored bats within LBL. The most frequently used species of roost tree, the mockernut hickory, has a crown depth of over 50% of the tree’s height when growing in a site with more open canopy (Coladonato 1992). Solar radiation, which can be important in roost selection (Kunz and Fenton 2006), was sufficient for extensive crown growth in the mockernut hickories used by the tri-colored bats studied with average crown depth being more than half of the average tree height (77 ± 6%).

The two plot variables that were significantly associated with roost trees were tree basal area and proximity to a road. The selection for roosting in plots with greater basal area was consistent with the preference of tri-colored bats for older and more mature stands when the overall high DBH across sites is taken into account (Leput 2004; Perry and Thill 2007; O’Keefe 2009). The preference for a comparatively high distance from roads is not generally reflected in other studies of tri-colored bats. However decreased use of habitat by small insectivorous bats was reported in areas 1-6 km from major road edges in a study of road effects on bats (Berthinussen and Altringham 2012). Differences in road density among areas may explain differences in results for tri-colored bats, but the road density is unavailable for other study areas (USFS 2004; O’Keefe 2009). There were approximately 2 roads/km² within LBL.
When examining roost selection on a larger scale, and including points further from the selected roosting tree, there were significant differences between the roosting and random sites. The preference for a comparatively high distance from roads was repeated, and a selection for lower elevations than random appeared. The significance of these variables indicated that a selection for distances further from roads was valid as a general relationship. The selection for lower elevations on only the larger scale may suggest selection for roosting sites within low basins including both sites of capture used for transit or foraging and sites for roosting.

Roost sites were also associated with secondary forest that had a developed understory. Tri-colored bats have high maneuverability compared with other foliage roosting bat species and can more easily fly through vegetation (Norberg and Rayner 1987). Due to the maneuverability of the species, the areas of medium density understory were not exclusionary. The selection for medium understory vegetation density was different from other studies that found selection for more open understory. The methodology for understory measurements are not identical among studies, however (Veilleux et al. 2003; Leput 2004; Perry and Thill 2007). The selection for medium understory density may have been a way of balancing costs of maneuvering with other benefits of flying and roosting in areas of higher vegetation density such as increased cover and potential community shifts of insects and predators (Thysell et al. 2000).

The movements and roost switching behaviors exhibited by tri-colored bats at LBL were very similar to those reported in previous studies of tri-colored bat roosting. The average distance moved of 1200 m from capture site to first roost in my study was within the range of values reported by studies in Indiana, Nova Scotia CA, and South
Carolina (300–5000 m) with an average among studies of 790 m (Veilleux 2001; Leput 2004; Quinn and Broders 2007). The average number of days per roost (2.7 days) at LBL was equal to the average among studies throughout the tri-colored bat’s range of 0.85–4.31 days (Kurta et al. 1999; Menzel et al. 1999; Veilleux 2001; Veilleux et al. 2003; Leput 2004; Perry and Thill 2007; Quinn and Broders 2007; O’Keefe 2009; Poissant et al. 2010). The area of ranges at LBL (0.79 ha), calculated with ≥ 3 roosts per bat, was close to the average across studies of 0.53 ha with a range of 0.002–1.1 ha (Krishon et al. 1997; Kurta et al. 1999; Menzel et al. 1999; Veilleux et al. 2003; Leput 2004; O’Keefe 2009). The roosting ranges of tri-colored bats were also similar to the ranges reported (0.004-2.6 ha) for other foliage roosting bat species (Lacki et al. 2007). The proportion of individuals (27%) who returned to at least one previous roost was near to the average reported between other studies (35%) with a range of 28–44% (Veilleux 2001; Leput 2004; O’Keefe 2009).

Movement similarities across regions and studies suggested a common basis for roost switching. Roost switching is common for many tree roosting bats (Veilleux et al. 2003; Lacki et al. 2007). Frequent switching has been thought to be an adaptive response to avoid predators, parasites, seek better microclimates, reduce commuting costs, or become familiar with alternative roosts (Lewis 1995; Kunz and Fenton 2006). Regardless of the reason, the consistent use of many roosts within a relatively small area informs potential conservation plans for these species. Although not tested during my study, the only tri-colored bats that were tracked during early night foraging (n = 3) appeared to use small (0.5–3 m width) dry and wet ephemeral stream beds near their roosting areas to commute to riparian foraging grounds. Thus, stream corridors are another potentially
important habitat and movement resource. Use of stream corridors was likely associated with the inclusion of the slope factor in the best fit model (Table 5), in which tri-colored bats showed a preference for localized small valleys that were defined by an ephemeral or perennial creek bed (i.e., bottomland).

Overall, roost selection by tri-colored bats at LBL was characterized by use of leaf clusters in mature deciduous trees that had broad deep crowns, were surrounded by a selection of similar mature trees, and were relatively near to bottomland corridors and relatively far from roads. Due to the high amount of roost switching and distinct range areas, there are several management implications that stem from these data. It is likely necessary to conserve and manage for large parcels of heterogeneous forest >10 km², with high numbers of mature trees to adequately protect habitat for the remnant populations of tri-colored bat that persist on the landscape. In LBL and likely large areas within the southeastern United States, live trees, including mockernut hickory and oaks of DBH ≥30 cm, may be very important as roost trees. These roost trees should also be included in relatively densely forested stands with > 7 m²/ha basal area, and potentially protected in parcels from fragmentation due to gaps such as roads.

Looking amongst the studies on tri-colored bat tree roost use, many characteristics of habitat use across differing regions, such as frequent roost switching and use of tall large trees near to water, remain consistent. Management to protect snags, decaying trees, exfoliating bark, or cavities does not appear to protect roosts for this species. In my study two bats each used only one roost tree throughout the time that they were tracked (6 and 11 days, respectively). The roost tree was in forest undergoing an active logged thinning treatment. The persistence of the bats in this area as well as their use of only one roost,
may have been due to lack of alternatives or by choice. Persistence of tri-colored bats in actively managed forests, especially long-term roost use in an area before, directly after, and some period after recovery from different management activities has begun, is an area of suggested further study with practical implications.

Some species of bat already have federal protection of their habitat (USFWS 2015, 2016); however, the roosting needs of those species are not consistent with those of the tri-colored bat. Currently, two federally-protected sympatric bat species occur within Kentucky and Tennessee that also use tree roosts during the summer, the Indiana bat (*Myotis sodalis*) and the northern long-eared bat (*Myotis septentrionalis*). The habitat that these species utilize differs from the habitat selected for by the tri-colored bat. Also, the range of these three species does not completely overlap (USFWS 2015, 2016; The Center for Biological Diversity and Defenders of Wildlife 2016). Trees with exfoliating bark, cracks, crevices, or hollows and habitat with linear openings are important habitat features for the Indiana and northern long-eared bat but are not factors that have been determined to be important for tri-colored bats (Veilleux et al. 2003; The Center for Biological Diversity and Defenders of Wildlife 2016; USFWS 2016). Therefore, additional survey and habitat protection measures will be necessary to conserve and manage for remnant tri-colored bat populations. I recommend that further studies are undertaken to gather more data about tri-colored bat roosting habitat in novel areas and in areas where the tri-colored bat has undergone severe population decline due to WNS.

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