Bat Occupancy Estimates and Species Richness at Cache River National Wildlife Refuge

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Running title: Occupancy Estimates of Bats of Cache River National Wildlife Refuge

Abstract

Six bat species of special concern, threatened or endangered, may occur in one of Arkansas' largest bottomland hardwood forests, the Cache River National Wildlife Refuge (CRNWR). However, inventory of bat species throughout the refuge has been lacking and management plans may not be adequate in promoting bat conservation. The objectives of this study were to inventory bat species in the CRNWR, and determine bat-habitat associations via occupancy estimates. From May–August 2014 and 2015, we mist-netted from sunset for 5 hours. We also deployed bioacoustic devices throughout 5 habitat types (cypress-tupelo [dominantly *Taxodium distichum* and *Nyssa aquatica*], emergent wetland, mature forest, hardwood reforestation, and managed hardwood). Mist-netting yielded 460 bat captures with Rafinesque's big-eared bats (*Corynorhinus rafinesquii; n* = 156) being the most common capture, followed by eastern red bats (*Lasiurus borealis; n* = 104), southeastern myotis (*Myotis austroriparius; n* = 91), evening bats (*Nycticeius humeralis; n* = 58), tri-colored bats (*Perimyotis subflavus; n* = 54), and a big-brown bat (*Eptesicus fuscus; n* = 1). Based on 3,896 calls identified with 85% certainty, evening bats and rarer big-brown bats tended to occupy managed hardwood forests more than any other habitat (occupancy probabilities ± SE: $\Psi = 0.75 \pm 0.13$ and 0.38 ± 0.19, respectively). Tri-colored bats tended to be more present in mature forest habitats ($\Psi = 0.91 \pm 0.09$), and *Myotis* species tended to have highest occupancy rates in cypress-tupelo stands ($\Psi = 0.59 \pm 0.15$). Not all species were detected with both methods. Thus, we encourage future studies to combine mist-netting and acoustic surveying methods to minimize bias in species presence estimate. This would ensure management practices that would benefit all present species.

Introduction

Since the colonization by European settlers, America's bottomlands have been greatly reduced and converted for agricultural use (Dahl 1980; Hank and Gosselink 1990). Only 10% of the original wetland habitat in the Mississippi Alluvial Plain remains today (Stanturf *et al.* 2000). The Cache River National Wildlife Refuge (CRNWR), when combined with other nearby conservation holdings, forms the second largest contiguous tract of forested wetland in Arkansas. The 27,315-ha refuge, founded in 1986 and located within Jackson, Woodruff, Monroe, and Prairie counties, is composed of bottomland hardwood forests (19,592 ha), reforested land (6,282 ha), and cropland and moist-soil units (1,441 ha). The CRNWR also borders several state wildlife management areas (WMA) such as Sheffield Nelson Dagmar WMA and Rex Hancock Black Swamp WMA as well as land owned by Arkansas Natural Heritage Commission. The CRNWR is listed on The Ramsar Convention of Wetlands as one of the Wetlands of International Importance in the United States, with 510 species of fauna and 120 species of trees and shrubs within the refuge (The Annotated Ramsar List: United States of America 2013).

Arkansas supports 16 bat species, 10 of which occur in the same counties as the CRNWR (Sealander and Heidt 1990). Two of these 10 species have some level of federal protection: the Indiana bat (*Myotis sodalis*), is listed as federally endangered, and the northern long-eared bat (*M. septentrionalis*) is threatened. Additionally, in Arkansas, the Rafinesque's big-eared bat (*C. rafinesquii*) and southeastern myotis (*M. austroriparius*) are species of special concern, and the little brown bat (*M. lucifugus*) is listed as a species of greatest conservation concern.

Several studies have examined the distribution of bats in bottomland forests of Arkansas (Fokidis *et al.*

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2005; Medlin 2006; Medlin *et al.* 2006). However, no study has exclusively focused on the bats of the CRNWR. Our first objective was to inventory bat species of the CRNWR. Our second objective was to estimate bat occupancy (i.e., the probability that a site selected at random is occupied by a species) in different habitats within the refuge. We hypothesized that habitat usage of the southeastern myotis and Rafinesque's big-eared bat is similar because both species are known to roost in cypress-tupelo dominated habitat (Jones and Manning 1989; Rice 1957, 2009; Stuemke *et al.* 2014). Therefore, we predicted that occupancy of *Myotis* bats and Rafinesque's big-eared bat would be highest in cypress-tupelo habitat compared to other habitats. Additionally, we hypothesized that habitat usage for other bat species is more flexible because of their wider distribution. Therefore, occupancy among species should be similar and reflect availability of other habitat types (Sealander and Heidt 1990; Fokidis *et al.* 2005; Medlin *et al.* 2006). The findings and conclusions in this article are those of the author(s) and do not necessarily represent the views of the U.S. Fish and Wildlife Service.

Materials and Methods

We captured bats over 45 nights during May 15 – August 15, 2014 and 2015 using 3–4 triple-high, 38-mm meshed mist-nets (AviNet Inc., New York, USA) of varying lengths. We sampled 21 netting locations, 1-3 times each, and spread our netting effort across the reproductive season. We chose netting locations in corridors with enclosed low canopies (e.g., roadways, waterways) to funnel bats into nets. We opened nets at sunset for 5 hours and checked for bat captures every 10 min, following U.S. Fish and Wildlife Service (USFWS) Indiana Bat protocol (USFWS 2016). All capture and handling procedures followed the guidelines of the American Society of Mammalogists for animal use (Sikes *et al.* 2011) and were approved by the Arkansas State University Institutional Animal Care and Use Committee (protocol 451729-1).

We used 2 bioacoustics approaches. In approach A, AnaBat SD2 Active Bat Detectors (Titley Electronics, Columbia MO) complemented netting efforts in both years. We deployed SD2 units, placed in modified ammunition boxes, before sunset on a 1-m tall PVC pipe anchored to the ground within 75 m of net-sites in fields, corridors or the interior of the forest. We collected detectors while nets were being closed for the night. Additionally, in approach B, from May–August, 2015, we collected search-phase echolocations of bats using 5

AnaBat SD2 units in 5 pre-defined habitat types: cypress-tupelo (dominated by *Taxodium distichum* and *Nyssa aquatica*; covering 7% of the CRNWR), reforestation (most trees were 10-20 years old; 21% coverage), mature forest (i.e., extant forest never cleared for agriculture; 65% coverage), managed hardwood (received some sort of silvicultural treatment; 4% coverage), and emergent wetland (moist-soil units, agricultural or open fields; 3% coverage). Each detector recorded calls for 3-5 consecutive nights in 16-20 stands (replicates) for each of the 5 habitat types, for a total of 91 sites. We programmed detectors to sample 30 min before sunset until 30 min after sunrise.

We classified search-phase echolocation calls of bats to species using Bat Call Identification version 2.7c (BCID, Kansas City, Missouri). We included only bats species whose range overlaps with the CRNWR in the analysis, i.e., eastern red bat (*Lasiurus borealis*), tri-colored bat (*Perimyotis subflavus*), Rafinesque's big-eared bat, big brown bat (*Eptesicus fuscus*), hoary bat (*L. cinereus*), evening bat (*Nycticeius humeralis*), and *Myotis* bats. The distribution of Brazilian free-tailed bats (*Tadarida brasiliensis*) only overlaps the southernmost portion of Prairie County and was therefore not included in the analysis. Although the Seminole bat's (*L. seminolus*) range overlaps with the CRNWR, reference libraries were not available in BCID or in EchoClass. We restricted calls to those containing at least 5 pulses (Mora *et al.* 2011) and we only retained those with a probability of correct species identification of ≥0.85. We then visually vetted retained calls with Analook 4.1 (Titley Electronics, Columbia, Missouri) to ensure accuracy. However, due to similarities in call structure between the Indiana bat, southeastern myotis, little brown bat, and northern long-eared bat, we placed all *Myotis* calls into one category.

We used only acoustic data collected under approach B to estimate single-season occupancy and probability of detection for each species in Program PRESENCE version 10.5 (US Geological Survey, Laurel, Maryland). Single-season occupancy models have 3 assumptions that must be met (MacKenzie *et al.* 2002): (1) sites are closed to changes in occupancy, which we met by having short sampling periods during which changes in occupancy are least likely to occur through volancy, death, or recruitment; (2) species are never falsely detected when absent, which we attempted to address by visually vetting calls; and (3) detection of a species at a site is independent of detecting the species at all other sites, which we met by having a single acoustic detector in each habitat type during each survey. We visually vetted calls by split-screen

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comparisons of collected and known calls, and by using an acoustic guidebook provided by Titley Electronics (Columbia, Missouri). Occupancy models estimate the probability of detection (p), i.e., the proportion of animals present that are detected, and the occupancy (Ψ) corrected by p, i.e., the probability that a site selected at random or sampling unit in a single area is occupied by a species (MacKenzie *et al.* 2006). For each species group, we conducted our analyses in three steps. First, we compared models with constant and survey-specific p while keeping Ψ constant. Models with constant p assign each survey effort the same probability of detection and estimate the highest probability of detection, whereas survey-specific p models assign probabilities of detection for each night of each survey effort. Second, starting with the best general structure for p, we compared p models with covariates (Julian date for a possible temporal trend and habitat types), keeping Ψ constant. Finally, using the best p model, we selected the best Ψ model with Julian date and habitat type as possible covariates. For all comparisons, an Akaike Information Criterion (AIC) was used to select the best model, i.e., the model with the lowest AIC (Burnham and Anderson 2002).

Results

Mist-netting yielded 460 bat captures for 45 trap-nights and 21 sites. The most common capture was the Rafinesque's big-eared bat ($n = 156$; Fig. 1), followed by eastern red bat ($n = 104$), southeastern myotis ($n = 91$), evening bat ($n = 58$), tri-colored bat ($n = 54$), and a Prairie County record for big brown bat ($n = 1$).

Table 1 – Bat occupancy model selection with constant ($p.$) versus survey-specific (p_s) detection probability. AIC, ΔAIC, and AICwt are Akaike Information Criterion, the difference in AIC for each pair of models, and the relative support of the model, respectively. The estimate of constant detection probability is indicated with its standard error.

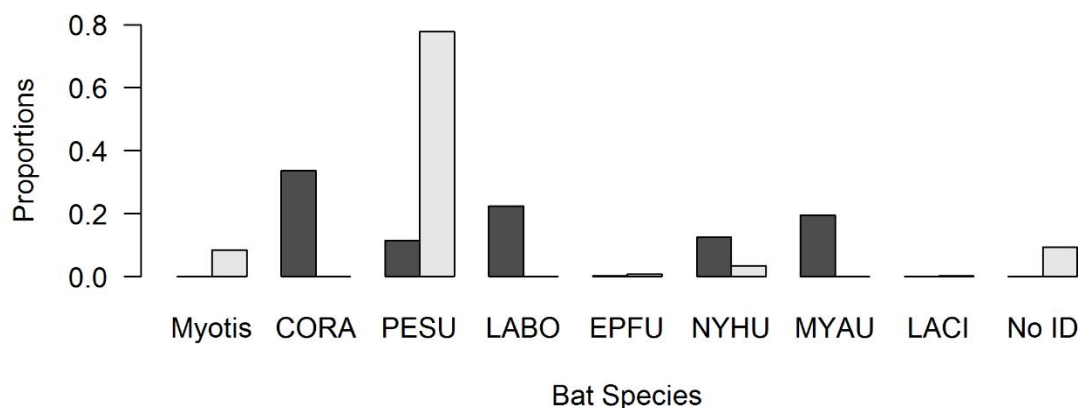| Models	AIC	ΔAIC	AICwt	p ($\pm$ SE)
Myotis bat				
$p.$	196.16	0.00	0.970	0.344 $\pm$ 0.071
p_s	203.12	6.96	0.030	
Tri-colored bat				
$p.$	324.64	0.00	0.755	0.833 $\pm$ 0.023
p_s	330.04	2.25	0.245	
Eastern red bat				
$p.$	99.19	0.00	0.959	0.073 $\pm$ 0.068
p_s	105.50	6.31	0.041	
Evening bat				
$p.$	347.17	0.00	0.881	0.423 $\pm$ 0.047
p_s	354.12	4.01	0.119	
Big brown bat				
$p.$	161.43	0.00	0.778	0.225 $\pm$ 0.074
p_s	163.94	2.51	0.222	

Figure 1. Proportions of individual bats captured through mist- netting (black) and call files collected through bioacoustics (gray), in 2014–2015, for Rafinesque's big-eared bat (CORA), eastern red bat (LABO), southeastern myotis (MYAU), *Myotis* species, evening bats (NYHU), tri-colored bat (PESU), big-brown bat (EPFU), hoary bat (LACI), and bat calls not identified to species (No ID). *Myotis* species were pooled together for the bioacoustics count because these species have similar calls and could not be distinguished with certainty. Bioacoustic data collected with two approaches (i.e., by net sites both years, and in 5 pre-defined habitat types in 2015 only) were pooled.

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Acoustic data collected with both approaches totaled 4,640 call files identified to species (approach A: $n_A = 744$ calls; approach B: $n_B = 3,896$ calls) and 483 call files recognized as bats but not identified to species. By decreasing order, 4,010 files ($n_B = 3,434$) were identified as tri-colored bats (Fig. 1), 426 ($n_B = 277$) as *Myotis* bats, 180 ($n_B = 166$) as evening bats, 39 ($n_B = 34$) as big-brown bats, 9 ($n_B = 9$) as hoary bats, 4 ($n_B = 3$) as eastern red bats, and 2 as Rafinesque's big-eared bats ($n_B = 0$).

Occupancy models with a constant (as opposed to survey-specific) probability of detection were the best models for all species and habitat types (Table 1). Probability of detection depended on habitat types for *Myotis* species and tri-colored bats (Table 2). *Myotis* were significantly less detected in managed forest than in cypress-tupelo stands, whereas tricolored bats were significantly more detected in managed hardwood than in mature forest (Table 3). On the contrary, detectability was constant for evening, big brown, and eastern red bats (Table 2).

The overall occupancy was (constant model) was highest for tri-colored bats (0.840 ± 0.039 [SE]), followed by evening bats (0.599 ± 0.069) and *Myotis* (0.301 ± 0.067). Big brown bats' occupancy (0.319 ± 0.101) (0.319 ± 0.101) did not differ from *Myotis* or evening bats, and the estimate for eastern red bats (0.475 ± 0.425) was associated with large uncertainty (Fig. 2). Constant occupancy was the most supported model for evening, tri-colored, and big brown bats, indicating no habitat was significantly more occupied by any of these species (Tables 3 & 4). However, tri-colored bats tended to occupy mature forest slightly more and evening bats had relatively higher occupancy in managed hardwood

Table 2 – Model selection for bat detection probability modeled as constant or as a function of habitat types and Julian date. Occupancy was kept constant. AIC, ΔAIC, and AICwt are Akaike Information Criterion, the difference in AIC for each pair of models, and the relative support of the model, respectively.

Models	AIC	ΔAIC	AICwt
Myotis			
Habitat	193.28	0.00	0.553
Habitat + Julian date	195.25	1.97	0.206
Constant	196.16	2.88	0.131
Julian date	196.51	3.23	0.110
Tri-colored bat			
Habitat	321.72	0.00	0.591
Habitat + Julian date	323.69	1.97	0.221
Constant	324.64	2.92	0.137
Julian date	326.61	4.89	0.051
Eastern red bat			
Constant	99.19	0.00	0.425
H	99.85	0.66	0.306
Julian date	101.19	2.00	0.156
Habitat + Julian date	101.85	2.66	0.112
Evening bat			
Constant	347.17	0.00	0.667
Julian date	348.76	1.59	0.301
Habitat	353.91	6.74	0.023
Habitat + Julian date	355.69	8.52	0.009
Big brown bat			
Constant	161.43	0.00	0.701
Julian date	163.41	1.98	0.261
Habitat	167.89	6.46	0.028
Habitat + Julian date	169.88	8.45	0.010

forests (Table 3). Occupancy for *Myotis* increased with time ($\text{slope}_{JD} = 0.019 ± 0.003$), but did not vary among habitat types although they tended to be more present in cypress-tupelo habitats (Table 3).

For eastern red bats, the best model indicates that occupancy varied among habitat types (Table 4), but it could not be estimated for two habitat types and the uncertainty for the estimated occupancy in the other three types was large (Table 3). Rafinesque's big-eared bats had too few confirmed calls to run occupancy analysis.

Discussion

The CRNWR's bat community included 6 species detected via acoustics that were also physically confirmed via capture in mist-nets. One other species,
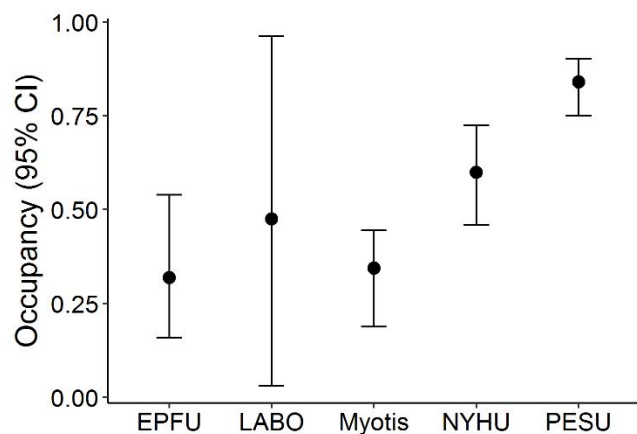

Figure 2 - Overall occupancy using constant model for eastern red bat (LABO), southeastern myotis (MYAU), *Myotis* species, evening bats (NYHU), tri-colored bat (PESU), and big-brown bat (EPFU) with 95% confidence intervals.

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Table 3 – Occupancy estimates (Ψ) and detection probabilities (*p*) for *Myotis* species, tri-colored, eastern red, evening, and big-brown bats in each pre-defined habitats (i.e., CT = cypress-tupelo; EW = emergent wetland, MF = mature forest, RF = reforestation, and MH = managed hardwood) of the Cache River National Wildlife Refuge for 2015. All *p* estimates are from models with Ψ constant and habitat-dependent *p*, whereas Ψ estimates are from models with habitat-dependent Ψ and *p* from the best species-specific model in Table 2.

Habitat	Ψ (95% CI)	*p* (95% CI)
Myotis bat		
CT	0.593 (0.296 – 0.835)	0.453 (0.284 – 0.634)
EW	0.174 (0.296 – 0.436)	0.255 (0.048 – 0.700)
MF	Not estimable	0.042 (0.005 – 0.273)
RF	0.229 (0.083 – 0.496)	0.341 (0.091 – 0.729)
MH	Not estimable	0.141 (0.043 – 0.375)
Tri-colored bat		
CT	0.834 (0.591 – 0.946)	0.845 (0.719 – 0.921)
EW	0.896 (0.661 – 0.975)	0.883 (0.773 – 0.943)
MF	0.909 (0.548 – 0.988)	0.674 (0.529 – 0.792)
RF	0.753 (0.522 – 0.894)	0.835 (0.699 – 0.917)
MH	0.843 (0.608 – 0.949)	0.909 (0.798 – 0.962)
Eastern red bat		
CT	Not estimable	0.092 (0.013 – 0.443)
EW	0.602 (0.070 – 0.968)	0.055 (0.007 – 0.341)
MF	Not estimable	Not estimable
RF	0.200 (0.016 – 0.793)	0.018 (0.001 – 0.212)
MH	0.210 (0.017 – 0.808)	0.037 (0.004 – 0.289)
Evening bat		
CT	0.588 (0.314 – 0.817)	0.488 (0.305 – 0.675)
EW	0.623 (0.343 – 0.840)	0.352 (0.206 – 0.533)
MF	0.454 (0.205 – 0.729)	0.407 (0.214 – 0.634)
RF	0.545 (0.288 – 0.781)	0.459 (0.269 – 0.662)
MH	0.750 (0.426 – 0.923)	0.419 (0.272 – 0.583)
Big brown bat		
CT	0.279 (0.076 – 0.646)	0.202 (0.049 – 0.556)
EW	0.363 (0.109 – 0.727)	0.287 (0.092 – 0.613)
MF	0.210 (0.045 – 0.600)	0.107 (0.019 – 0.424)
RF	0.362 (0.106 – 0.731)	0.257 (0.089 – 0.552)
MH	0.377 (0.111 – 0.747)	0.195 (0.067 – 0.450)

Table 4 – Model selection for bat occupancy modeled as constant or as a function of habitat types and Julian date. Detection probabilities were constant for eastern red, evening, and big brown bats, but were modeled as a function of habitat types for Myotis and tri-colored bats. AIC, ΔAIC, and AICwt are Akaike Information Criterion, the difference in AIC for each pair of models, and the relative support of the model, respectively.

Models	AIC	ΔAIC	AICwt
Myotis			
Julian date	192.31	0.00	0.388
Constant	193.28	0.97	0.239
Habitat + Julian date	193.67	1.36	0.196
Habitat	193.88	1.57	0.177
Tri-colored bat			
Constant	321.72	0.00	0.953
Habitat	327.75	6.03	0.047
Julian date	394.06	72.34	0.000
Habitat + Julian date	402.06	80.34	0.000
Eastern red bat			
Habitat	97.97	0.00	0.408
Habitat + Julian date	98.66	0.69	0.289
Constant	99.19	1.22	0.222
Julian date	101.19	3.22	0.082
Evening bat			
Constant	347.17	0.00	0.678
Julian date	349.04	1.87	0.266
Habitat	352.77	5.60	0.041
Habitat + Julian date	354.75	7.58	0.015
Big brown bat			
Constant	161.43	0.00	0.621
Julian date	162.57	1.14	0.351
Habitat	168.58	7.15	0.017
Habitat + Julian date	169.52	8.09	0.011

the hoary bat, was detected acoustically only. The presence of 4 of these 6 species (eastern red, big brown, evening, and tri-colored bats) was not surprising, as they are fairly common throughout the state (Fokidis *et al.* 2005; Sealander and Heidt 1990). However, proportions of captures and calls were not in agreement (Fig. 1). Only

two calls were recorded for the most commonly captured species, the Rafinesque's big-eared bat, whereas the most common species acoustically, the tri-colored bat, was the second least common capture. Rafinesque's big-eared bats may have been under-detected due to the ineffectiveness of zero-cross systems such as AnaBat systems (Hein *et al.* 2009) and because bats within the genus *Corynorhinus* echolocate at low intensities that are hard to detect, hence their nickname of "whispering bats" (Fenton 1982; Lacki and Bayless 2013; Loeb *et al.* 2015; Stihler 2011). Additionally, the tri-colored bat was the second least common capture via mist-netting but dominated acoustic surveys accounting for 81% of all identified call files. These findings are

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similar to other acoustic studies that reported low numbers of captures but high numbers of confirmed calls for this species (Young and Gruver 2011; Jordan 2014). The probability of detection for tri-colored bats was higher than for all other species. The amplitude of the species' echolocation is higher than in other species, which may inflate their detectability by zero-cross devices (Ryan Allen, *pers. comm.*; MacDonald *et al.* 1994), but may not necessarily reflect their relative abundance. Such inflated detection has the potential to bias conclusions. Also, although tri-colored bat calls are rather unique, we cannot rule out the possibility of some calls of other species being misclassified as tricolored bats. Although habitat type affected detection probabilities, the tri-colored bat was seemingly a generalist, not preferring any one habitat. They had a tendency to be more present in mature forest, as expected based on availability since mature forest represented the main habitat type (65%) in the refuge.

The loudness of tri-colored bats and quietness of Rafinesque's big-eared bat may lead to overestimated and underestimated occupancy estimates, respectively. Furthermore, the eastern red bat had the second highest physical capture rate among our 6 species, but it was also among the least common identified bat calls, despite higher frequencies than Rafinesque's big-eared bat. Eastern red bat calls may have been misclassified as evening bats (Britzke 2003), and these two species may need to be considered as one LABO/NYHU group in future studies (Cox *et al.* 2016).

As expected, *Myotis* bats tended to have higher occupancy at cypress-tupelo stands more than any other habitat type even though cypress-tupelo stands comprised 7% of the refuge. Although *Myotis* bats were placed into one category due to similarities of call structure, the *Myotis* bats' affinity toward cypress-tupelo stands could be reflective of the strong associations with bottomland hardwood forests of the southeastern myotis, the only Myotine bat captured during the study (Gooding and Langford 2004; Jones and Manning 1989; Rice 1957; Stuemke *et al.* 2014). Thus, the higher occupancy in cypress-tupelo habitats may suggest dominance of southeastern myotis over other *Myotis* bats. Due to similarities of *Myotis* calls, presence of the northern long-eared bat and Indiana bat on the CRNWR should not be excluded. An Indiana bat had possibly been detected acoustically in Jackson County in the summer of 2013 (Richard Crossett, *pers. observ.*). Capture at emergence and radio-tracking may provide more data to inform us about the likelihood of these species using the Delta in general and CRNWR specifically. Finally, we were not able to test the

prediction of a higher occupancy of Rafinesque's big-eared bats in cypress-tupelo habitats because of a lack of acoustic data, but we confirmed its presence in the refuge.

Acoustic data were in agreement with mist-netting data for big brown bats, both suggesting its rarity within the refuge. Although studies in highlands of the Ouachita Mountains (Saugey *et al.* 1989) and in the southeastern portion of the state where bottomland forests are present (Baker and Ward 1967) showed low capture rates of both Rafinesque's big-eared bats and southeastern myotis, higher captures of both species were reported in the east-central portion of the state (Fokidis *et al* 2005; Medlin *et al.* 2006). Higher numbers in the CRNWR may relate to overall suitability of the refuge for these two species. The CRNWR is also on the westward edge of their distribution (Arroyo-Cabrales and Álvarez-Castañeda 2008a,b) and the Arkansas Delta represents their core population areas in the state (Fokidis *et al.* 2005; Medlin *et al.* 2006). Big brown bats as well as evening bats tended to have higher occupancies in managed hardwood forest, which suggests that these two species may share a preference for habitats with a more open canopy as a result of silviculture treatments (Timpone *et al.* 2006; Istvanko *et al.* 2016). Therefore, our results only partially supported our prediction that habitat use would reflect habitat availability.

Conclusions

The results of this study show the value of a two-pronged method to surveying bats. Passive surveying methods such as bioacoustics can complement physical methods. Despite the similar echolocation calls among *Myotis* species and the current inability of bioacoustic devices to detect low-frequency calls of big-eared bats, acoustic monitoring is becoming a more standard and cheaper approach to bat research and can be used to assess spatiotemporal patterns of bat activity. Similarly, mist-netting provides physical evidence of a species presence although high-fliers such as hoary bats are more likely to be missed (Brown 1997). Therefore, it is recommended to combine acoustic surveys with mist-netting to confirm species presence or absence (Kaiser and O'Keefe 2015). If land managers of the CRNWR based management decisions solely off acoustic data, these decisions would not necessarily promote the most common bat species (i.e., Rafinesque's big-eared bat). In addition, due to zero-cross systems' seemingly ineffective ability at detecting low-amplitude bat calls, land managers could consider using full-spectrum

detectors to increase their chances of detecting the "whispering bats". Finally, this study provides land managers with a weighted guideline of how management practices in certain habitat types may affect bat species and can provide guidance during their decision making process.

Acknowledgements

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