



U.S. Fish and Wildlife Service Southeast Region Inventory and Monitoring Branch I&M Interim Report

Bat Species Occurrence and Long-Term Bat Population Monitoring on Refuges Using Acoustical Detection - 2012-2015 Summary

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Abstract: Long-term trend monitoring efforts for bats on National Wildlife Refuges have been prompted by a paucity of significant population information and precipitous declines in many bat species across the eastern United States. Acoustical detection of search-phase echolocations of bats provides an efficient means to identify bats to species without capture. We provide preliminary summary data for bat species richness and relative abundance using acoustical detection from road-based transects at 56 National Wildlife Refuges and 2 Ecological Field Offices across USFWS Regions 2, 3, and 4 from 2012-2015. We detected 13 species of bats, with species richness varying considerably (1-12) and more northern locales demonstrating higher richness. Bat relative abundance (detections/mile) varied among species and sites and may be influenced by habitat characteristics along transects. The most commonly detected species were tricolored bat, eastern red bat, and evening bat. We conducted prospective power analysis for two widely distributed species which supported the current annual survey design for long-term monitoring. Integration of monitoring data from this project with the North American Bat Program will provide a mechanism to explore larger geographic scale changes in bat species metrics across their range.

INTRODUCTION

Bats are integral to sustaining biodiversity in forested ecosystems of the eastern United States. Nonetheless, we know comparatively little about forest-dwelling bat population abundance, although many species are thought to be declining. In North America, the acute decline in several species is attributed to white-nose syndrome while forest habitat alteration is believed to affect many other species. In general, baseline inventories of bats are lacking on national wildlife

refuges (NWR) though some Comprehensive Conservation Plans and Habitat Management Plans identify bats as species of concern.

Inventory and monitoring of bats is complicated by their small size, mobility, nocturnal habits, and variation in diurnal roosting sites, and among foliage roosting species, limited roost fidelity. Moreover, foliage roosting species are particularly difficult to monitor using direct counts or traditional estimates derived from mark-recapture methods. However, search-phase echolocations of bats provide a means to both inventory and monitor bats because call parameters typically allow species to be uniquely identified. In addition, acoustic surveys of bats along road-based transects provides a robust and efficient method to sample bats across large areas.

We initiated this mobile acoustic bat monitoring project to address four primary objectives regarding the inventory and long-term monitoring of forest-dwelling bats as well as their habitat and landscape associations across multiple scales.

Objectives:

1. Provide a baseline inventory (i.e., occurrence) of bat species on National Wildlife Refuges,
2. Conduct long-term trend monitoring of bat occupancy or abundance at local and landscape scales using a standardized survey protocol,
3. Integrate indices of species abundance and richness with other agencies and partners to support broad-scale Strategic Habitat Conservation Initiatives for bats, and
4. Develop local and landscape-scale bat species habitat associations.

To date, this project has primarily addressed objectives 1 and 2 and is working to identify a cooperator to investigate objective 4. Integration of the data with other partners (objective 3) through the North American Bat Initiative is expected in the near future. In addition to the above objectives, we conducted prospective power analysis and repeated sampling efficacy analysis to evaluate our ability to detect bat population trends (objective 2) based on two years of empirical data.

This report provides a preliminary overview of the results regarding each of the primary project objectives as well as exploratory population trend analysis and sampling design. These results should be viewed with appropriate limitations based only four years of data.

STUDY AREA

We conducted mobile acoustical bat surveys on or adjacent to USFWS field stations, primarily NWRs and Ecological Service Offices in Region 4. In 2013, we expanded the study area to include NWRs in the eastern portion of Region 2 and southern portion of Region 3 with additional NWRs added as sample sites in subsequent years (Figure 1). Sites were not randomly selected; instead, sampling was opportunistic based on the willingness of a field station to conduct the surveys.



Habitat types varied extensively across the study area. Major vegetative classifications consisted of forested wetlands (bottomland hardwood, bald cypress [*Taxodium distichum*], and water tupelo [*Nyssa aquatica*]), upland hardwoods, pine (loblolly [*Pinus taeda*] and shortleaf [*P. echinata*]), and agriculture (cereal grains, cotton, and hay). Survey transects at field stations were constructed across multiple vegetative classifications.

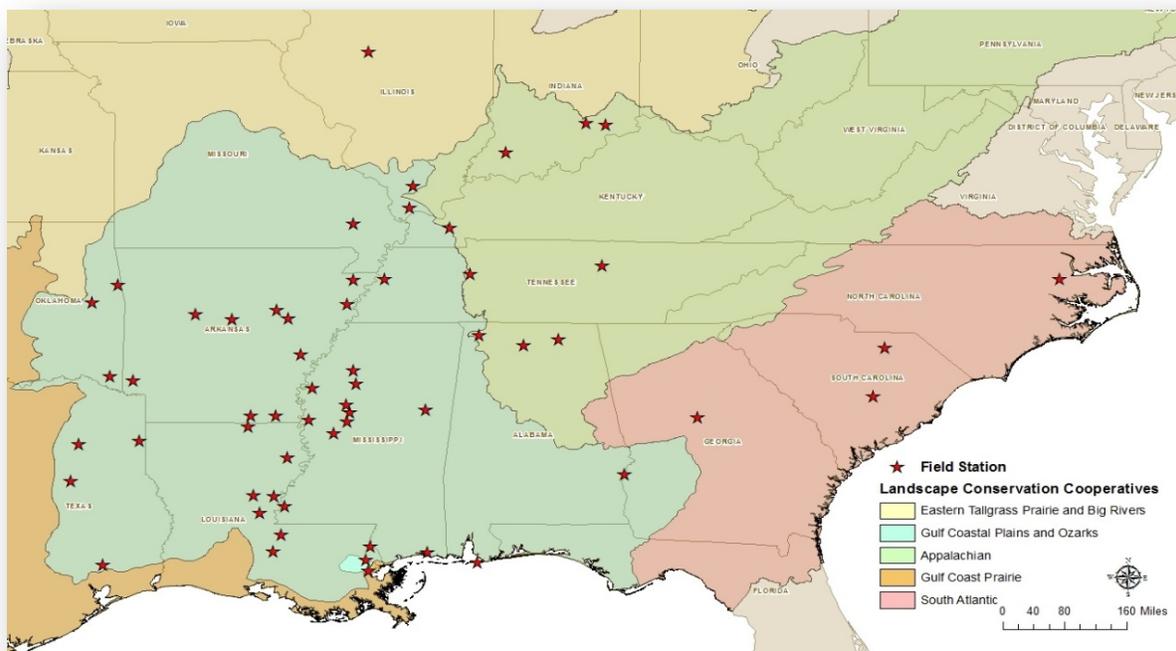


Figure 1. Location of the 58 field stations participating in mobile acoustical bat monitoring during 2012-2015.

METHODS

Measuring the relative abundance and habitat association of bats was done following a standardized mobile acoustical bat monitoring (MABM) survey protocol. We collected search-phase echolocations of bats along road-based transects using an AnaBat SD2 detector (Titley Scientific, Inc.) with a roof-mounted, directional microphone pointed vertically on a vehicle traveling at approximately 20 mph. Transects were constructed around local road infrastructure and based on factors of accessibility, safety, and annual repeatability. Transects were non-random and while an attempt was made to superimpose transects across refuge lands many extended along public roads and private land ownerships. Transect length was determined by road constraints with a target length of 20-30 miles. A global positioning system unit attached to the AnaBat detector enabled call locations to be georeferenced. Surveys were designed to be conducted from June 1 – July 15 though survey data collected between May 16 and July 30 were included in the analysis. Survey effort was initially based on repeated sampling of transects 1-3 times in 2012. Annual sampling was reduced to 1-2 surveys during 2013-2015. Bat calls were recorded using zero-crossing analysis techniques to a compact flash card in the AnaBat detector.

Acoustical data were initially filtered through CFCread Storage ZCAIM interface (ver. 4.4n). To standardize the call classification processes, only call sequences consisting of 5 or more calls were analyzed. We treated bat calls separated by greater than one second as unique; these calls were subsequently used to characterize call sequences to species. Search-phase echolocations of bats were auto-classified to species using the BCID Eastern USA software package (Bat Call Identification, ver. 2.7). To minimize false-positive detections, we limited call classifications at a given field station only to those bat species with summer geographic ranges overlapping the field station. We anticipated possible false-negative detections (i.e., the species is present but misclassified) for Brazilian free-tailed bat (*Tadarida brasiliensis*), northern yellow bat (*Lasiurus intermedius*), and Seminole bat (*L. seminolus*) due to the software lacking classifiers for these species. However, only the Seminole bat is expected to occur across most field stations and would likely be classified as an eastern red bat. We expected very infrequent detections of Brazilian free-tailed bat and northern yellow bat which have more restricted ranges or habitat usage.

Species Diversity and Relative Abundance

Bat diversity for each field station was characterized based on naïve species richness, Shannon-Weiner index (H), and Evenness (E). Acoustical detections for each field station and associated transects were pooled across survey years. Relative abundance was described as the average number of bats per mile of transect surveyed. For stations that had multiple transects, a weighted average based on the length of each transect was used to compute the relative abundance metric.

Power Analysis

The ability to detect a change in a population when one exists given a predefined level of precision is paramount to understanding the required sample size and years of surveying effort for effective mobile acoustical bat monitoring. We conducted a prospective power analysis to assess the efficacy of the sampling design and monitoring protocol. We used data from the 2012 and 2013 sampling periods to explore our ability to detect 25% and 50% population declines (effective size) over a 25 year period, corresponding to an annual rate of decline of -1.14% and -2.73%, respectively. These target estimates of effective size were based on the IUCN red-list categories for assigning species vulnerability to extinction. We also assessed a catastrophic population decline of 5% annually which could reflect a crash attributed to white-nose-syndrome. We used data from two species widely distributed among the field stations (99%) but with disparate abundances and coefficient of variation: the tricolored bat (*Perimyotis subflavus*) was detected at 0.66 bats/mile (CV = 0.75), and the big brown bat (*Eptesicus fuscus*) was detected at 0.13 bats/mile (CV = 1.33). Modeling variation in abundance was done through a Monte Carlo simulation approach in a series of generalized linear mixed models using the package *lme4* in program R. Power was evaluated for P = 0.05.

Repeated Sampling Efficacy

Initially, each transect was surveyed 1-3 times annually. Repeated sampling was intended to address concerns of variation in bat detections related to time and the influence of weather and daily bat activity which could reduce precision in population estimates across years. However, if



repeated sampling across the 6-week sampling period results in a violation of the closed population assumption, due for example to the addition of volant pups, biased estimates of relative abundance could result. To assess the influence of repeated annual sampling, we used data from 7 stations that conducted weekly (4 stations) or biweekly (3 stations) surveys from June 1-July 15 in 2013. We restricted analysis to four bats commonly detected across all stations: big brown bat, eastern red bat (*Lasiurus borealis*), evening bat (*Nycticeius humeralis*), and tricolored bat. Relative abundance was modeled for each species and the four-species combined using generalized linear models in the package *lme4* in program R. In 2014 and 2015, we reduced sampling effort to 2 times annually.

RESULTS

Survey Effort

Fifty-eight field stations (56 NWRs and 2 Ecological Services Field Offices) in three FWS administrative regions (2, 3, and 4) across 14 states participated in the survey during 2012-2015 (Table 1). Most field stations were located within the Gulf Coastal Plains and Ozarks Landscape Conservation Cooperative (LCC) but also extended into 4 other LCCs (Figure 1). Additional field stations contributed each year, although the number of field stations able to conduct sampling varied from 39 to 56 annually (Table 2). Field station personnel surveyed 77 transects on 1-14 occasions over the four years of sampling. Of the 58 survey sites involved in the monitoring, only 3 stations (Bayou Savage, Bogue Chitto, and Bon Secour NWRs) have discontinued the survey.

Relative Abundance

We detected 23,162 bat call sequences representing at least 13 species during 2012-2015 (Table 2). Of these, 88% were classified to species, while 2725 calls (12%) were categorized as 'unknown'. In addition, 96% of call sequences were successfully georeferenced along transects. There was a general increase in the number of sites sampled annually but total number of calls detected decreased. The decreased call detection corresponded to the reduction in sampling intensity of 3-6 times from 2012-2013 to two times annually in 2014-2015.

Most classified calls were attributable to tricolored bat, evening bat, and eastern red bat, all of which were detected on more than 97% of transects (Figure 2). Rafinesque's big-eared bat (*Corynorhinus rafinesquii*), northern long-eared bat (*Myotis septentrionalis*), eastern small-footed bat (*M. leibii*), and Ozark big-eared bat (*C. townsendii*) were infrequently detected at less than a third of the relevant transects. As a group, *Myotis* species were detected across most refuges where they potentially exist; however, the relative abundance was quite low (Table 3).

The relative abundance of total bat species combined at a field station varied considerably across the study area (Table 3). Highest abundances were recorded at Bayou Cocodrie NWR, LA (5.06 bats/mile) and Tensas River NWR, LA (5.60 bats/mile). Lowest abundances were observed at Sequoyia NWR., OK (0.55 bats/mile) and Middle Mississippi NWR, IL (0.70 bats/mile).



Species Diversity

Thirteen bats species were detected from at least one field station (Table 3). Bat diversity varied considerably across the study area with naïve species richness highest at Patoka River NWR, IN (12) and lowest at Bayou Sauvage NWR, LA (1). These same two sites also had the highest and lowest Shannon Weiner index of diversity, respectively. Overall, field stations within the central and more northern portions of the study area had higher species richness attributed to greater overall expected species occurrence at those locales (Figure 3). Field stations on the southerly portion of the study area had a potential species richness of only 6 compared to as many as 12 species at more northern locales. Most stations detected greater than 75% of the bat species expected to be present during the survey period for that site (Table 3).

Table 1. Geographic location of USFWS field stations conducting mobile acoustical bat monitoring and number of transects being sampled.

	AL	AR	GA	IL	IN	LA	KY	MO	MS	NC	OK	SC	TN	TX	Total
No Stations	5	10	1	3	3	9	1	1	12	1	3	2	3	4	58
No Transects	7	14	1	4	5	10	1	1	12	2	4	3	5	4	77

Table 2. Summary of sampling effort and number of search-phase call sequences classified to species and georeferenced from 2012-2015.

Survey Year	Number of Field Stations	Number of Bat Calls				Total Calls
		Classified to Species	Classified as Unknown	Georeferenced	Non-Georeferenced	
2012	39	5833	1121	6701	253	6954
2013	41	6365	630	6759	236	6995
2014	56	4227	330	4411	143	4557
2015	48	4013	643	4491	165	4656
Total	58 ^a	20438	2725	22326	800	23162

^a Represents the cumulative number of unique field stations participating during the 4 years of the project.



Table 3. Station-level survey route data, species richness, Shannon diversity index (H), evenness (E), and species-specific and total mean relative abundance (detections/mile) of bat search-phase echolocations from mobile acoustical bat monitoring conducted 2012-2015 (CORA-Rafinesque's big-eared bat, COTN-Ozark big-eared bat, EPFU-Big brown bat, LABO-Eastern red bat, LACI-Hoary bat, MYAU-Southeastern bat, MYGR-Gray bat, MYLE-Eastern small-footed bat, MYLU-Little brown bat, MYSE-Northern long-eared bat, MYSO-Indiana bat, NYHU-Evening bat, PESU-Tricolored).

Field Station	State	# Years	# Survey Nights	Transect Length (mi)	Expected Species Richness ¹	Naïve Species Richness	Shannon Index (H)	Evenness (E _H)	Total Bat Calls	CORA	COTN	EPFU	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYSU	PESU	UNKN	TOTAL
AR ES – Fifty-Six	AR	3	6	18.2	10	6	1.36	0.76	172	0	- ²	0.06	0.35	0.03	0	-	-	0.05	0	0	0.49	0.65	0.01	1.08
AR ES - Millcreek	AR	4	8	20.2	10	6	1.26	0.71	145	0	-	0.07	0.19	0.03	0	-	-	0.02	0	0	0.34	0.25	0.02	0.90
Atchafalaya	LA	2	4	15.3	6	5	1.21	0.75	257	0	-	0.02	1.39	-	0.16	-	-	-	-	-	1.70	0.82	0.11	4.21
Bald Knob	AR	2	4	21.2	10	5	1.24	0.69	101	0	-	0.05	0.21	0	0	-	-	0.05	0	0	0.22	0.66	0.01	1.17
Bayou Cocodrie ³	LA	3	10	25.7	6	6	1.34	0.69	1200	0.01	-	0.12	1.65	-	0.06	-	-	-	-	-	2.03	0.96	0.23	5.06
Bayou Sauvage	LA	1	1	15.9	6	1	Und ⁴	Und ⁴	2	0	-	0.06	0	-	0	-	-	-	-	-	0	0	0.06	0.13
Big Branch Marsh	LA	1	2	21.8	6	4	1.07	0.77	92	0	-	0.25	0.50	-	0	-	-	-	-	-	1.19	0.11	0.05	2.11
Big Lake	AR	3	4	17.9	10	4	1.06	0.65	119	0	-	0	0.20	0	0	-	-	0.03	0	0	0.32	1.06	0.06	1.66
Big Oaks ³	IN	3	9	55.3	9	8	1.72	0.83	758	-	-	0.51	0.27	0.30	-	-	0.01	0.08	0	0.01	0.30	0.19	0.10	0.57
Bogue Chitto	LA	1	1	30.5	6	5	1.32	0.82	49	0	-	0.03	0.30	-	0.03	-	-	-	-	-	0.25	0.20	0	0.80
Bon Secour	AL	1	1	5.83	6	3	1.05	0.96	5	0	-	0.17	0.34	-	0	-	-	-	-	-	0.34	0	0	0.83
Cache River ³	AR	4	7	42.6	10	9	1.07	0.48	429	0.01	-	0.03	0.26	0.01	0.01	-	-	0.07	0	0.01	0.33	1.48	0.04	2.25
Caddo Lake	TX	2	4	32.2	6	5	1.39	0.86	258	0.01	-	0.42	0.37	-	0	-	-	-	-	-	0.58	0.58	0.04	2.00
Carolina Sandhills	SC	4	9	27.5	8	5	1.22	0.76	610	0	-	0.07	0.85	-	0	-	-	0.04	0	-	0.97	0.47	0.07	2.46
Cat Island	LA	2	4	9.51	6	4	1.17	0.85	56	0	-	0.13	0.50	-	0	-	-	-	-	-	0.60	0.13	0.11	1.47
Catahoula	LA	3	6	9.43	6	5	1.22	0.76	162	0	-	0.07	0.88	-	0.02	-	-	-	-	-	0.88	0.94	0.08	2.86
Chickasaw	TN	3	8	30.5	11	8	1.28	0.62	671	0	-	0.02	0.55	0.01	0.01	-	0	0.11	0	0.02	0.74	1.25	0.05	2.75
Clarks River	KY	4	8	32.8	11	10	1.41	0.61	367	0.01	-	0.02	0.38	0.01	0.01	-	0	0.05	0.01	0.01	0.59	0.27	0.06	1.39
Coldwater River	MS	4	10	25.5	6	4	1.08	0.78	380	0	-	0.01	0.29	-	0	-	-	-	-	-	0.55	0.64	0.01	1.49
Crab Orchard ³	IL	3	7	40.1	12	9	1.31	0.63	460	0.01	-	0.10	0.22	0.02	0	0.01	0	0.06	0	0.01	0.31	0.63	0.03	1.77
Cypress Creek	IL	4	8	18.3	12	8	1.58	0.72	199	0	-	0.08	0.33	0.01	0	0.01	0	0.05	0.01	0	0.36	0.44	0.05	1.35
Dahomey	MS	4	10	27.7	7	5	1.09	0.61	201	0	-	0.01	0.14	0	0.01	-	-	-	-	-	0.43	0.11	0.01	0.72
Eufala ³	AL	4	10	15.6	7	7	1.30	0.66	438	0.01	-	0.18	0.55	-	0.01	-	-	0.02	-	-	0.48	1.13	0.15	2.74
Felsenthal	AR	4	12	20.7	7	6	1.62	0.83	722	0	-	0.55	0.44	0.06	0	-	-	-	0.01	-	0.77	1.04	0.10	2.91
Fern Cave	AL	4	8	30.3	11	7	1.24	0.64	626	0	-	0.11	0.44	0.07	0	0.02	-	0.05	0	0	0.30	1.44	0.15	2.58
Hillside	MS	4	8	31.0	6	5	1.16	0.72	327	0	-	0.01	0.26	-	0.02	-	-	-	-	-	0.52	0.48	0.04	1.31
Holla Bend	AR	3	8	24.8	11	7	1.34	0.69	337	0	-	0.05	0.34	0.03	0.01	-	0	0.03	0	0	0.49	0.68	0.07	1.70
Key Cave	AL	4	9	30.0	11	7	1.16	0.60	507	0	-	0.02	0.30	0	0	0.10	-	0.03	0	0.01	0.22	1.11	0.08	1.88
Lake Ophelia	LA	4	8	11.4	7	4	1.13	0.81	256	0	-	0.13	0.65	-	0	-	-	-	0	-	1.49	0.51	0.04	2.82
Little River	OK	2	4	29.9	8	6	1.53	0.85	101	0	-	0.15	0.18	-	0.01	-	-	0.04	0	-	0.16	0.28	0.02	0.84
Little Sandy	TX	2	3	29.6	6	5	1.42	0.88	64	0	-	0.01	0.22	-	0.22	-	-	-	-	-	0.28	0.18	0.01	0.72



Field Station	State	# Years	# Survey Nights	Transect Length (mi)	Expected Species Richness ¹	Naive Species Richness	Shannon Index (H)	Evenness (E _H)	Total Bat Calls	CORA	COTN	EPFU	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYSU	PESU	UNKN	TOTAL
Mathews Brake	MS	4	8	2.8	6	3	0.78	0.71	50	0	-	0.40	-	0	-	-	-	-	-	-	1.82	0.30	0	2.52
Middle MS River	IL	3	6	13.8	11	5	1.27	0.79	58	0	-	0.10	0.05	0.07	0	0	0	0	0	0	0.08	0.39	0.01	0.70
Mingo	MO	4	10	25.4	11	8	1.30	0.62	1090	0	-	0.08	0.80	0.02	0.01	-	0.01	0.15	0	0	1.60	1.51	0.12	4.29
MS Sandhill Crane	MS	2	4	17.4	6	5	1.08	0.60	73	0.01	-	0.02	0.20	-	0	-	-	-	-	-	0.64	0.09	0.04	1.06
Morgan Brake	MS	4	8	15.8	6	4	1.10	0.68	355	0	-	0.02	0.48	-	0	-	-	-	-	-	1.53	0.72	0.06	2.81
Muscatatuck	IN	3	9	23.7	9	9	1.72	0.78	366	0	-	0.50	0.24	0.18	-	-	0.01	0.06	0.01	0.01	0.26	0.40	0.04	1.71
Neches River	TX	2	3	29.8	6	5	1.34	0.83	118	0	-	0.12	0.46	-	0.02	-	-	-	-	-	0.45	0.27	0	1.32
Noxubee ³	MS	4	14	41.5	7	7	1.45	0.74	1210	0.02	-	0.29	0.55	0.02	-	-	-	-	-	-	0.65	0.73	0.08	2.34
Overflow	AR	3	7	38.7	7	7	1.35	0.75	204	0.01	-	0.07	0.20	-	0.01	-	-	-	0.01	-	0.25	0.38	0.03	0.75
Ozark Plateau ³	OK	3	9	81.9	11	10	1.64	0.71	832	-	0.01	0.03	0.21	0.01	-	0.08	-	0.05	0.01	0.01	0.30	0.39	0.06	1.13
Panther Swamp	MS	4	6	29.7	6	5	1.08	0.67	541	0	-	0.01	0.86	-	0.01	-	-	-	-	-	1.42	0.62	0.12	3.03
Patoka River ³	IN	4	8	40.7	11	11	1.67	0.70	439	0.01	-	0.12	0.39	0.05	-	0.01	0.01	0.06	0.01	0.01	0.40	0.44	0.09	1.58
Piedmont	GA	1	2	29.5	10	7	1.25	0.70	145	0	-	0.95	0.47	-	0	-	-	0.02	0	0.02	0.86	0.08	0.05	2.46
Pond Creek	AR	3	7	29.9	9	7	1.29	0.66	589	0	-	0.13	0.61	0.02	0.02	-	-	0.06	0	-	0.47	1.39	0.11	2.81
Roanoke River ³	NC	3	5	13.2	9	7	1.61	0.83	349	0	-	0.15	0.88	-	0.10	-	-	0.23	0.10	-	0.80	0.92	0.15	1.53
Santee ³	SC	4	7	29.2	7	6	1.30	0.73	389	0	-	0.01	0.28	-	0.01	-	-	0.03	-	-	0.24	0.31	0.06	1.80
Sequoyah	OK	2	4	25.5	11	7	1.62	0.83	57	0	-	0.02	0.14	0.02	-	0.02	0	0.04	0	0	0.17	0.13	0.03	0.55
St. Catherine Creek	MS	3	8	20.7	6	4	1.22	0.88	309	0	-	0.10	0.64	-	0	-	-	-	-	-	0.82	0.48	0.10	2.13
Tallahatchie	MS	4	14	30.0	7	6	1.12	0.62	793	0	-	0.01	0.43	0.01	0.01	-	-	-	-	-	0.93	0.48	0.03	1.89
Tennessee NWR	TN	4	11	27.6	12	8	1.34	0.65	612	0	-	0.02	0.57	0	0	0.01	0	0.08	0.01	0.01	0.58	0.69	0.07	2.01
TN ES-Bump Mills	TN	2	2	31.0	12	7	1.59	0.76	183	0	-	0	0.48	0	0.03	0.08	0	0.21	0	0.05	0.56	1.09	0.13	1.67
TN ES-Gap Creek	TN	1	1	27.6	12	8	1.51	0.72	164	0	-	0.18	0.87	0.14	0.03	0.03	0	0.14	0	0	0.58	1.45	0.22	2.64
TN ES-Roaring R.	TN	4	8	24.8	12	9	1.06	0.48	629	0	-	0.04	0.40	0.03	0	0.05	0.01	0.08	0	0.02	0.29	2.13	0.13	3.17
Tensas River	LA	4	9	31.2	6	5	1.14	0.71	1570	0	-	0.23	2.24	-	0.01	-	-	-	-	-	2.23	0.67	0.20	5.60
Trinity River	TX	1	2	23.1	6	5	1.36	0.84	100	0	-	0.30	0.64	-	0.04	-	-	-	-	-	0.55	1.05	0.07	2.16
Upper Ouachita	LA	3	6	22.2	7	4	1.21	0.87	326	0	-	0.12	0.64	-	0	-	-	-	0	-	0.55	1.05	0.07	2.44
Wapanocca	AR	4	7	18.9	10	5	1.08	0.67	254	0	-	0.01	0	-	0	-	-	0.02	0	0	0.84	0.72	0.02	1.92
Wheeler	AL	4	8	30.5	11	10	1.78	0.77	442	0.01	-	0.32	0.04	0.17	0.01	0.38	-	0.06	0	0.04	0.15	0.59	0.04	1.81
White River ³	AR	4	9	23.3	8	6	1.20	0.62	630	0	-	0.02	0.47	0	0.01	-	-	0.09	-	-	0.82	1.68	0.06	3.14
Yazoo	MS	4	7	30.6	6	5	1.03	0.64	496	0	-	0.01	0.66	-	0.01	-	-	-	-	-	1.18	0.34	0.11	2.31

¹Expected species richness is based on range distributions for bats which could occur during the sampling interval (May 16 – July 31) and bat species that can be classified using the software package Bat Call ID. Brazilian free-tailed bat, northern yellow bat, and Seminole bat occur across a portion of the field stations but are not discriminated from other species using the auto-classification software and are not included in the potential species richness total.

²-(Dash) denotes that the species was not considered to occur at that location during the sampling period.

³Data from multiple transects for the field station were combined. Weighted means were generated for relative abundance when the number of survey nights differed within a year.

⁴Species diversity and evenness are undefined for sites with only 1 species.



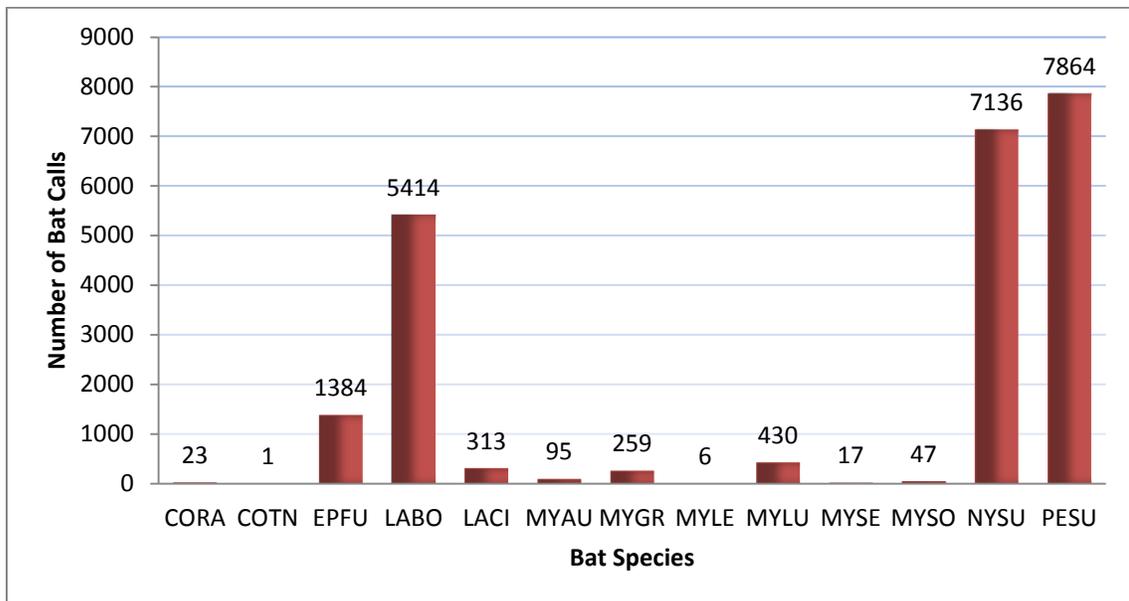


Figure 2. Species classification of bat search-phase echolocations (n=20438) detected at 58 field stations conducting mobile acoustical bat monitoring from 2012-2015. Classifications were based on outputs from BCID Eastern USA software package (ver. 2.7) (CORA-Rafinesque’s big-eared bat, COTN-Ozark big-eared bat, EPFU-Big brown bat, LABO-Eastern red bat, LACI-Hoary bat, MYAU-Southeastern bat, MYGR-Gray bat, MYLE-Eastern small-footed bat, MYLU-Little brown bat, MYSE- Northern long-eared bat, MYSO-Indiana bat, NYHU-Evening bat, PESU-Tricolored).

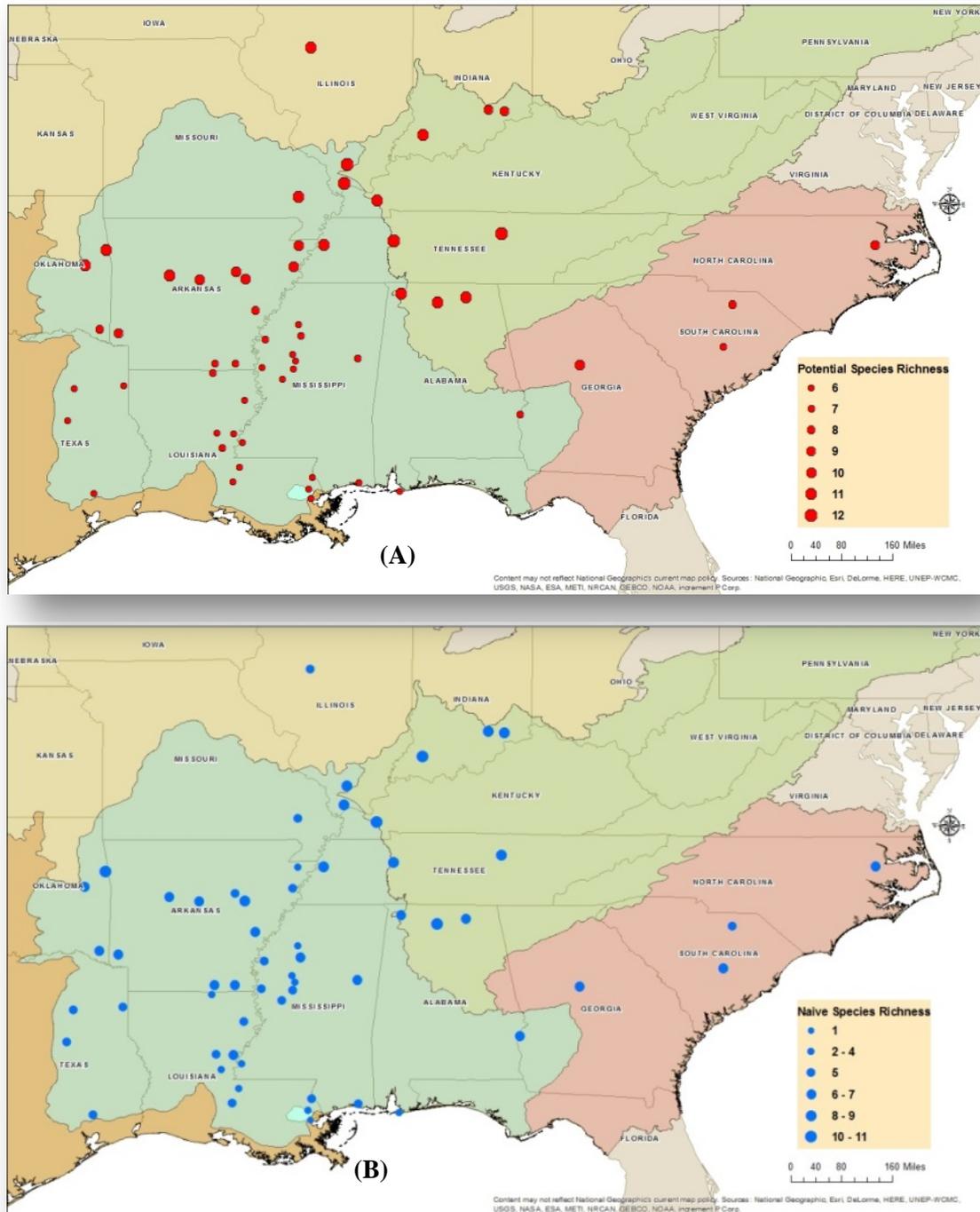


Figure 3. Potential bat species richness (A) and naïve species richness (B) at 58 field stations conducting mobile acoustical bat monitoring from 2012-2015.

Repeated Sampling Efficacy

Relative bat abundance for each of the four species modeled generally increased over the six-week survey period for both the 4-stations doing weekly surveys and to a lesser magnitude for all seven stations (0.3-3.0 % daily). This increase was significant for eastern red bat and tricolored bat ($P < 0.001$). A similar pattern of increased abundance ($P < 0.001$) across sampling periods occurred



when the four bat species were combined into a single data set (Figure 4). Variability on mean estimates increased over time suggesting data collected latter in the survey periods may exhibit greater variability than data collected in the early season.

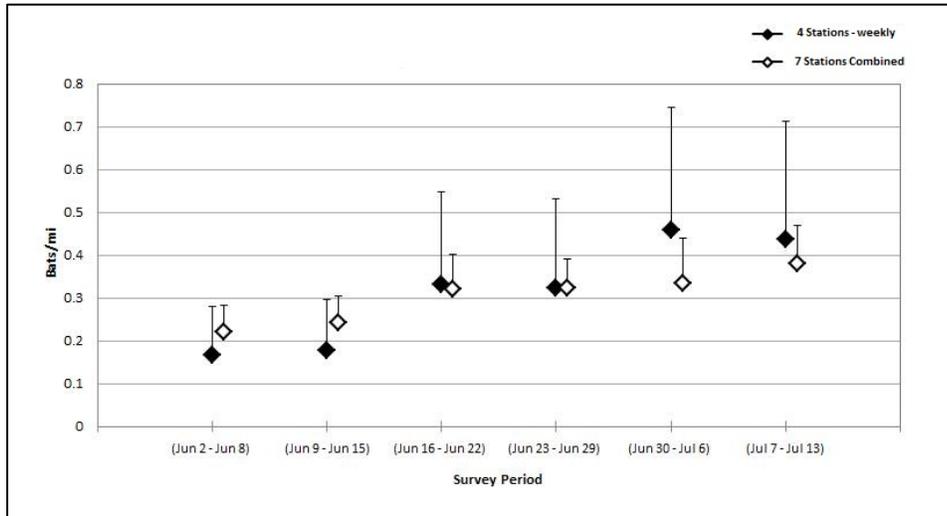


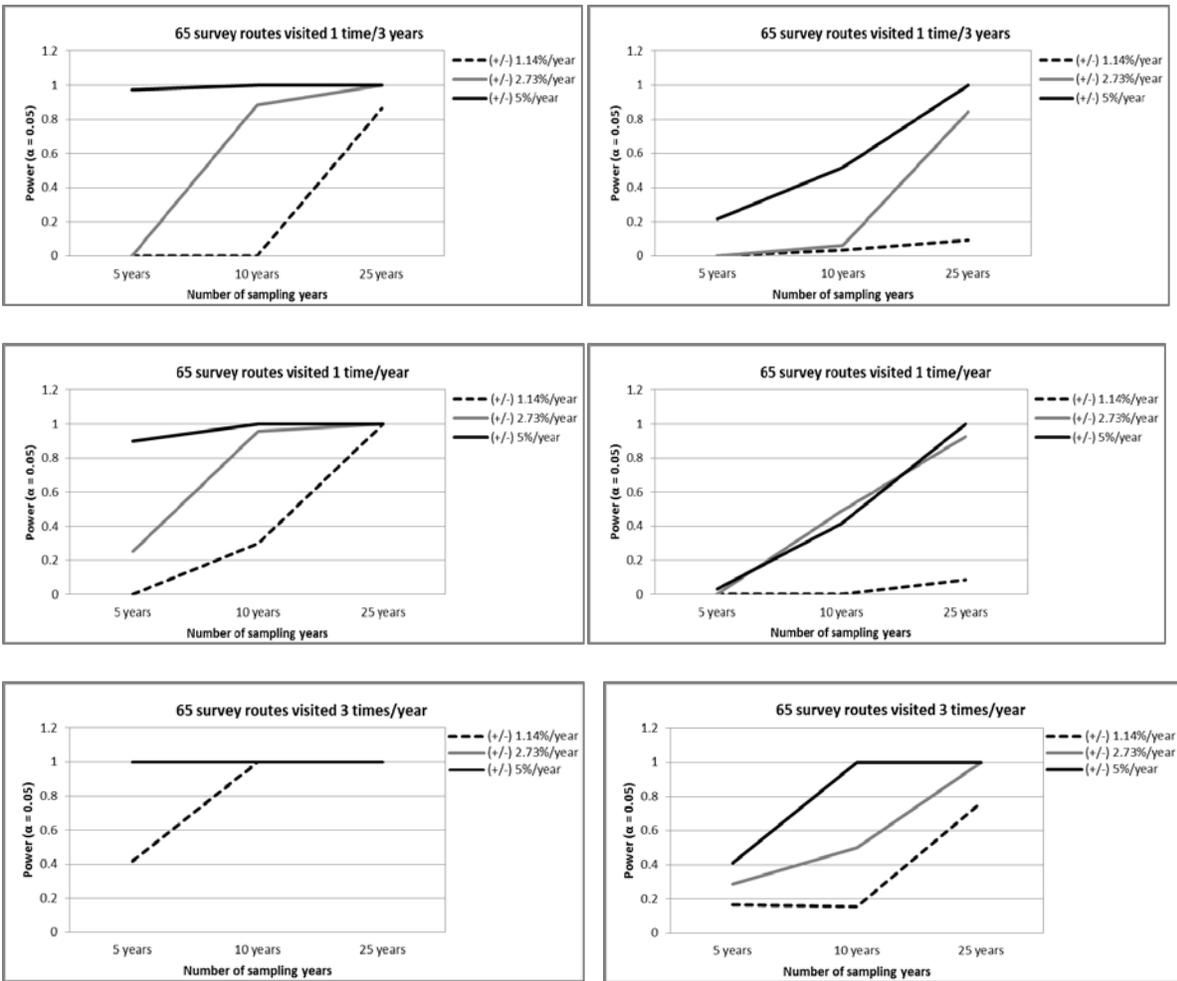
Figure 4. Relative abundance (bat detections/mi \pm 95% confidence interval) of big brown bat, eastern red bat, evening bat, and tricolored bat combined by weekly survey period for four stations doing weekly surveys and three additional stations combined doing biweekly sampling across a six-week period in 2013.

Power Analysis

Tricolored Bat: Power to detect a relatively small population decline (25% decline over 25 years, -1.14% per year) in relative abundance of tricolored bats is <30% with 10 years of sampling across 65 transects (Figure 5). A change in sampling to every third year would have very poor power until >20 years of sampling. However, power exceeds 85% if sites are surveyed three times annually over a 10-year period. The power to detect a 50% decline over 25 years (-2.73% per year) is not probable with only 5 years of monitoring but improves to >75% after 10 years of monitoring. A sudden population crash (-5% per year) can be detected with a probability of 90% and 100% after 5 and 10 years of sampling, respectively.

Big Brown Bat: Power to detect a 1.14% annual decline in relative abundance of big brown bat is very poor unless 65 transects were sampled 3 times yearly for a 25 year period (Figure 5). Sufficient power to detect a more precipitous decline of 50% over 25 year will still require 25 years of monitoring to obtain power >80%. The power to detect a sudden population crash (-5% over 25 years) is 0% at 5 years but improves to near 100% if 65 transects are surveyed 3 times annually over 10 years.





Tricolored Bat

Big Brown Bat

Figure 5. Simulated power ($\alpha = 0.05$) to detect a 1.14%, 2.73%, and 5% change in tricolored bat and big brown bat mean relative abundance when 65 survey transects are sampled 3 times per year (bottom), 1 time per year (middle), and 1 time every 3 years (top) estimated from generalized linear mixed regression models in package *lme4* in program R.

DISCUSSION

The project to date has addressed three of the major objectives and also completed preliminary study design analysis to inform future sampling efforts and efficiency. We discuss the major findings of the project below and provide recommendations for future efforts.

Participation in the MABM survey has steadily increased. The first four years of this long-term bat monitoring project have demonstrated its efficacy to develop refuge scale bat inventories based on acoustical detections. Central to this effort has been the reliance the Region 4 Inventory and Monitoring Branch to facilitate data management and analysis. This has allowed more field stations to contribute towards landscape monitoring of bats. The development of mechanisms to centralize data storage and auto-generate annual station-level summary reports has greatly facilitated feedback to the field stations in a timely fashion. As more field stations become involved with this project, it will be important to continue to provide oversight and develop procedures for autonomous data integration, analysis, and report generation.

Species Diversity

The MABM survey detected all 13 bat species expected within the study area capable of auto-classification using BCID software. Species diversity appeared to be limited in more southerly areas as most species of myotis do not occur throughout the southern study area. Species richness based on acoustical detection provides the foundation for initial bat inventory at participating field stations. However, the species inventory was developed along road-based transects and likely does not represent a complete inventory of bats nor a comprehensive understanding of habitat association. Few field stations detected all the expected species at a field station. To accomplish a comprehensive bat species inventory and fully understand habitat associations, more intensive surveys using a combination of sampling methodologies is necessary. In some instances, this will require an acoustical detector to be passively deployed for several nights at targeted locations for species which produce low volume echolocations, or exhibit more restrictive habitat use (e.g., Rafinesque's big-eared bat). Moreover, three bats species (Brazilian free-tailed bat, northern yellow bat, and Seminole bat) cannot be auto-classified using current software programs and we recognize this as a limitation to the survey analysis; validation of these species will need to be done through direct captures (i.e., netting) or visual vetting of the bat calls. Also, the hoary bat and silver-haired bat (*Lasiorycteris noctivagans*) are migrant species throughout most of the field stations in this study; surveys for these two species should be undertaken in September – November or early spring (before May) to detect them within their winter range.

Relative Abundance

An objective of the project is to conduct long-term population monitoring of multiple species of bats at the landscape scale. The initial four years of the survey have provided significant insights into the diversity and relative abundance of bats at the local and regional scales. We expect to conduct an analysis of species trend data after 10 years at which point any major changes (i.e., 25%) in a species population could be detected.



We observed great disparity between individual bat species abundance as well as total bat relative abundance among stations (0.55 – 5.50 bats/mile). There does not appear to be a relationship between potential bat species richness (number of species expected to occur at a station) and the detected relative abundance at a station to explain this difference. Rather, it is hypothesized that this relationship may be influenced by habitat characteristics along transects. Plotting calls along transects overlaid with photos or topographic layers supports the notion that the preponderance of bat calls are located within forested areas or along habitat edges (see example Figure 6). A more comprehensive analysis of the relationship between detections and habitat associations is being pursued.

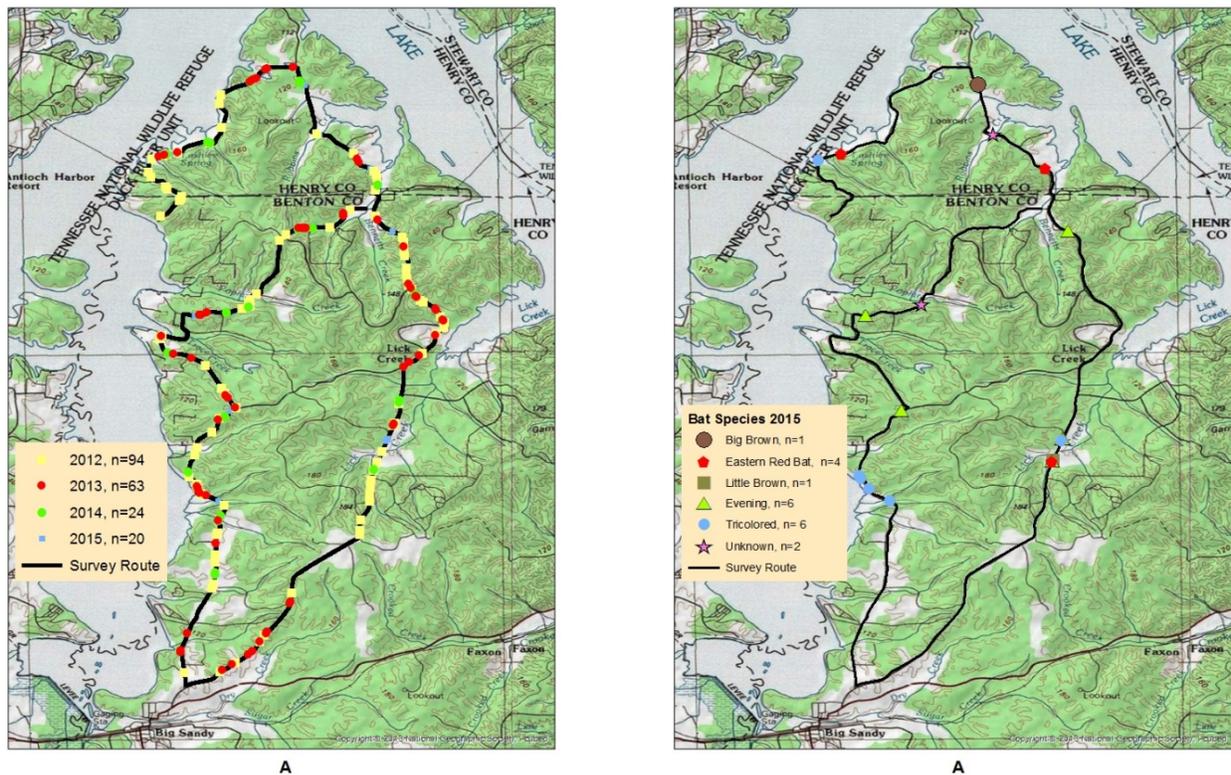


Figure 6. Examples of bat call locations of all bat species detected from 2012-2015 along the survey transect at Tennessee NWR (Left) and location of individual species in 2015 (Right).

Repeated Sampling Efficacy and Power Analysis

The presumption that repeated sampling on a weekly or bi-weekly basis over the six week survey period would increase precision of the mean estimate of bats was not supported. In fact, both the mean estimate of bats as well as variability of that estimate tended to increase. Though we are uncertain as to the mechanism, it may reflect several factors including pups becoming volant during the latter part of the sampling period as well as shifts in bats using roadways for foraging and travel between roosts. Based on these results, it is important to restrict repeated sampling efforts to a narrow sampling period (~ 2weeks) to reduce bias by better meeting the assumption of a closed population and avoiding potential seasonal changes in behavioral patterns. Repeated sampling within a given year also has implications regarding the power to detect changes in bat



populations over a much shorter period. Field stations should make efforts to be certain transects are sampled at least 2 times annually within the prescribed period.

The prospective power analysis of two widely distributed and commonly detected bat species provides information about the ability to detect small as well as catastrophic annual population changes with existing sampling efforts. For tricolored bats, probable detection of small annual population changes (1.14%) during ten years of sampling is very poor unless repeated sampling is done 3 times annually. Presently, efforts to accomplish repeated samples within a 2 week period are difficult and it is unlikely many field stations would be able to increase efforts to a third sample. More significant changes in population ($\geq 50\%$ over 25 years) can be more readily detected with the existing effort and a single annual sample. Efficacy of changing the sampling effort to every third year would substantially reduce our ability to discern changes in tricolored bats. Therefore it is important to retain the existing sampling design.

However, the big brown bat which had much greater variability in relative abundance across the field stations and the ability to detect population declines of 25-50% over 25 years is extremely unlikely within a 10-year sampling period. For even a catastrophic decline of 5% annually, it would take 25 years before the decline was indicated.

The outcome of the power analysis of these two species provides a foundation for considering this protocol design for evaluating long-term population changes of many bat species. A broader and more robust modeling of the 11 other bat species detected in this project is needed to more fully understand which species can be monitored. The MABM survey under existing sampling effort is unlikely to provide an appropriate method to monitor population change of species that have high variability in relative abundance or limited geographic range (e.g., *Myotis spp.*). This underscores the importance of collaborating with other partners using the same monitoring protocol to improve power through increased sampling efforts.

Habitat Associations

Analysis of acoustical bat detections and their corresponding habitat characteristics along each transect may provide important information regarding bat use of different habitat types. In addition, this information may provide an understanding of observed differences in relative abundance of species. We have collected 22,236 geo-referenced bat calls since 2012 which will be used to develop a model of landscape habitat association by bats. Within the context of the roadside sampling design, we recognize that biases may occur and habitat use may not reflect all potential habitats. We are working to identify researchers to assist in modeling these relationships and expect to complete this major objective over the next year.

Integrated Bat Conservation Monitoring

The value of a cooperative bat monitoring program is vested in the sampling effort and geographic scale at which one can examine changes in species abundance. For example, the precipitous decline of many bat species which hibernate in caves in northeastern North America due to white nose-syndrome has been clearly documented through on-going cooperative winter surveys. However, many non-cave winter roosting bats may also be undergoing effects of white-nose



syndrome and other stressors (e.g., habitat alteration, wind energy). The mobile acoustical bat monitoring survey is an approach to identify changes in these other species. While the monitoring of bats on NWRs is important to understand local scale bat demographics and response to on-going management, a broader contribution of the data exists by combining survey efforts across a larger geographic scale (e.g., LCC) to understand changes of species throughout their range. We are exploring mechanism to integrate the on-going data from this survey with the North American Bat Initiative. This national bat inventory and monitoring effort uses the same mobile sampling methodology to derive relative abundance of bats. This increased sampling effort should substantially improve the ability to detect changes in bats at finer scales of reference and in a shorter period of sampling. Data submission and oversight for integration with North American Bat Initiative would be done through the Region 4, NWRS Inventory & Monitoring Branch.

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