# OVERWINTERING BRAZILIAN FREE-TAILED BATS (*TADARIDA BRASILIENSIS*) IN CENTRAL TEXAS: BASELINE POPULATION ESTIMATES AND MICROCLIMATE HABITAT ANALYSIS

THESIS

Presented to the Graduate Council of Texas State University-San Marcos in Partial Fulfillment of the Requirements

for the Degree

Master of SCIENCE

by

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San Marcos, Texas August 2012

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#### ACKNOWLEDGMENTS

First, I would like to thank Dr. Simpson, my advisor, for the many years of learning and guidance he has provided me. This thesis and the knowledge and skills I have gained would not have been possible without him. I would also like to thank my other committee members, Dr. Baccus and Dr. Weckerly, for their wealth of knowledge and for answering the many questions I had throughout my project. Thank you to Dr. Shwartz's lab for supplying field equipment that made my data collection possible. Thank you to Fran Hutchins, Jim Kennedy, and Mylea Bayless with Bat Conservation International who supplied equipment, field assistance, helped with my experimental design, and conducting my population estimates. I would like to include a special thank you to the land owners that provided me access to my sites. I also want to thank my friends and lab mates for all the help in the field and for the priceless advice I was given during the course of my degree. Finally, I want to thank my family, namely my husband and children for allowing me to complete my thesis. The countless trips into the field and hours away from home were made so much easier by your support and love. I could not have done any of this without you. And to my aunt Judy and my brother Robert who inspired me to pursue a degree in Wildlife Ecology. I love you.

This manuscript was submitted on July 24, 2012

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## ABSTRACT

# OVERWINTERING BRAZILIAN FREE-TAILED BATS (*TADARIDA BRASILIENSIS*) IN CENTRAL TEXAS: BASELINE POPULATION ESTIMATES AND MICROCLIMATE HABITAT ANALYSIS

by

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August 2012

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Colonies of Brazilian free-tailed bats (*Tadarida brasiliensis*) roost in central Texas from March through November. These bats have historically migrated south in late fall leaving summer roosts unoccupied during winter. Recently, new overwintering populations have been discovered in central Texas. The objectives of my study were to confirm the presence or absence of overwintering free-tailed bat colonies at six summer roosts, conduct baseline population estimates, and evaluate temperature and absolute humidity of habitats with and without overwintering bats during winters of 2010-2011 and 2011-2012. I placed 3 Hygrochron iButton data loggers within each roost and 1 outside to monitor temperature and humidity hourly. I entered each roost once a month (November-February) to determine bat presence. I recorded digital images with a laser caliper at 2 roosts with overwintering Brazilian free-tailed bat populations. I analyzed and estimated population sizes using ImageJ software. I estimated population size at a bridge using crevice measurements combined with previously established roosting densities for bridges. I estimated the population at a bat house roost containing Brazilian free-tailed bats using the percentage of space occupied based on dimensions and previously established roosting densities for bat houses. Two roosts were vacant and treated as control sites. My results indicated a difference in temperature (P = 0.086) but not absolute humidity (P = 0.813) between unoccupied and occupied overwintering roosts ( $\alpha = 0.10$ ). The populations increased at all occupied roosts from 2010 to 2011. I also report the northern-most occurrence of overwintering Brazilian free-tailed bats known in Texas. Overall, these data may lead to a greater understanding of the natural history of Brazilian free-tailed bats and identification and protection of potential overwintering sites in central Texas.

#### **CHAPTER I**

#### INTRODUCTION

The Brazilian free-tailed bat (*Tadarida brasiliensis*; hereafter free-tailed bat) is one of the most abundant, wide ranging mammalian species in North America (McCracken 2003). Free-tailed bats range throughout Mexico to Oregon and from California to North Carolina (Wilkins 1989). These bats are of great economic importance throughout the region because major crop pests are their main food sources, and they perform a vital role in ecosystems as insectivores (Cleveland et al. 2006; Lee and McCracken 2002). Cleveland et al. (2006) estimated that free-tailed bats provided \$741,000/year in economic assistance in pest control in south-central Texas and models predicted they reduced cotton crop damages by 43%-50% (Federico et al. 2008).

The estimated summer free-tailed bat population in central Texas was historically hundreds of millions (Constantine 1967; Davis et al. 1962; Wahl 1993). Betke et al. (2008) suggested a population decline due either to earlier inaccurate surveying methods or an overall natural decrease in population size. Free-tailed bats inhabited caves, bridges, and abandoned mines and tunnels in central Texas from March through October or November. These populations have long been considered migratory, traveling to more southern latitudes during winter (Glass 1958; Villa 1956; Villa and Cockrum 1962). Small populations of free-tailed bats have been documented as overwintering in Texas for many years (Glass 1982; Keeley and Keeley 2004; Krutzsch and Sulkin 1958; Spenrath and LaVal 1974; Tuttle 2003). Recent observations indicated overwintering populations

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had established at new roosts in central Texas with several believed to be increasing in size (pers. comm., F. Hutchins, Bat Conservation International, and S. Fulton, Bamberger Ranch). This may be related to multiple factors, including but not limited to the presence of a previously unavailable food source such as overwintering migratory moths (Vaughan 1976; Westbrook 2008), changes in roosting sites within the winter range (Kunz 1982; Tuttle 1977), as well as global climate change (Frick et al. 2010; Popa-Lisseanu and Voigt 2009; Scheel et al. 1996). Climate change has been suggested as a key factor influencing migratory behavior of birds (Bezzel and Jetz 1995; Bradley et al. 1999; Pulido and Berthod 2010) and might also influence the migratory behavior of bats (Popa-Lisseanu and Voigt 2009). Minimal survey and environmental data have been collected on overwintering populations of free-tailed bats in central Texas because this phenomenon is relatively new. Recent research conducted in New Mexico verified an overwintering population at Carlsbad Cavern (Geluso 2008); however, no published studies on overwintering populations in Texas exist. Populations of free-tailed bats overwintering in central Texas may be disturbed by guano mining during winter at large summer roost sites. Thus, there is a need for assessing the impacts of guano mining in winter roosts.

Baseline surveys are necessary for biologists to properly evaluate population trends and fluctuations, changes in habitat parameters, and responses to management (Duff and Morrell 2007). General knowledge of colony size is also required to analyze and interpret the ecology of these gregarious animals (Hristov et al. 2010), and the limited information about the status of free-tailed bat populations, such as colony size, in Texas as well as North America are continuing concerns (McCracken 2003). Recently, the need for information about overwintering populations has become increasingly important due to the spread of White-Nose Syndrome, a cold-adapted fungus that has killed millions of bats throughout the eastern United States. White-Nose Syndrome was discovered in an Oklahoma cave used by free-tailed bats for roosting during summer (Cohn 2012); however, the fungus has not been verified as existing in a bat population in a Texas cave.

Microclimate is a key factor influencing roost site selection during winter (Martin et al. 2006). Minimal data have been collected on the microclimate of overwintering roosts compared to the microclimate of roosts used by free-tailed bats when large maternity colonies are formed throughout central Texas (Herreid 1963; Kunz and Robson 1995; Reichard et al. 2009),

The objectives of my study were to a) confirm the presence or absence of overwintering (November, December, January, and February) free-tailed bat colonies at known summer roosts, b) obtain baseline estimates of overwintering populations, c) measure microclimate variables of temperature and absolute humidity at 6 known summer roosts during winter 2010-2011 and 2011-2012, and d) examine microclimate differences between roosts with and roosts without overwintering free-tailed bat populations.

### **CHAPTER II**

# MATERIALS AND METHODS

I visited 6 free-tailed bat summer roosts in central Texas (Fig. 1) during winter (November, December, January, February) 2010-2011 and 2011-2012. Four of these summer roosts had overwintering bat populations, including Bracken Bat Cave (Comal County, 29°41'13"N, 98°21'34"W), D'Hanis Bridge (Medina County, 29°19'34"N, 99°17'40"W), Old Tunnel State Park (Kendall County, 30°06'01"N, 98°49'14"W), and the Chiroptorium (Blanco County, 30°11'14"N, 98°28'34"W). Two roosts did not have confirmed overwintering populations--Davis Blowout Cave (Blanco County, 30°27'17"N, 98°34'07"W) and James River Bat Cave (Mason County, 30°34'14"N, 99°19'59"W). I visited each roost once per month (total of 8 visits per location during 2 winters). I did not use data from a seventh roost with a confirmed overwintering population, Frio Bat Cave (Uvalde County), due to data logger problems.

*Population Estimation.---* I estimated overwintering bat populations in December of 2010 and 2011. I used different methods to estimate populations at each roost because of structural diversity of roosting locations as well as roosting habits of bats. I used digital photography and a laser caliper with a setting of 15.2 cm for roosting area estimates and population counts at Bracken Bat Cave and Old Tunnel State Park (Meretsky et al. 2010). I took digital images perpendicular to roosting clusters using a Canon EOS Rebel T2i with a 70-300mm lens and a Canon Speedlite 430EX II flash. The laser caliper and camera were mounted on a metal plate with two bubble levels parallel to the camera lens axis. The complete assembly was stabilized with a tripod. I reviewed the digital quality of the images as pictures were taken in the field. Disturbance to roosting bats was reduced whenever possible by only using enough light from the laser caliper to identify clusters and their outlines and focusing my camera. I used ImageJ (National Institute of Health, USA), a public domain software program, to analyze all photos (Abramoff et al. 2004).

At Bracken Bat Cave, I included the entire cluster of roosting bats in 1 image and used the known distance from the laser caliper to set the scale in ImageJ. This allowed me to draw polygons around a roosting cluster and determine roosting surface area. Polygons were drawn 10 times in each photo and an average of resultant areas was determined. However, bats roosted approximately 38 m from the cave floor during both seasons, which prevented me from obtaining the resolution necessary to count individual bats for density estimates. Current and historical estimates of Brazilian free-tailed bat populations using extrapolation calculated densities at either 2,153 or 3,229 adult bats/m<sup>2</sup> (Constantine 1967; Kennedy 2003). I conservatively estimated the population density at 2,153/m<sup>2</sup> and extrapolated to the area based on the average size of polygons.

Bats were spaced much closer at Old Tunnel State Park, and I took quality photos for enhancement. I used ImageJ to enhance images and estimate numbers of  $bats/m^2$ . I created a 30.5 x 30.5 cm square on the enhanced images using the laser caliper distance as a reference. I counted each bat inside of this square. A digitized point was placed on each bat's nose to eliminate double counting. Old Tunnel State Park presented its own unique challenges. The roosting surface covered by bats was too long to incorporate into a single photo or stitch together in a series of images. The bats also did not cover the entire area evenly. Therefore, I took pictures at random points within the occupied roost area and measured the roost width along the tunnel length occupied by bats. I then placed a grid on each photo using ImageJ and estimated percent area covered by bats in each square to get a mean percent cover of bats in each photo. The mean percent cover of bats in each photograph was then averaged to estimate the percentage of the entire roost covered by bats. I multiplied the measured area by total percent cover and number of bats/m<sup>2</sup> to produce a final estimate. This gave me an estimated roost population.

Bats roosted inside crevices at D'Hanis Bridge. A previously established range of 431-538 bats/m<sup>2</sup> was averaged and combined with crevice measurements of occupied roosting space taken with a 100-m measuring tape (pers. comm., Mylea Bayless, Bat Conservation International). Lastly, the Chiroptorium contains 7 bat houses of varying sizes, which housed all overwintering bats during both seasons. I measured dimensions of each bat house and took pictures of colonies inside to visually estimate the percent of each bat house occupied. I then used the accepted measurement of 2 bats/linear inch (pers. comm., Mylea Bayless, Bat Conservation International) and applied this to total linear inches occupied.

*Microclimate Habitat Analysis.---*I placed 3 Hygrochron iButton data loggers (Maxim Dallas Semiconductor Corp., Dallas, Texas) inside and 1 outside each roost to monitor microclimate data on the first visit to each location in November 2010. I positioned data loggers inside each roost as close to roosting bats as possible (most within 1 m). I could not position data loggers in Bracken Bat Cave close enough to roosting bats to obtain measurements representative of the roosting environment, and therefore, I eliminated this

roost from microhabitat analysis. I placed data loggers in areas occupied by bats during summer at unoccupied winter roosts as evidenced by the presence of guano at a minimum of 15 m apart. Each data logger recorded temperature and relative humidity at 60 min intervals during November, December, January, and February. I downloaded data in February 2011 and 2012. I converted relative humidity to absolute humidity (g/m<sup>3</sup>) using the recorded temperature and relative humidity data (Vaisala 2010):

$$AH = 216.679 * \frac{P_w}{T_k}$$
$$P_w = P_{ws} * \frac{RH}{100}$$
$$P_{ws} = 6.1162 * 10^{\circ} \frac{7.5892 * T_c}{T_c + 240.71}$$

where *AH* represents absolute humidity,  $P_w$  represents water vapor pressure in hectoPascals (hPa),  $T_k$  and  $T_c$  represent temperature in degrees Kelvin and Celsius, respectively,  $P_{ws}$  represents saturation water vapor pressure in hPa and *RH* represents relative humidity. I used absolute humidity because it is a better measure of air moisture than relative humidity and is independent of temperature (Colloff 2009; Hillman et al. 2009).

Safety precautions were taken by all who entered roosting sites due to the risk of histoplasmosis, potential exposure to rabies, and concern for (WNS). I used incandescent flashlight headlamps and respirators in all locations where histoplasmosis was a concern and bats were never handled. I also followed the U.S. Fish and Wildlife Service's quick reference for White–Nose Syndrome (WNS) containment and decontamination procedures for cave activity (United States Fish and Wildlife Service 2011). All research was conducted under the Texas State University IACUC permit number 1029\_0909\_24.

*Statistical Analysis.---*I determined confidence intervals for population estimates for each roost individually due to differences in methodology. I calculated the 95% confidence intervals using standard deviation of area estimates for Bracken Bat Cave. I used standard deviation of the number of bats estimated/m<sup>2</sup> to calculate confidence intervals at Old Tunnel State Park. I did not determine confidence intervals for the Chiroptorium because bat house estimates were based on assumed amount of space required for a single individual. No range of densities has been reported and no methods for determining bat house population estimates have been published. Finally, I calculated the D'Hanis Bridge confidence intervals using the range of bats estimated/m<sup>2</sup> (pers. comm., Mylea Bayless, Bat Conservation International). I analyzed differences between mean weekly temperature and absolute humidity of roosts with over-wintering bats and roosts without bats with linear mixed-effects models (LME). I considered a *P*-value < 0.10 statistically significant due to low sample size. Microclimate analyses were done using R statistical software program (R Development Core Team 2004).



Figure 1. Locations of six Brazilian free-tailed bat roosts (Bracken Bat Cave, D'Hanis Bridge, Old Tunnel State Park, Chiroptorium, Davis Blowout Cave, and James River Bat Cave) I studied in winter 2010-2011 and 2011-2012.

#### **CHAPTER III**

## RESULTS

Free-tailed bats occupied Bracken Bat Cave, the Chiroptorium, Old Tunnel State Park, and D'Hanis Bridge during November, December, January, and February of 2010-2011 and 2011-2012. James River Bat Cave was unoccupied both years. Davis Blowout Cave was occupied only in February of 2011 and 2012 by migratory populations of freetailed bats. Also, the Old Tunnel State Park population increased in November 2010 (pers. comm., Nyta Brown, Texas Parks and Wildlife Department). I assumed free-tailed bat migration was in progress during November and February and restricted analyses to data from non-migratory winter months of December and January.

The largest free-tailed bat population during winter 2010-2011 was at D'Hanis Bridge with 216,128 bats ( $\pm$  24,013). I estimated Old Tunnel State Park had 65,015 ( $\pm$  1,948) free-tailed bats and Bracken Bat Cave had 15,233 ( $\pm$  1,223) free-tailed bats during winter 2010-2011. The Chiroptorium contained the smallest population of 635 free-tailed bats (Table 1).

I observed the largest population of free-tailed bats, 254,502 ( $\pm$  9,255), during winter 2011-2012 at Old Tunnel State Park. D'Hanis Bridge had the second largest population with an estimated 226,349 ( $\pm$  25,150) free-tailed bats. I estimated Bracken Bat Cave contained 88,834 ( $\pm$  2,232) free-tailed bats. The Chiroptorium again had the smallest estimated population at 885 free-tailed bats. Populations of free-tailed bats at all roosts increased from 2010 to 2011 with Bracken Bat Cave and Old Tunnel State Park roosts showing the largest increases of 480% and 277%, respectively (Table 1).

James River Cave had the lowest mean absolute humidity during both seasons (5.98 g/m<sup>3</sup> in 2010-2011 and 8.00 g/m<sup>3</sup> in 2011-2012). The highest mean absolute humidity was recorded at Davis Blowout Cave for both seasons (9.65 g/m<sup>3</sup> in 2010-2011 and 11.9 g/m<sup>3</sup> in 2011-2012). The lowest mean temperature was recorded in Old Tunnel State Park for both seasons (8.58°C in 2010-2011 and 9.21°C in 2011-2012). The highest mean temperature was recorded in Davis Blowout Cave for 2010-2011 and 2011-2012 at 16.6 °C and 17.3 °C, respectively. Temperatures appeared to be most stable at roosts without bats. The temperature at Davis Blowout Cave only fluctuated 8.34 °C during 2010-2011 and 6.84 °C during 2011-2012. Temperature ranges at James River Bat Cave were similar to Davis Blowout Cave at 8.85 °C during 2010-2011 and 8.35 °C during 2011-2012 (Tables 2 and 3).

Internal temperatures for both Davis Blowout Cave and James River Bat Cave did not follow the same trends as external temperatures during either season (Figs. 2 and 3). James River Bat Cave had similar internal absolute humidity fluctuations as the external environment; however, Davis Blowout Cave did not (Figs. 2 and 3). Old Tunnel State Park most closely followed external temperatures and absolute humidity during both seasons (Fig. 4). Both the Chiroptorium and D'Hanis Bridge had trends similar to the external environment with occasional divergences (Figs. 5 and 6).

Results of the LME indicated a statistical difference in winter temperature (P = 0.086, Table 4) but not absolute humidity (P = 0.813, Table 5) between roosts with and roosts without overwintering free-tailed bats. There was also a difference in mean weekly

temperatures between roosts (P = 0.001, Table 4) and a difference in absolute humidity

between years (P < 0.001, Table 5).

Table 1. Population estimates and 95% confidence intervals for Brazilian free-tailed bats at 4 overwintering roosts in December 2010 and 2011.

	Dec	ember 2010	December 2011		
Site	Estimate	95% CI	Estimate	95% CI	
Bracken	15,233	14,101-16,456	88,334	86,102-90,565	
Chiroptorium	635	N/A	885	N/A	
D'Hanis Bridge	216,128	192,115-240,140	226,349	201,199-251,499	
Old Tunnel	65,015	63,067-66,963	254,502	245,248-263,757	

	2010-2011					
-	Average Abs. Humidity	Average Temp.	Minimum Temp.	Maximum Temp.	Temp. Range	
Site	<u>(g/m<sup>3</sup>)</u>	( <sup>0</sup> C)	( <sup>0</sup> C)	( <sup>0</sup> C)	( <sup>0</sup> C)	
Davis <sup>a</sup>	9.65	16.61	12.47	20.81	8.34	
James River <sup>a</sup>	5.98	14.15	9.62	18.47	8.85	
Chiroptorium	7.25	11.68	3.93	28.31	24.38	
D'Hanis	7.94	13.00	7.11	19.48	12.37	
Old Tunnel	7.44	8.58	0.06	16.95	16.89	

Table 2. Temperature (°C) and absolute humidity (g/m<sup>3</sup>) summaries of all roosts from December 2010 through January 2011.

<sup>a</sup>without bats

Table 3. Temperature (°C) and absolute humidity (g/m<sup>3</sup>) summaries of all roosts from December 2011 through January 2012.

	2011-2012						
Site	Average Abs. Humidity	Average Temp	Minimum Temp	Maximum Temp	Temp. Range		
Site	(g/m)	( )	$(\mathbf{c})$	( C)	( C)		
Davis <sup>a</sup>	11.90	17.30	14.47	21.31	6.84		
James River <sup>a</sup>	8.00	14.94	11.29	19.64	8.35		
Chiroptorium	9.53	11.73	4.60	24.48	19.88		
D'Hanis	10.27	13.11	7.45	19.31	11.92		
Old Tunnel	8.63	9.21	1.91	17.95	16.04		

<sup>a</sup> without bats

Variable	Estimate	SE	d.f.	t	р
Intercept	16.51	1.43	82	11.51	0.000
Bats Present	-4.56	1.81	3	-2.52	0.086
Week	-0.19	0.05	82	-3.58	0.001
Season	0.43	0.27	82	1.60	0.114

Table 4. Linear mixed effect model (LME) results between mean weekly temperatures (°C) at roosts with and without overwintering Brazilian free-tailed bats from December-January of 2010-2011 and 2011-2012.

Table 5. Linear mixed effect model (LME) results on mean weekly absolute humidity  $(g/m^3)$  at roosts with and without overwintering Brazilian free-tailed bats from December-January of 2010-2011 and 2011-2012.

Variable	Estimate	SE	d.f.	t	р
Intercept	8.04	1.17	82	6.87	0.000
Bats Present	-0.38	1.47	3	-0.26	0.813
Week	-0.03	0.05	82	-0.65	0.520
Season	2.01	0.25	82	7.99	0.000



Figure 2. Average weekly patterns of internal versus external temperature (°C) (A) 2010-2011 and (B) 2011-2012 and absolute humidity  $(g/m^3)$  (C) 2010-2011 and (D) 2011-2012 at James River Cave.



Figure 3. Average weekly patterns of internal versus external temperature (°C) (A) 2010-2011 and (B) 2011-2012 and absolute humidity  $(g/m^3)$  (C) 2010-2011 and (D) 2011-2012 at Davis Blowout Cave.



Figure 4. Average weekly patterns of internal versus external temperature (°C) (A) 2010-2011 and (B) 2011-2012 and absolute humidity  $(g/m^3)$  (C) 2010-2011 and (D) 2011-2012 at Old Tunnel.



Figure 5. Average weekly patterns of internal versus external temperature (°C) (A) 2010-2011 and (B) 2011-2012 and absolute humidity  $(g/m^3)$  (C) 2010-2011 and (D) 2011-2012 at D'Hanis bridge.



Figure 6. Average weekly patterns of internal versus external temperature (°C) (A) 2010-2011 and (B) 2011-2012 and absolute humidity  $(g/m^3)$  (C) 2010-2011 and (D) 2011-2012 at the Chiroptorium.

### **CHAPTER IV**

#### DISCUSSION

I confirmed the presence of overwintering free-tailed bat populations and established baseline estimates at 4 roosts. Recoveries of banded free-tailed bats in central Texas during winter were reported as early as 1952 (Glass 1982). However, no attempts were made to determine roosting locations. It was also concluded that they did not occur above the southern edge of the Edward's Plateau region of Texas, and their winter range was between 29°N and 20°N latitudes (Glass 1982). All roosts with overwintering bats occurred north of the 29°N latitude. I also discovered populations overwintering at 2 bridges in Hays County, Texas. One located in San Marcos at I-35 and Centerpoint Road (Hays County, 29°49'40"N, 97°59'07"W), and another in Wimberley on Ranch Road 12 at the Cypress Creek crossing (Hays County, 29°59'48"N, 98°05'51"W). Both locations are also north of 29°N latitude. Another overwintering, breeding population was reported at McNeil Bridge on I-35 (Round Rock, Travis County, 30°29'59"N, 97°40'45"W) (Keeley and Keeley 2004). A large number of free-tailed bats died at this site after prolonged freezing temperatures in late January and early February of 2011. This to my knowledge is the northern-most occurrence of free-tailed bats during winter in Texas north of 30°N latitude. These results might indicate a northward expansion in Texas of the winter range of the species.

The combined free-tailed bat populations from all winter roosts totaled 297,011 bats in 2010 and 570,070 in 2011, almost doubling from 2010 to 2011. This increase might be due to weather driven fluctuations rather than an increasing overwintering population. Eads et al. (1957) reported no overwintering bats at Bracken Bat Cave during 3 consecutive winters suggesting a slight northward expansion of the winter range over the last 50 years. Continued monitoring of overwintering roosts and populations are needed to document any further northward expansion of the overwintering range of freetailed bats.

I found conducting population estimates of the free-tailed bats was challenging. Combining digital photographs with a laser caliper greatly increases the accuracy of population estimates when clusters of free-tailed bats can be included in 1 photograph or stitched together in a series of images (Meretsky et al. 2010). However, this was not possible at Old Tunnel State Park since the population roosted in groups up to 68 m in length and 4.88 m wide. Use of thermal imaging equipment would appear to be the best method in such situations (Betke et al. 2008). However, thermal imaging requires bats to emerge from their roosting site. Bats do not predictably emerge during winter (Geluso 2008). Studies on the activity patterns of overwintering free-tailed bats in central Texas are needed to determine the best method for population estimates at Old Tunnel State Park. It is important to note photos should be taken at the same zoom and distance from the roost if digital photography is used for this site. There are irregularly shaped guano mounds located randomly throughout the tunnel that make this a challenge. Analyzing photographs also does not account for irregularities in walls and ceilings of roosts. Therefore, my population estimates for Old Tunnel State Park and Bracken Bat Cave are conservative.

I did not obtain confidence intervals for the bat house population estimates in the Chiroptorium due to a lack of published methodology for accepted roosting densities. It is not clear how and where density estimates were established (pers. comm., Jim Kennedy, Bat Conservation International). Other methods for obtaining population estimates for bat houses include emergence counts (Brittingham and Williams 2000). This was not a feasible option because of the unpredictability of bat emergence in response to variable winter weather conditions. I could not determine bat roosting densities in Bracken Bat Cave because of challenges due to equipment capabilities. Lighting was insufficient to count individual bats due to the distance of bats from the camera. I recommend a brighter flash to obtain high quality photos in future winter surveys.

Microclimate results of my research indicate free-tailed bats are selecting colder, less stable environments during winter in central Texas. Free-tailed bats enter obligate torpor when temperatures are below 22 °C with thermal conductance remaining independent of ambient temperatures from 13.6-26 °C (Soriano et al. 2002). They survive only 1/3 to 1/4 as long as hibernating vespertilionid species when exposed to conditions similar to hibernating environments (Herreid 1963). While mean temperatures were below 22 °C at non-wintering sites, the mean temperatures were 4.56 °C higher (Table 2), and more stable than roosts with bats. Hays et al. (1992) and Speakman and Racey (1991) suggested that bats inhabiting winter sites with strongly correlated internal and external roost temperatures can evaluate external conditions without emerging, which is energetically costly. Correlation of internal to external roost temperatures has been discovered in hibernacula of several other species of bats (Daan 1973; Ransom 1968). It has also been demonstrated that bats preferentially emerge during warmer evenings (Avery 1985; Brack and Twente 1985; Ransome 1968) and will not emerge on cold nights when food is unavailable (Hays et al. 1992). The temperature patterns at sites without overwintering free-tailed bat populations did not appear to correlate with external temperatures as strongly as roosts with free-tailed bats based on visually analyzing graphed data and might be an explanation why free-tailed bats were not found at James River Bat Cave and Davis Blowout Cave.

Only 1 other bat species was encountered during both years of my study. A single cave myotis (*Myotis velifer*) was discovered in the Chiroptorium in January of 2012. No other hibernating species were found at James River Bat Cave or Davis Blowout Cave, which supports conclusions that higher temperatures along with stable temperatures are problematic for free-tailed bats during winter. Hibernating cave myotis (Tinkle and Patterson 1965) and tri-colored bats (*Perimyotis subflavus*) (Briggler and Prather 2003) require thermally stable hibernacula and both hibernate in sites throughout central Texas (Fitch et al. 1981; Fujita and Kunz 1984), but both species were not found at my roosts without overwintering free-tailed bats. This suggests higher temperatures may also influence roost selection for hibernating bats during winter.

Absolute humidity was not a factor influencing roost site selection in my study. James River Bat Cave had the lowest absolute humidity and Davis Blowout Cave had the highest both years. The range of humidity values recorded at my sites was within the accepted range for survival of free-tailed bats (Herreid 1963; Twente 1956). Only 2 of my winter roost sites lacked overwintering populations, therefore, humidity as a factor influencing roost site selection cannot be ruled out due to low sample size. It is possible that humidity may be a factor at other sites. Also, there might be other variables that I did not measure influencing roost site selection, such as airflow (Tuttle and Stevenson 1981).

While Davis Blowout Cave did not have an overwintering free-tailed bat population during my study, Krutzsch and Sulkin (1958) reported a few free-tailed bats occurred in the cave during winter. It is not mentioned whether these bats survived through winter or were transients staying at the cave for any length of time. It is possible the James River and Davis Blowout caves are transient sites used briefly but not as permanent winter roosts. I only entered each site once a month, so bats could have been present in either James River or Davis Blowout Cave when external temperatures were warm enough to allow successful foraging. However, on several visits to these sites, external temperatures were above 22 °C and bats were not present. Therefore, I do not think these sites were used between visits.

Two subspecies of Brazilian free-tailed bats are documented in Texas, the Brazilian free-tailed bat, also called guano bat (*T. b. mexicana*), and Le Conte's freetailed bat (*T. b. cynocephala*) (Schwartz 1955). The subspecies are usually identified based on their migratory differences. The Mexican free-tailed bat is considered migratory and occurs throughout Texas, while Le Conte's free-tailed bat is resident and typically found in east Texas (Davis 1974; Schmidly 1977). I did not attempt to distinguish between subspecies during my study, so it could be argued bats at my sites are Le Conte's free-tailed bats that are expanding their range westward from east Texas. However, Le Conte's are known to form much smaller colonies, typically in man-made structures such as buildings (McCracken 2003). McCracken and Gassel (1997) suggested genetic differences between subspecies are within range of what would be expected at the species level. It should be determined which subspecies is overwintering in central Texas. If it is concluded they are *T. b. mexicana* this could give additional support to claims that there is not a need for subspecies classification. Brazilian free-tailed bats display a wide range of behaviors (McCracken and Gassel 1997); therefore, distinguishing subspecies based on behavioral differences may be a specious conclusion.

It is not clear at this time if free-tailed bats present during winter are summer residents of the respective roost sites, or northern migrants that are short stopping historical migration routes into Mexico (Geluso 2008). In either case, shifts in winter range of free-tailed bats could be related to their prey species. Free-tailed bats primary food sources during summer in central Texas are the corn earworm (*Helicoverpa zea*) and fall armyworm (*Spodoptera frugiperda*) moths. Both are facultative migratory insects considered major crop pests in Texas (Lee and McCracken 2002; Whitaker et al. 1996). McCracken et al. (2008) suggested distribution of free-tailed bats in Texas can be associated with distributions of these migratory moths. It is possible that as free-tailed bats are expanding their winter range northward, so are their prey species, which could have economic implications for both Texas and Mexico due to their profound impacts on the agricultural industry (Cleveland et al. 2006; Federico et al. 2008).

If global climate change is impacting shifts in overwintering range for free-tailed bats, then continued changes in winter distribution can be expected (Popa-Lisseanu and Voigt 2009; Scheel et al. 1996). It is also important to continue monitoring the bat populations as White-Nose Syndrome spreads throughout the United States (Blehert et al. 2009; Cohn 2012). It is not known what impact, if any, White-Nose Syndrome will have on free-tailed bats, but it affects sympatric species (Blehert et al. 2009), so the potential for free-tailed bats to spread the disease is of great concern.

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