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TECHNICAL MEMORANDUM

Date: November 2020

To: Christopher Lewis, Courtney Arthur, Industrial Economics, Inc.

From: Kevin Lager Murray, Western EcoSystems Technology, Inc. (WEST)

Subject: Sauget Acoustic Analysis

Background

Industrial Economics, Inc. contracted Western EcoSystems Technology, Inc. (WEST) to analyze bat acoustic data associated with the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois. Acoustic analyses entailed review of bat call data recorded in 2018 with an emphasis on five species: gray bat (*Myotis grisescens*, federally and state-endangered), Indiana bat (*Myotis sodalis*, federally and state-endangered), northern long-eared bat (*Myotis septentrionalis*, federally and state-threatened), little brown bat (*Myotis lucifugus*), and tri-colored bat (*Perimyotis subflavus*). WEST used both quantitative (Kaleidoscope Pro Software) and qualitative methods to analyze the call data. The acoustic dataset included call files from six sites recorded from summer through fall of 2018.

Methods

The objective of the WEST call analysis was to review acoustic data from 2018 to determine if the echolocation calls of the five target bat species were present within the Project dataset. The five target species were the gray bat (MYGR), little brown bat (MYLU), northern long-eared bat (MYSE), Indiana bat (MYSO) and tri-colored bat (PESU). The acoustic analysis dataset contained all call files from six sites (Ag Field, Arsenal Island, Borrow Pit Lake, Dead Creek, Site P and Site Q) recorded in 2018.

Automated Call Identification (Kaleidoscope)

Bat calls were initially identified using the USFWS-approved automated identification program Kaleidoscope Pro (version 5.1.0; Wildlife Acoustics Inc.). The Bats of North America classifier 5.1.0 was used within Kaleidoscope. We used the Bats of Illinois (Feldhamer et al. 2015) and the USFWS Environmental Conservation Online System (https://ecos.fws.gov/ecp) to determine which bat species were likely to occur in St. Clair County, Illinois. We included the following species in the Kaleidoscope model: big brown bat (*Eptesicus fuscus*), silver-haired bat

(*Lasionycteris noctivagans*), eastern red bat (*Lasiurus borealis*), hoary bat (*Lasiurus cinereus*), MYGR, MYLU, MYSO, MYSE, evening bat (*Nycticeius humeralis*), and PESU. Minimum number of pulses was set to "2" (default) and the bat classifier setting was set to "0" Balanced (Neutral)" (USGS 2019). Defaults were used for all other software settings. The Disperse command in Analook software (version 4.4a) was used to separate call files from each site and survey night into separate folders within each station based upon the Kaleidoscope identification and label. Follow-up qualitative call analysis (i.e. manual vetting) was conducted for all call files identified as MYGR, MYLU, MYSE, MYSO or PESU by Kaleidoscope. In addition, files that Kaleidoscope could not identify (labeled as NoID) were reviewed.

Follow-up Qualitative Review

Qualitative call analysis involves visual review and identification of bat echolocation calls by an acoustic analyst based on comparison of qualitative and quantitative call characteristics to calls in a known call library. Call characteristics (e.g. minimum frequency, maximum frequency, minimum slope) and sequence characteristics (consistency of minimum frequency and call shape) are used to assign species identifications to unknown calls. Qualitative call analysis was conducted by an experienced acoustic analyst (Dr. K. L. Murray). The WEST acoustic analyst used Analook software to conduct qualitative call analysis for each site. All files within the Kaleidoscope target species folders were identified and labeled as the appropriate species, species group, or as unknown. Additionally, the acoustic analyst reviewed all files in no identification folders (NoID) and identified and labeled any target species that were observed within these folders. Calls determined to be non-target species were not identified and the original NoID label was retained for these files. Files that Kaleidoscope identified as non-target species or as noise files were not reviewed or labeled. Bat calls were identified using a known call library and published echolocation call parameters for bat species in the eastern U.S. (Murray et al. 2001 [pg. 732], Britzke et al. 2011 [Table S1], USFWS 2019 [pg. 9]).

Foraging Call Analysis

All calls identified in qualitative analysis were examined for evidence of foraging calls. Bats use a progression of call types while foraging: search-phase calls while searching for prey items, approach-phase calls when they detect and approach prey items and terminal-phase or feeding buzz calls just prior to capturing prey items in their mouth, wing or tail membrane. The presence of approach-phase and/or feeding buzz calls are evidence that bats are actively detecting and pursuing prey items in a given area. Bat calls files with approach-phase calls, feeding buzz calls, or both were considered to be foraging calls and were labeled as "Approach". Specifically, foraging calls were defined as call sequences that exhibited a visually perceptible increase in pulse repetition rate in which the time between successive pulses eventually decreased to 65 milliseconds (ms) or less. Call files were coded as foraging or non-foraging. We did not quantify the number of individual foraging sequences within a single call file.

Results

Kaleidoscope Pro analyzed 455,695 call files from six sites (Table 1). Acoustic survey start dates were May 23, 2018 for each site but end dates varied, ranging from October 11 to November 23,

2018. Kaleidoscope Pro identified 300,348 bat calls to species. A total of 5,716 files were identified as Myotis calls or tri-colored bat calls (PESU) and these files were qualitatively identified in a follow-up analysis. All 73,010 NoID files were also reviewed by qualitative analysis. NoID files contain either noise that the software cannot distinguish from bat calls or bat calls that the software cannot identify with statistical confidence.

Table 1. Summary of the acoustic dataset analyzed with Kaleidoscope Pro for the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois. Follow-up qualitative analysis was conducted on all calls identified by Kaleidoscope Pro as Myotis species, tri-colored bats (PESU) or No Identification (NoID) (in bold).

Site ID	Survey Dates	Total Files	Bat Calls Identified	Myotis	PESU	NoID	Noise
Ag Field	May 23-Oct. 13	135,713	83,545	**627**	**660**	**26,793**	25,375
Arsenal Island	May 23-Oct. 11	100,330	65,775	**1,530**	**293**	**15,847**	18,708
Borrow Pit Lake	May 23-Nov. 17	70,299	47,413	**452**	**221**	**11,632**	11,254
Dead Creek	May 23-Nov. 22	75,230	50,096	**695**	**448**	**10,376**	14,758
Site P	May 23-Nov. 23	29,116	18,610	**186**	**141**	**2,978**	7,528
Site Q	May 23-Oct. 27	45,007	34,909	**247**	**216**	**5,384**	4,714
Total		455,695	300,348	**3,737**	**1,979**	**73,010**	82,337

Kaleidoscope analysis indicated that low frequency (LF) bat species including big brown bats, hoary bats, and silver-haired bats, respectively, had the highest activity rates (Table 2). Low frequency bats species are defined as species with minimum call frequency less than 30 kHz. Together these three species accounted for 93.4% of total bat activity recorded with big browns calls making up 71.3% of total bat activity. According to Kaleidoscope, the four *Myotis* species accounted for 1.2% of bat activity (3,737 calls). However, follow-up qualitative analysis indicated that most of the calls identified as *Myotis* species by Kaleidoscope were misidentifications (Tables 2 and 3). Only 23 calls were identified as *Myotis* species by qualitative analysis (12 MYGR, 10 MYLU, and 1 MYSO). Kaleidoscope identified 1,979 calls as tri-colored bats whereas qualitative analysis identified 1,324 calls as this species (Table 2). A more detailed comparison of Kaleidoscope and qualitative analyses for each target species is provided below.

We used follow-up qualitative analysis to review a total of 78,726 call files (17.3% of the dataset) including 3,737 potential *Myotis* species files, 1,979 potential PESU files, and 73,010 NoID files. We identified 5,901 call files which included the 5,716 potential *Myotis* and PESU calls plus an additional 185 reclassified NoID files. The remaining 72,825 files were reviewed but no target species calls were observed. So, the original Kaleidoscope NoID labels of these files were retained. Most of the 5,901 identified calls (76.9%) were reclassified as either eastern red bats

(3,530 calls), high-frequency (HF) unknowns (900 calls) or Noise (106 calls). However, we did identify 1,324 calls (22.4%) as tri-colored bats and another 23 calls (< 0.1%) as one of three species of Myotis (Table 3).

Gray bats were identified in low numbers at all six survey sites (Tables 3 and 4) ranging from 1 to 4 calls per site. Nearly all gray bat calls were recorded in the early and late fall (Table 5). While Kaleidoscope identified 1,104 files as potential gray bats, nearly all of these files were reclassified. Calls identified as MYGR by Kaleidoscope were reclassified as eastern red bats (651 calls), HF unknowns (301 calls), tri-colored bats (116 calls), and Noise (24 calls). The Kaleidoscope identifications of 12 call files as gray bats were confirmed by qualitative analysis.

We identified 10 calls as little brown bats (Table 3). Of these 10 MYLU calls, five were identified as MYLU, four as MYSO and one as NoID by Kaleidoscope. MYLU calls were identified at four survey sites (Ag Field, Arsenal Island, Borrow Pit Lake, and Site P) ranging from 1 to 5 calls identified per site (Tables 3 and 4). All of the MYLU calls were identified in the summer or early fall (Table 5). Kaleidoscope identified 2,567 files as potential MYLU calls but nearly all of these files were reclassified by qualitative analysis. Calls identified as MYLU by Kaleidoscope were reclassified as eastern red bats (2,256 calls), HF unknowns (296 calls), Noise (8 calls) and big brown bats (2 calls).

We did not identify any calls as northern long-eared bats (Table 3). Kaleidoscope identified 27 files as potential MYSE calls but all of these files were reclassified by qualitative analysis. Calls identified as MYSE by Kaleidoscope were reclassified as big brown bats (14 calls), HF unknowns (12 calls) and Noise (1 calls).

We identified only one call as an Indiana bat (Table 3). This call was identified at Site P on the night of September 2, 2018 (Table 5). Kaleidoscope identified 39 files as potential MYSO calls but all but one of these files were reclassified by qualitative analysis. Calls identified as MYSO by Kaleidoscope were reclassified as eastern red bats (21 calls), HF unknowns (13 calls), and little brown bats (4 calls).

Tri-colored bats were the most commonly recorded target species at the site. PESU calls were identified at all six survey sites (Tables 3 and 4) ranging from 25 to 648 calls per site for a total of 1,324 PESU calls. The majority of PESU calls were recorded in the summer but a substantial number of calls were also recorded in the early fall (Table 5). Kaleidoscope identified 1,979 files as potential tri-colored bat and many of these identifications were confirmed by qualitative analysis (1,028). The remainder were reclassified as eastern red bats (602 calls), HF unknowns (276 calls), and Noise (73 calls). Also, 180 calls identified as NoID and 116 calls identified as MYGR by Kaleidoscope were reclassified as tri-colored bats by qualitative analysis.

We identified 569 foraging call files indicating that bats at all six surveys sites were actively foraging (Table 6). Foraging calls were observed for PESU, LABO, MYLU, EPFU, and NYHU. No foraging calls were observed for MYGR, MYSE or MYSO. In addition, foraging calls were observed for 13 call files identified as HF unknowns. Approximately 10% of call files identified by

qualitative analysis contained foraging calls. The Ag Field site had the highest percentage of foraging calls and Site P had the lowest percentage of foraging calls (Table 6). Foraging calls were most commonly observed in tri-colored bat call sequences with a total of 333 foraging calls or approximately 25% of PESU files identified by qualitative analysis.

The results of our acoustic analyses indicate that gray bats (federally and state-endangered species) and little brown bats are present within the site in relatively low numbers. Tri-colored bat calls, including foraging calls, were commonly recorded at all six survey sites. Only one Indiana bat call (federally and state-endangered species) was identified within the site. Because the uncertainty associated with Indiana bat call identification, the presence of a single call is not definitive evidence of presence but rather an indication that presence of this species is possible within the site. No northern long-eared bat calls were identified and therefore, there was no acoustic evidence that this species was present during bat surveys. Our analysis also indicated that eastern red bats, big brown bats, and evening bats were present within the site but these species were not the focus of the analysis.

Literature Cited

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US Fish and Wildlife Service (USFWS). 2019. Evaluating Acoustic Bat Surveys for ESA Compliance. Course Guidebook. May 2019. National Conservtion and Training Center.

Table 2. Bat calls identified by Kaleidoscope Pro® Version 5.1.0 for the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois. Numbers in parentheses indicate number of MYGR, MYLU, MYSE, MYSO and PESU calls identified by follow-up qualitative analysis.

Site ID	EPFU	LABO	LACI	LANO	MYGR	MYLU	MYSE	MYSO	NYHU	PESU
Ag Field	62,677	1,331	13,930	2,966	282 (1)	338 (2)	3 (0)	4 (0)	1,354	660 (648)
Arsenal Island	51,988	1,127	8,450	1,987	304 (1)	1194 (1)	16 (0)	16 (0)	400	293 (179)
Borrow Pit Lake	27,260	2,787	12,728	2,894	145 (2)	298 (2)	0 (0)	9 (0)	1,071	221 (136)
Dead Creek	37,411	1,769	7,666	1,279	146 (3)	541 (0)	6 (0)	2 (0)	828	448 (246)
Site P	14,271	1,182	1,098	1,015	101 (4)	76 (5)	1 (0)	8 (1)	717	141 (25)
Site Q	20,761	861	10,969	1,148	126 (1)	120 (0)	1 (0)	0 (0)	707	216 (90)
Overall Total	**214,368** **71.4%**	**9,057** **3.0%**	**54,841** **18.3%**	**11,289** **3.8%**	**1,104 (12)** **0.4%**	**2,567 (10)** **0.9%**	**27 (0)** **< 0.1%**	**39 (1)** **< 0.1%**	**5,077** **1.7%**	**1,979 (1,324)** **0.7%**

EPFU = big brown bat (*Eptesicus fuscus*); LABO = eastern red bat (*Lasiurus borealis*); LACI = hoary bat (*Lasiurus cinereus*); LANO = silver-haired bat (*Lasionycteris noctivagans*); MYGR = gray bat (*Myotis grisescens*); MYLU = little brown bat (*Myotis lucifugus*); MYSE = northern long-eared bat (*Myotis septentrionalis*); MYSO = Indiana bat (*Myotis sodalis*); NYHU = evening bat (*Nycticeius humeralis*); PESU = tri-colored bat (*Perimyotis subflavus*).

Table 3. Bat calls identified by follow-up qualitative analysis for the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois.

Site ID	EPFU	LABO	MYGR	MYLU	MYSE	MYSO	NYHU	PESU	HF	Noise	Total Calls Identified
Ag Field	0	507	1	2	0	0	0	648	182	49	1,389
Arsenal Island	11	1,400	1	1	0	0	0	179	241	30	1,863
Borrow Pit Lake	0	416	2	2	0	0	0	136	125	6	687
Dead Creek	5	723	3	0	0	0	0	246	171	12	1,160
Site P	0	194	4	5	0	1	0	25	97	4	330
Site Q	0	290	1	0	0	0	2	90	84	5	472
Overall Total	**16** **0.3%**	**3,530** **59.8%**	**12** **0.2%**	**10** **0.2%**	**0** **0.0%**	**1** **< 0.1%**	**2** **< 0.1%**	**1,324** **22.4%**	**900** **15.3%**	**106** **1.8%**	**5,901**

EPFU = big brown bat (*Eptesicus fuscus*); LABO = eastern red bat (*Lasiurus borealis*); MYGR = gray bat (*Myotis grisescens*); MYLU = little brown bat (*Myotis lucifugus*); MYSE = northern long-eared bat (*Myotis septentrionalis*); MYSO = Indiana bat (*Myotis sodalis*); NYHU = evening bat (*Nycticeius humeralis*); PESU = tri-colored bat (*Perimyotis subflavus*); HF = high-frequency unknown.

Table 4. Summary of qualitative identifications of five target species for the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois.

Bat Species	Common Name	Number of Sites	Number of Calls
Myotis grisescens	gray bat	6 of 6	12
Myotis lucifugus	little brown bat	4 of 6	10
Myotis septentrionalis	northern long-eared bat	0 of 6	0
Myotis sodalis	Indiana bat	1 of 6	1
Perimyotis subflavus	tri-colored bat	6 of 6	1,324

Table 5. Timing of target species recordings for the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois.

Bat Species	Summer (May 23 to July 31)	Early Fall (Aug. 1 to Sept. 30)	Late Fall (Oct. 1 to Nov. 23)
gray bat	1 call June 7	**7 calls** Aug. 25 to Sept. 29	4 calls Oct. 8 to Oct. 15
little brown bat	4 calls May 24 to June 22	**6 calls** Aug. 11 to Sept. 14	0 calls
Indiana bat	0 calls	**1 call** Sept. 2	0 calls
tri-colored bat	**828 calls** May 23 to July 31	457 calls Aug. 1 to Sept. 30	39 calls Oct. 1 to Nov. 21

Table 6. Number and percentage of foraging calls identified by qualitative analysis for the Sauget Industrial Corridor (SIC) natural resource damage assessment (NRDA) in St. Clair County, Illinois.

Site	EPFU	LABO	MYLU	NYHU	PESU	HF Unknown	Summary
Ag Field	na	23 (4.5%)	1 (50.0%)	na	192 (29.6%)	2 (1.1%)	218 of 1,389 calls (15.7%)
Arsenal Island	2 (18.2%)	111 (7.9%)	0 (0.0%)	na	15 (8.4%)	5 (2.1%)	133 of 1,863 calls (7.1%)
Borrow Pit Lake	na	21 (5.0%)	0 (0.0%)	na	29 (21.3%)	2 (1.6%)	52 of 687 calls (7.6%)
Dead Creek	0 (0.0%)	44 (6.1%)	na	na	81 (32.9%)	2 (1.2%)	127 of 1,160 calls (10.9%)
Site P	na	3 (1.5%)	3 (60.0%)	na	0 (0.0%)	2 (2.1%)	8 of 330 calls (2.4%)
Site Q	na	14 (4.8%)	na	1 (50.0%)	16 (17.8%)	0 (0.0%)	31 of 472 calls (6.6%)
Total	**2 (12.5%)**	**216 (6.1%)**	**4 (40.0%)**	**1 (50.0%)**	**333 (25.2%)**	**13 (1.4%)**	**569 of 5,901 calls (9.6%)**

EPFU = big brown bat (*Eptesicus fuscus*); LABO = eastern red bat (*Lasiurus borealis*); MYLU = little brown bat (*Myotis lucifugus*); NYHU = evening bat (*Nycticeius humeralis*); PESU = tri-colored bat (*Perimyotis subflavus*); HF Unknown = high-frequency unknown