Bat Presence, and Evaluation of a Lower-Cost Passive Acoustic Monitoring Device, at a Passive Treatment System for Acid Mine Drainage

U.S. Department of the Interior
Office of Surface Mining Reclamation and Enforcement

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Acknowledgements. The Author would like to thank AmeriCorps volunteer Cassandra Krul for assistance in deployment and maintenance of detectors, OSM hydrologist Larry Shores for assistance in maintenance of detectors, OSM ecologist Craig Walker for assistance in detector maintenance and review of this report, and University of Pittsburgh Assistant Professor Justin Kitzes for equipment loan and review of the report.

Cover. View of passive treatment pond and wetland vegetation within the Flight 93 National Memorial Grounds. Image Credit: Brian Dailey, OSM.

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By Brian Dailey

Technical Study Report

September 24, 2020

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Office of Surface Mining Reclamation and Enforcement

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Bat Presence, and Evaluation of a Lower-Cost Passive Acoustic Monitoring Device, at a Passive Treatment System for Acid Mine Drainage

By Brian Dailey

Executive Summary

This study found that bats are present at ponds/wetlands that treat acid mine drainage (AMD) within the Flight 93 Memorial Grounds and the AudioMoth 1.0.0 capable of recording bat calls for auto-identification with an off-the-shelf classifier software. Based on auto-identified calls from the Wildlife Acoustics SM4BAT-ZC and AudioMoth 1.0.0 detectors it is inferred that six bat species were present during this study across all passive treatment wetlands and five were present at the natural wetland. The greatest species richness of bat calls auto-identified by both detectors was at a pond treating AMD. In general, the AudioMoth had a greater number of auto-identified calls and species identified at each of the deployment locations than the SM4BAT-ZC.

Wildlife Acoustics SM4BAT-ZC and AudioMoth 1.0.0 passive acoustic monitoring devices (detectors) were deployed at four locations, three ponds/wetlands treating AMD and one natural pond fringed with wetland vegetation, within the Flight 93 National Memorial Grounds. The detectors were secured to existing woody vegetation with microphones approximately two meters off the ground and set to record from July 25, 2018 through August 30, 2018. The SM4BAT-ZC recorded data throughout the deployment. The AudioMoth did not record data for all days deployed. The lack of AudioMoth data is attributed to either a malfunction of the detector caused by corrosion, issues with formatting the memory cards, or battery exhaustion. Kaleidoscope Pro Version 5.1.0 was used to parse the continuously recorded WAV files from the AudioMoth to WAV files with a max duration of 15 seconds. Kaleidoscope Pro - Bats of North America 5.1.0 was used to auto-identify SM4BAT-ZC files and parsed AudioMoth WAV files.

Future study of AudioMoth efficacy should focus on a direct comparison of WAV files recorded from full-spectrum recorders. While the results of this study show that the lower-cost AudioMoth 1.0.0 can record ultrasonic frequency calls, and identification is possible using an existing auto-identification software, a direct analysis between detectors may not be drawn due to the difference in recording formats of the two detectors. To confirm species present within the Flight 93 National Memorial Grounds, mist net surveys could be conducted in conjunction with passive acoustic monitoring. Finally, passive treatment systems on a regional scale could be monitored for bat presence along with insect diversity and abundance, water quality parameters, and analysis of metal concentration of captured bats. Evaluating bat presence on a larger scale could provide insight into how bat populations and communities utilize ponds designed for the treatment of AMD across the greater landscape.

Introduction

Mining within the Flight 93 National Memorial Grounds (Grounds) began with surface mining in 1967 and five coal seams were mined. Beginning in the 1990's, three underground mines operated within the Grounds with two mining the Lower Kittanning and one mining the Middle Kittanning coal seams. All mining within the Grounds ceased operation in 2004 (Office of Surface Mining 2015). Mining coal can have associated environmental problems involving the interaction between surface or ground water and acid-forming materials. Acid mine drainage (AMD) is the result of the interaction between acid-forming materials containing sulfide minerals (e.g. pyrite) and oxygen, bacteria, water, or weathering processes, to form acids that may create acid drainage (Brady, Smith, and Schueck 1998; Watzlaf et al. 2004). Mining resulted in AMD at several locations within the Grounds. Left untreated, this AMD may have detrimental effects to downstream aquatic ecosystems and therefore a treatment system has been employed to mitigate the effects.

In general, there are two designs of treatment systems for AMD: active and passive. Active treatment of AMD involves the addition of alkaline chemicals to remove contaminants while passive treatment uses natural chemical and biological processes (Watzlaf et al. 2004). Passive AMD water treatment facilities have been constructed over the past thirty years to treat AMD and over 300 passive treatment systems have been designed and installed in Pennsylvania (Stream Restoration, Inc. & 241 Computer Services 2020). Passive treatment facilities take advantage of the natural chemical and biological processes, such as those occurring in wetlands or associated with limestone, to decrease metal concentrations, neutralize acidity, and minimize detrimental effects to aquatic ecosystems (Watzlaf et al. 2004). Within the Grounds, water from the underground mines is pumped from a well installed into the underground mine pool to the surface where the 775 to 1,200 gallons per minute (GPM) of AMD passes through a series of nine passive treatment ponds and one wetland. Passive acoustic monitors were deployed at three of the passive treatment ponds and one natural pond within the Grounds (Figure 1).

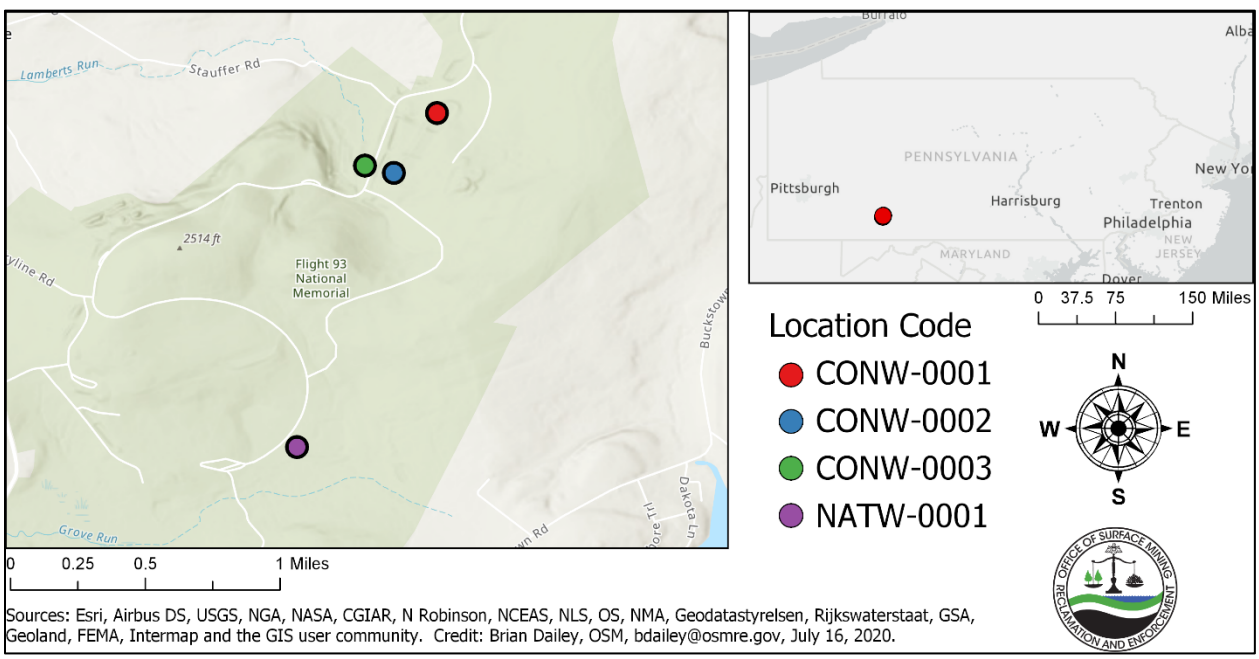

Figure 1. Passive acoustic monitor (Detector) locations within the Flight 93 National Memorial Grounds. CONW locations indicate ponds/wetlands designed to treat AMD. NATW is a pond/wetland not designed to treat AMD.

Passive acoustic monitoring for bat presence involves deploying detectors in areas of suspected bat presence to record ultra-high frequency sound. Detectors typically record files using either heterodyne (e.g. Pettersson D100), frequency division (e.g. Wildlife Acoustics SM4BAT-ZC), or full spectrum (e.g. AudioMoth 1.0.0) outputs (Parsons, Boonman, and Obrist 2000). Heterodyne detectors produce an audible output that allows for real time analysis with no call structure preserved (Parsons, Boonman, and Obrist 2000). Frequency division produces a representation of the sound of the bat with its frequency divided by a constant. Frequency division files require less storage space but do not include all the spectral information. Full spectrum recording produces a file that contains all spectral information including time, frequency, and amplitude. Full spectrum files use considerably more storage space, however, they provide the ability to view intensity and harmonics of a call which may aide in species identification (Browning et al. 2017). Spectral data of frequency division and full spectrum outputs are capable of analysis for species identification via specialized classifier software[1] using either zero-crossing, Fourier analysis, or instantaneous frequency techniques (Parsons, Boonman, and Obrist 2000). Detectors are deployed in locations of suspected bat activity such as forested corridors or adjacent to waterbodies then set to record on a schedule when bats are known to be active, e.g. dusk to dawn, for days or weeks at a time. Detectors are relatively inexpensive and are easily deployed in multiple locations across a landscape. Lower-cost detectors, e.g. AudioMoth, could increase the scale of a study by adding 10 to 20 additional recorders for the same investment as other full spectrum recorders.

Natural freshwater resources such as ponds, wetlands, and streams provide ecosystem services for bats such as drinking or foraging for insects (Butchkoski et al. 2016). Wetlands constructed for mitigation purposes may also provide these services (Maslonek 2009). Like natural freshwater and constructed wetland resources, constructed passive treatment systems may provide similar ecosystem services to bats and may contribute to a wetland habitat network that may be critical to bats (Lookingbill et al. 2010; Sievers et al. 2018). However, because passive treatment systems are designed to capture and store metals at potentially toxic levels to bats it is important to determine if bats are present and if so, how they are using these resources to better understand what the impact of the passive treatment systems may have to bat populations and communities. This study aims to evaluate: 1) bat presence at AMD passive treatment wetlands/ponds within the Grounds, and 2) how a lower-cost acoustic monitoring device functions for this purpose. Results from this study may be used to inform further study of bat use of active and abandoned mine lands, and specifically passive treatment ponds/wetlands.

Methods

Equipment

Audiomoth 1.0.0

The Open Acoustic Devices AudioMoth 1.0.0 is a lower-cost (approximately $43) open-source full-spectrum acoustic recorder capable of recording uncompressed audio into the ultrasonic frequencies (Hill et al. 2018). The AudioMoth 1.0.0 has a built in microelectro-mechanical systems (MEMS) microphone, is powered by three “AA” batteries, and continuously records full spectrum WAV[2] files to a

[1] Examples of analysis software include: BCID Program (Ryan Allen, Bat Call Identification, Inc.), EchoClass (Eric Britzke, ERDC), Kaleidoscope® Pro (Wildlife Acoustics, Inc.), and SonoBat™ (Joe Szewczak).

[2] A usually uncompressed lossless computer file format used for the storage of digital audio data (Merriam-Webster 2015).

microSD card (Rogers, Hill, and Prince 2017; Hill et al. 2018). Recording schedule and device settings are configured via direct connection to a computer running the AudioMoth Configuration App. Alternatively, the recorder may be set to record on a default schedule.

Wildlife Acoustics SM4BAT-ZC

The Wildlife Acoustics SM4BAT-ZC (approximately $650 with external microphone) is a native zero-crossing recorder capable of recording in the ultrasonic frequencies (Wildlife Acoustics 2018). The SM4BAT-ZC is powered by four "D" batteries, equipped with a Wildlife Acoustics SMM-U1 external microphone, and when triggered recorded zero-cross files to a standard SD card. Recording schedule and device settings may be either configured through direct connection via computer running the Song Meter SM4 Configurator or directly on the recorder. For this study the recorder was configured directly on the recorder.

Deployment

Wildlife Acoustics SM4BAT-ZC and AudioMoth 1.0.0 acoustic monitoring devices were deployed at four locations within the Flight 93 National Memorial Grounds, set to record on July 25, 2018, and retrieved on August 30, 2018[3] (Table 1). Figure 2 shows the deployment locations within the Grounds. NATW-0001 is an open water pond fringed by wetland vegetation collecting runoff from the adjacent forested and Grounds lands. CONW-0001 is the fourth settling pond in the series treating AMD within the Grounds and is fringed by sparse wetland and herbaceous vegetation. CONW-0002 is the second settling pond in the series treating AMD within the Grounds and fringed by sparse wetland and herbaceous vegetation. CONW-0003 is the sixth settling pond in the series treating AMD within the Grounds and is dominated by emergent wetland vegetation.

Table 1. Recorder deployment information.

Location Code	AMD Treatment	Latitude	Longitude	Detector Model	Date Deployed	Date Retrieved	Microphone Height (Meters)	Distance to Water (Meters)
NATW-0001	N	40.05156	-78.89588	AudioMoth1.0.0	7/25/2018	8/30/2018	2.1	5.9
NATW-0001	N	40.05156	-78.89588	SM4BAT-ZC	7/25/2018	8/30/2018	2.1	5.9
CONW-0001	Y	40.06949	-78.88835	AudioMoth1.0.0	7/25/2018	8/30/2018	2.6	10.8
CONW-0001	Y	40.06949	-78.88835	SM4BAT-ZC	7/25/2018	8/30/2018	2.6	10.8
CONW-0002	Y	40.06627	-78.89066	AudioMoth1.0.0	7/25/2018	8/30/2018	2.9	12.8
CONW-0002	Y	40.06627	-78.89066	SM4BAT-ZC	7/25/2018	8/30/2018	2.9	12.8
CONW-0003	Y	40.06668	-78.89222	SM4BAT-ZC	7/25/2018	8/30/2018	2.7	15.8
CONW-0003	Y	40.06668	-78.89222	AudioMoth1.0.0	7/30/2018	8/30/2018	2.7	15.8

[3] AudioMoth 1.0.0 was not deployed at FL93-CONW-0003 on July 25, 2018 due to battery/custom programming issues.

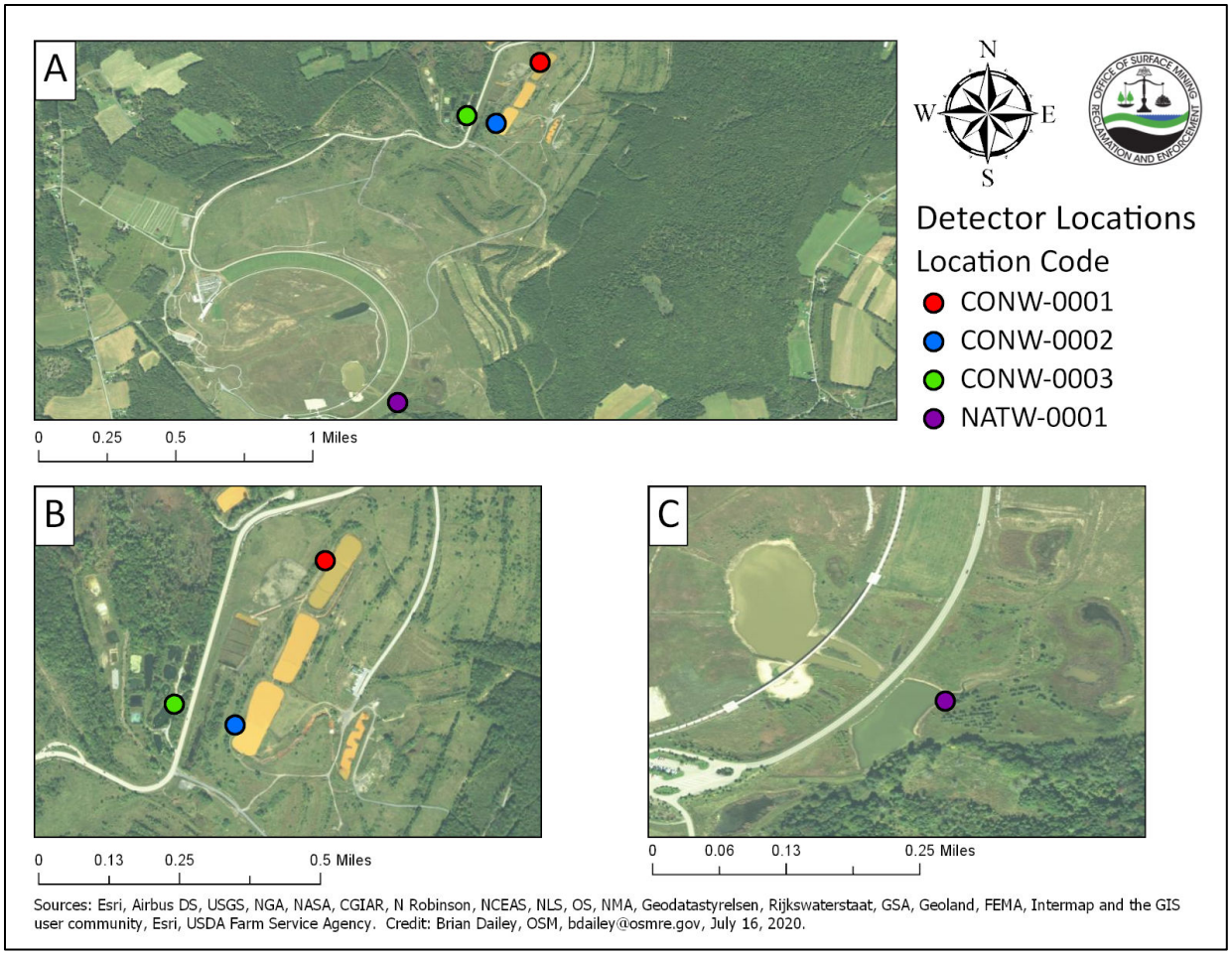

Figure 2. Detector locations within the Grounds. Panel A shows an overview of all locations, Panel B is zoomed into the AMD treatment ponds, and Panel C is zoomed into the natural pond. CONW-0002 is near the beginning of the series of treatment ponds and has the greatest iron precipitation as seen in the aerial imagery. Water flows from CONW-0002 to CONW-0001 and eventually to CONW-0003.

Bungee cords were used to secure the SM4BAT-ZC recorder and microphones to existing woody vegetation with microphones oriented towards the open water. The microphone was extended using a cable and constructed PVC pole from the SM4BAT-ZC recorder body at NATW-0001 and CONW-0002 and attached directly to the recorder body at CONW-0001 and CONW-0003. AudioMoth recorders were placed microphone up, and oriented towards the open water, in a quart Ziploc® freezer bag along with a 75mm x 95mm desiccant pack and secured to existing woody vegetation with zip-ties. Clutter from leaves and trees surrounding all microphones was minimized as much as possible.

Device settings were configured prior to initial deployment using their respective configuration apps to start recording approximately 30 minutes prior to sunset and stop approximately 30 minutes after sunrise. The SM4BAT-ZC automatically started recording earlier and ending later throughout the deployment to account for local changes in sunrise/sunset and the AudioMoth activation start time was manually changed once on August 15. AudioMoth batteries and microSD cards were refreshed and devices reconfigured in the field on four occasions during the deployment: July 30; August 6; August 9; and August 15.

Recording Process Workflow

Recorded WAV files from the AudioMoth were extracted following each refresh and all data from the SM4BAT-ZC were extracted following recorder retrieval and saved to an "Archive" folder for each location and recorder. "Working" folders were created for each location and recorder to preserve the integrity of the original data, in the Archive folders, during processing. North American Bat Monitoring Program (NABat) guidance was followed to rename and process all recorded files using Kaleidoscope Pro Version 5.1.0 (Reichert et al. 2018). AudioMoth files were split to a max duration of 15 seconds to parse bat echolocation calls into individual WAV files from the larger continuously recorded WAV files. Kaleidoscope Pro - Bats of North America 5.1.0 was used to auto-identify SM4BAT-ZC files and parsed AudioMoth WAV files. MS Excel was used for data organization, analysis, and figure generation. Specific information about file processing in Excel is shown in Appendix A.

Results

Figure 3 shows the total days with data recorded by the AudioMoth at each location with all recorders, aside from at CONW-0003 at 31 days, deployed for a total of 36 days[4]. Because the AudioMoth is recording continuously, data should be recorded if deployed. If no data were on the AudioMoth when retrieved, it was due to a malfunction in the device caused either by corrosion, issues with formatting the memory cards, or battery exhaustion. The SM4BAT-ZC recorded calls that were auto-identified by the Kaleidoscope Pro software two sites (CONW-0001 and NATW-0001) while the AudioMoth recorded calls at all four sites (Table 2).

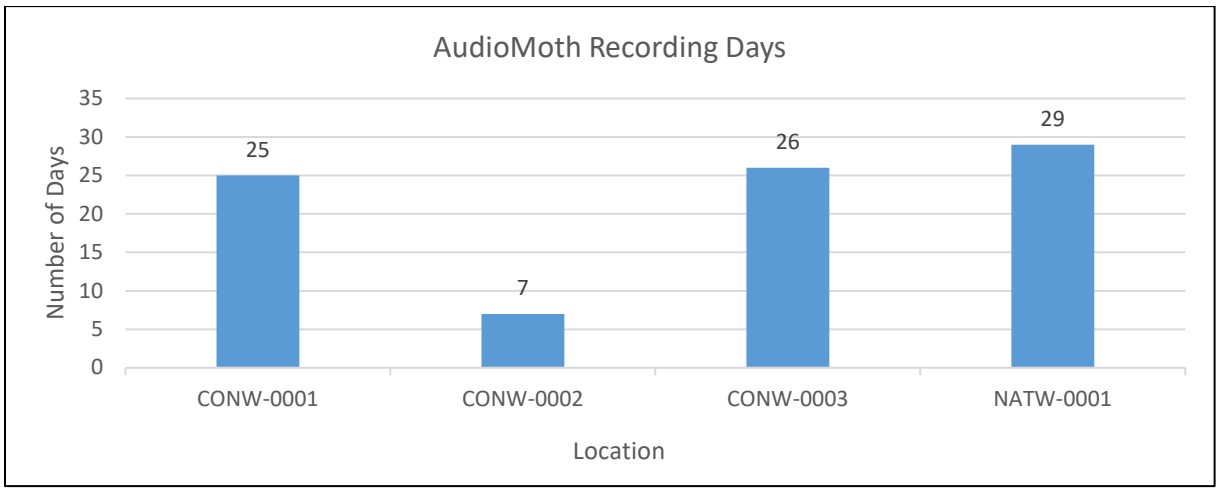

Figure 3. Total number of days with recordings on the AudioMoth at each location. Aside from CONW-0003 (31 days between July 30, 2018 and August 30, 2018) all detectors deployed for a total of 36 days between from July 25, 2018 and August 30, 2018. No data were recorded at any location after August 24, 2018, most likely due to exhausted batteries. A lower number of days with recordings at a location is attributed to either issues with batteries, SD card formatting issues, or recorder malfunction possibly due to corrosion (CONW-0002).

Species Presence

Processed calls were filtered to reduce the total number of auto-identified calls down to those with greater confidence in the identification using best industry practices. Only calls with nine or more pulses

[4] Data was recorded by the SM4BAT-ZC on all days during deployment.

and a match ratio greater than 0.66 were kept for analysis (best practices based on personal communication, Bat Survey Solutions staff, 2018). Table 2 shows the species identified at all locations by both recorders across all nights. At CONW-0001 the SM4BAT-ZC identified four species and the AudioMoth identified five species. The greatest species richness was seen at this location with a total of six species auto-identified. At CONW-0002 no species were auto-identified by the SM4BAT-ZC and AudioMoth files were classified as either Hoary bat or Silver haired bat. The AudioMoth was only functional for a total of seven days at the site and was damaged by corrosion early in the deployment which may have limited the species abundance. However, the SM4BAT-ZC functioned for the entire deployment of 36 days and did not auto-identify any bat species during its deployment. Of the auto-identified calls at CONW-0002 the majority were recorded between 12:00:00 AM and 7:00:00 AM with many calls identified nearly back to back, i.e. parsed at 15 second intervals which may indicate a bat circling the detector which is common behavior of the Hoary bat (Table 3). At CONW-0003 five species were auto-identified from AudioMoth recordings and no calls were identified from SM4BAT-ZC recordings with all calls recorded by the SM4BAT-ZC either auto classified as “NoID” or “Noise”. At NATW-0001 five species were auto-identified from AudioMoth recordings while only three species were identified from SM4BAT-ZC recordings. The Hoary and Silver haired bat were identified at all locations by AudioMoth files.

Table 2. Species listed by common name identified by detector at all locations deployed. Bats were identified by both detectors at CONW-0001 and NATW-0001 while only the AudioMoth identified bats at CONW-0002 and CONW-0003.

Common Name	Location and Detector					
	CONW-0001		CONW-0002	CONW-0003	NATW-0001	
	AudioMoth	SM4BAT-ZC	AudioMoth	AudioMoth	AudioMoth	SM4BAT-ZC
Big brown bat	x	x		x	x	x
Eastern red bat	x			x	x	
Evening bat		x				
Hoary bat	x	x	x	x	x	x
Silver haired bat	x	x	x	x	x	
Tri-colored bat	x			x	x	x

Table 3. Selection of sequential auto-identified calls at CONW-0002.

Location	Detector	Nightly Call Date	Time	Common Name
CONW-0002	AudioMoth	7/25/2018	4:32:29 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:32:59 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:33:14 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:33:29 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:33:44 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:34:14 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:34:29 AM	Hoary bat
CONW-0002	AudioMoth	7/25/2018	4:34:44 AM	Hoary bat

Detector Comparison

Confident calls were further filtered to analyze calls auto-identified on the same recording night by both detectors. Files that were made on the same recorder night, e.g. from 1800hrs to 0600 hrs., by both detectors within one hour of each other are shown in Table 4. Both detectors confirmed presence of Hoary bat at NATW-0001 on 7/27/2018 and CONW-0001 on 7/25/2018, 7/26/2018, and 8/15/2018. No other species was recorded by both detectors on the same night within a one-hour time window.

Table 4. Calls identified within one hour of each other by both detectors on the same nightly call date. Shaded rows highlight nightly calls to compare.

Location	Detector	Nightly Call Date	Time	Common Name
CONW-0001	AudioMoth	7/25/2018	9:02:00 PM	Hoary bat
CONW-0001	AudioMoth	7/25/2018	9:15:45 PM	Hoary bat
CONW-0001	AudioMoth	7/25/2018	9:16:00 PM	Hoary bat
CONW-0001	SM4BAT-ZC	7/25/2018	9:18:18 PM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	5:33:31 AM	Silver haired bat
CONW-0001	AudioMoth	7/26/2018	5:39:16 AM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	5:41:51 AM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	5:41:53 AM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	5:46:46 AM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	5:47:16 AM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	5:49:16 AM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	5:49:19 AM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	9:07:45 PM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	9:13:30 PM	Silver haired bat
CONW-0001	AudioMoth	7/26/2018	9:15:45 PM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	9:17:04 PM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	9:18:16 PM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	9:19:00 PM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	9:19:15 PM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	9:21:39 PM	Hoary bat
CONW-0001	SM4BAT-ZC	7/26/2018	9:22:04 PM	Hoary bat
CONW-0001	AudioMoth	7/26/2018	10:12:32 PM	Big brown bat
NATW-0001	AudioMoth	7/27/2018	9:01:45 PM	Hoary bat
NATW-0001	AudioMoth	7/27/2018	9:02:00 PM	Hoary bat
NATW-0001	AudioMoth	7/27/2018	9:06:30 PM	Hoary bat
NATW-0001	AudioMoth	7/27/2018	9:10:00 PM	Hoary bat
NATW-0001	AudioMoth	7/27/2018	9:10:45 PM	Hoary bat
NATW-0001	AudioMoth	7/27/2018	9:11:00 PM	Hoary bat
NATW-0001	AudioMoth	7/27/2018	9:11:45 PM	Hoary bat
NATW-0001	SM4BAT-ZC	7/27/2018	9:14:26 PM	Hoary bat
CONW-0001	AudioMoth	8/6/2018	10:24:30 PM	Silver haired bat
CONW-0001	SM4BAT-ZC	8/6/2018	10:30:18 PM	Big brown bat
CONW-0001	AudioMoth	8/6/2018	10:59:00 PM	Hoary bat
CONW-0001	AudioMoth	8/15/2018	3:35:17 AM	Hoary bat

Location	Detector	Nightly Call Date	Time	Common Name
CONW-0001	SM4BAT-ZC	8/15/2018	3:35:35 AM	Hoary bat
CONW-0001	SM4BAT-ZC	8/17/2018	8:21:12 PM	Evening bat
CONW-0001	AudioMoth	8/17/2018	8:22:15 PM	Eastern red bat

Figure 4 shows the spectrograms of calls auto-identified as Hoary bat and recorded on the same night at CONW-0001. Files were recorded within eight minutes of each other and show characteristics of a Hoary bat such as a reverse "J" shape around 20kHz. The AudioMoth spectrogram exhibits what appear to be echoes above 40kHz and noise below 10kHz. Figure 4 panels C and D are zoomed to show three pulses by each recorder with the full spectrum view turned off for the AudioMoth file. Both spectrograms show a similar call structure in zero cross mode with similar call shapes, maximum frequency, minimum frequency, and peak frequency.

Figure 5 shows files recorded within 90 seconds of each other at CONW-0001 on August 17, 2018. Panels A and C show what was identified as an Eastern red bat by an AudioMoth file. The spectrogram is not particularly clear; however, it is within +/- 5kHz of 40kHz frequency which is characteristic of the Eastern red bat. Panels B and D show what was identified as an Evening bat by the SM4BAT-ZC file.

Big brown bats were not recorded on the same recording night by both detectors during the deployment. Files made at CONW-0001 by the AudioMoth on recording night July 26, 2018 and a file by the SM4BAT-ZC on August 6, 2018 are compared (Figure 6 in Appendix B). Big brown bat spectrograms recorded by both detectors show similar structure and shape with a characteristic frequency around 25kHz. Similarly, Tri-colored bat files were analyzed at CONW-0001 from the AudioMoth and NATW-0001 for the SM4BAT-ZC from different nights (Figure 7 in Appendix B). Calls exhibit a hockey stick shape around 40kHz with calls produced by the SM4BAT more pronounced. Silver haired bat was only identified by the AudioMoth. A file on the recording night of August 26, 2018 shows a uniform oscillogram and a flat call pattern around 25 kHz typical of silver haired bat (Figure 8 in Appendix B). This file of the Silver haired bat is one of the clearest call files produced by the AudioMoth and visualized in Kaleidoscope Pro.

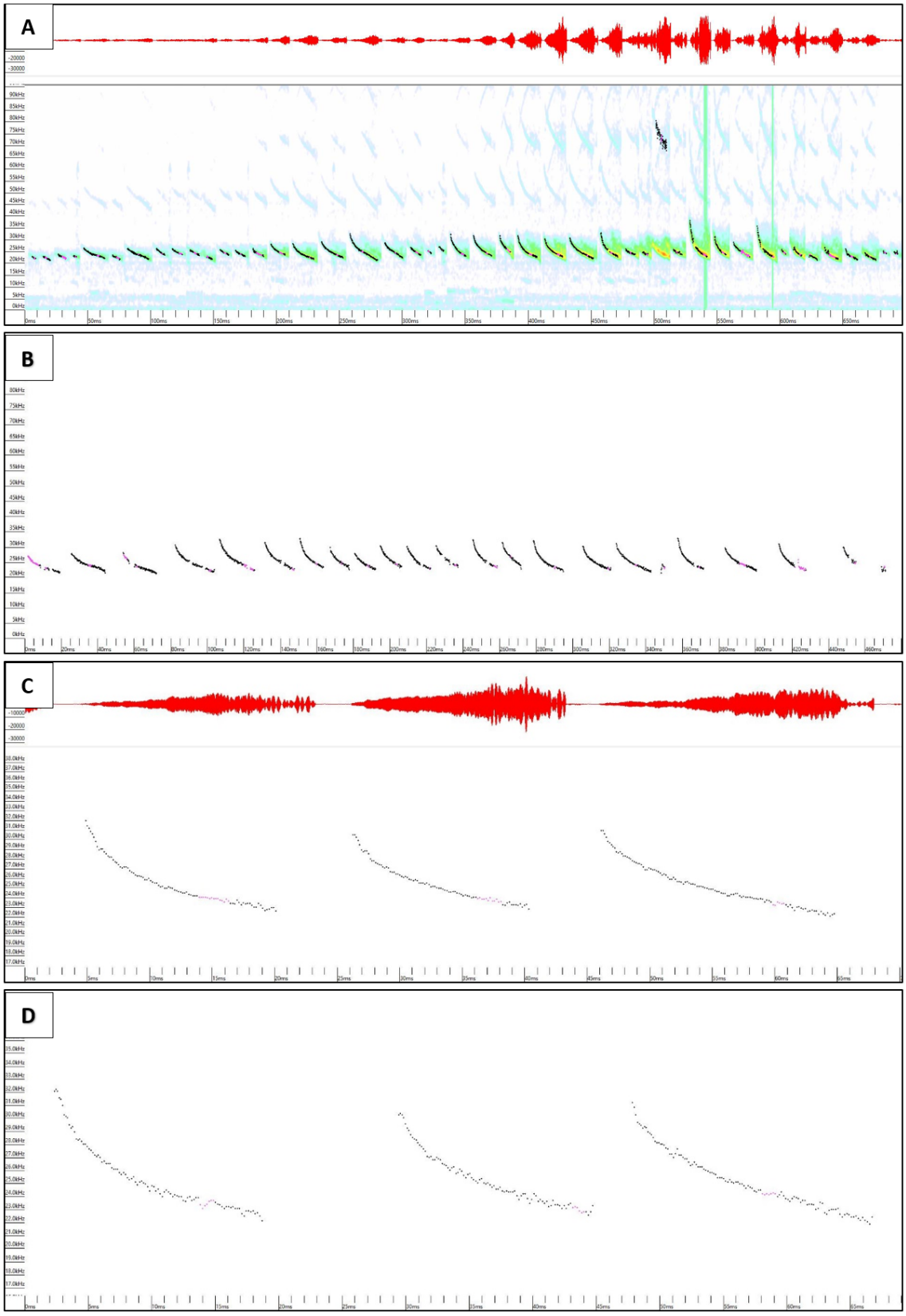

Figure 4. Spectrograms from nightly call date of July 6, 2018 and shown in compressed view with the frequency in kHz on the y-axis and the time in milliseconds (ms) on the x-axis. Panel A shows an image recorded by the AudioMoth and auto-identified as a Hoary bat at CONW-0001 at 05:39:16 AM. Panel B shows an image recorded by the SM4BAT-ZC and auto-identified as a Hoary bat at CONW-0001 at 05:41:53 AM (Note the apparent echoes in the AudioMoth file shown above 40kHz and oscillogram providing additional information to the full spectrum file (panels A & C). Panel C (AudioMoth with full spectrum view toggled off) and panel D (SM4BAT-ZC) are zoomed to show three consecutive pulses. Note the similar call shape, maximum frequency, minimum frequency, and peak frequency (indicated by pink dots) in all panels.

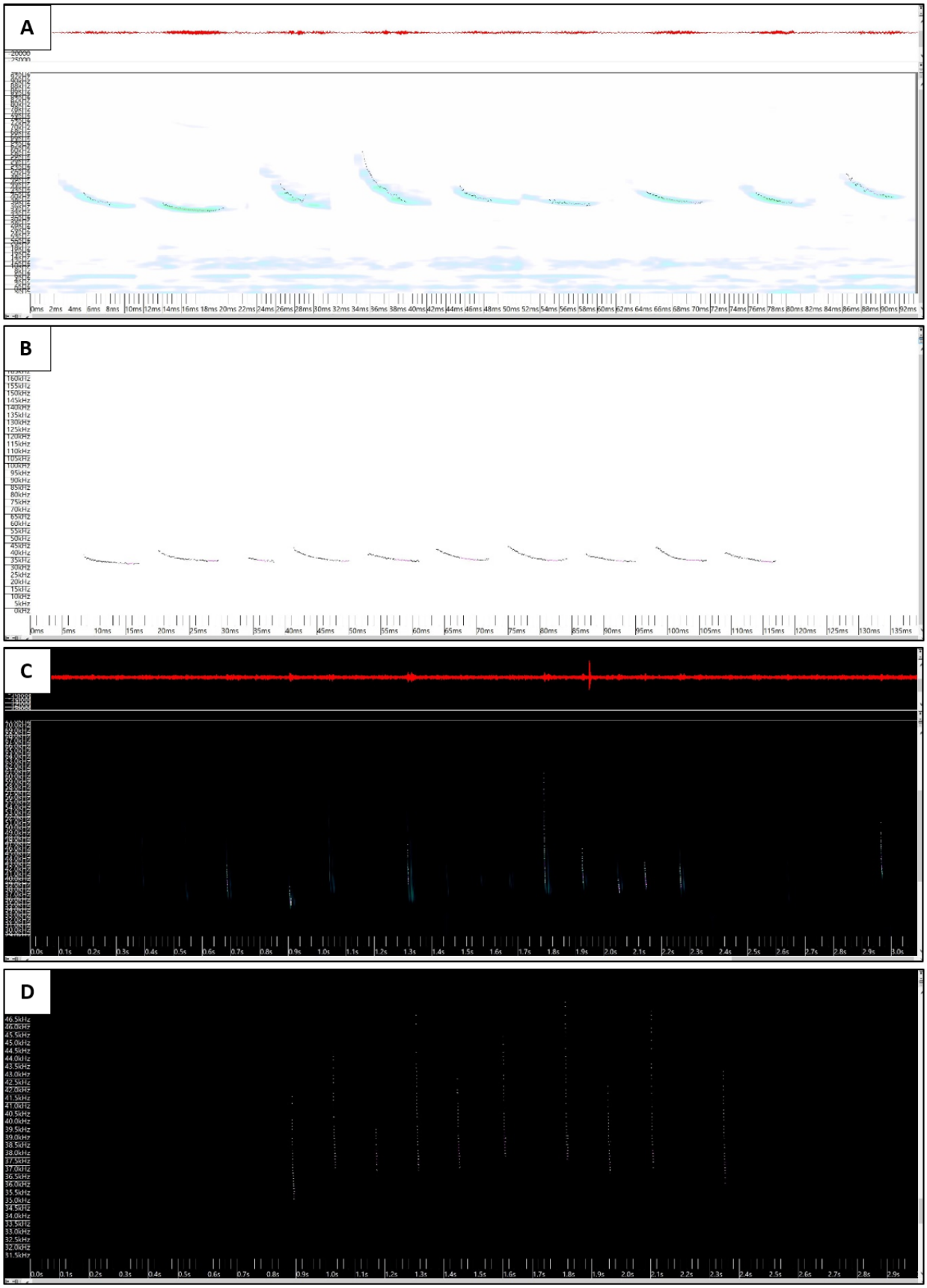

Figure 5. Files recorded within 90 seconds of each other at CONW-0001 on August 17. 2018. Panel A shows an image recorded by the AudioMoth and auto-identified as an Eastern red bat at 08:22:15 PM. Panel B shows an image recorded by the SM4BAT-ZC and auto-identified as Evening bat at 08:21:12 PM. Panels A and B show compressed view to eliminate spaces between pulses while C and D show real-time and in dark mode to highlight the pulses.

Discussion

This study shows that bats are present at ponds that treat AMD and a natural pond within the Flight 93 National Memorial Grounds. Additionally, the study shows that the AudioMoth 1.0.0 is capable of recording files which were then auto-identified as bats by an off-the-shelf classifier software. Based on auto-identified calls from both detectors it is inferred that six species were present during this study around all treatment ponds and that five were present at the natural pond. In general, future study should incorporate a greater number of treatment and non-treatment ponds/wetlands across a broad landscape to evaluate bat presence at AMD treatment ponds/wetlands and should measure additional variables such as: water chemistry, insect community, vegetation community/structure, and bat toxicology. Finally, mist netting in combination with acoustics could be performed at the park to confirm which species are using the landscape and how they may be using the water resources, e.g. foraging or drinking.

Several lessons were learned on deployment and post processing of the AudioMoth 1.0.0 for passive acoustic monitoring of bats:

- **microSD card formatting.** Data absence from multiple occasions on the AudioMoth appears to be attributed to refreshing with microSD cards that had not been reformatted prior to deployment. MicroSD cards were formatted to MS-DOS (FAT 32) prior to initial deployment of each AudioMoth. The cards were not always reformatted following data extraction and redeployment of the cards. This may have contributed to dates with no data logged. It is imperative that the microSD card be re-formatted prior to each deployment/refresh.

- **File parsing and auto-triggering.** Because the AudioMoth records one continuous file the recording must be split to a set duration and fifteen seconds was chosen as the duration on recorded AudioMoth files based on discussion with Wildlife Acoustics technical support staff (personal communication, August 2018). Further research indicated that 15 seconds is more often a duration for frequency division files while a maximum duration for full spectrum may be better limited to eight seconds. Some files ended up with what appeared to be multiple sequences of calls by either the same species or a different species. This could affect the output of the classification software and species could be missed. To avoid this problem the AudioMoth could be programmed to automatically trigger and only record within the frequency range of expected bat species.

- **Weatherproofing.** Recorded calls with the AudioMoth were noisy and spectrograms exhibited what appear to be echoes which may be attributed to placing the AudioMoth in Ziploc® bags for weather protection, may be from waves reflected off the trees, or may be due to poor microphone quality. Additionally, the detector deployed at CONW-0002 was damaged by moisture and no data were recorded following realization of the damage. Future study should take into consideration weather proofing alternatives such as a molded plastic enclosure and the potential for use of an acoustic cloth to minimize decibel loss and maximize protection from precipitation.

- **Auto-trigger for recording.** Setting AudioMoth to continuously record requires an enormous amount of data storage. Resultant continuously recorded WAV files took hours to process and required hundreds of gigabytes of storage space. This made data analysis cumbersome and archival retention of files problematic. Data processing and storage issues could be resolved by programming the device to auto-trigger so that it only recorded files that would be of an expected bat frequency.

- **Direct Full-Spectrum recorder comparison.** Future field study of AudioMoth efficacy should focus on a direct comparison of WAV files recorded from full-spectrum recorders such as the Wildlife Acoustics SM4BAT-FS or Pettersson D500x. While the results of this study show that the lower-cost AudioMoth 1.0.0 could record ultrasonic frequency calls, and identification is possible using an existing auto-identification software, a direct analysis may not be drawn due to the difference in recording formats of the two recorders.

Based on AudioMoth data alone, bat presence within the Grounds did not vary between the natural and AMD treatment ponds as the same species were identified across all ponds (aside from CONW-0002 where device damage may have influenced the results). SM4BAT-ZC files did not identify any species at CONW-0002 and CONW-0003 which may be attributed to a difference in detector performance, poor water quality at CONW-0002, and/or clutter at CONW-0003 around the SM4BAT-ZC and not the AudioMoth. SM4BAT-ZC deployment at CONW-0003 was more obstructed by tree branches and clutter due which may have impacted recording quality. CONW-0003 did not have as much open water as the other wetlands, was farther along in the sequence of in the AMD treatment system, and exhibited more emergent wetland vegetation than any other wetland. The presumed improved water quality would suggest that it could potentially support a more diverse and abundant range of forage organisms, however, the additional vegetation may not allow for CONW-0003 to be used as a drinking location. CONW-0002 was the first in the series of treatment ponds and had more visible iron flocculant than all other treatment ponds. This could have affected bat presence due to the potential for reduced organisms for forage or bats preferentially not choosing to drink from this pond. The most species were identified by both detectors at CONW-0001 and NATW-0001. This may be attributable to the open water aspect of the two sites and/or the water quality. CONW-0001 is the last in a series of four treatment ponds and did not exhibit the same amount of iron flocculant as CONW-0002. Therefore, there may have been additional forage, or the bats preferred to use this as a drinking location. It is not surprising that bats were present within the Grounds due to the abundance of open water. However, it is interesting that the species present at one of the treatment wetlands nearly mirrored the natural wetland. Mist netting is recommended at CONW-0001 and NATW-0001 to confirm detector results.

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Appendix A

AudioMoth File Processing

In Excel:

1. Create and populate LOCATION column.
2. Create and populate DETECTOR column.
3. Create and populate LATITUDE column.
4. Create and populate LONGITUDE column.
5. Remove file extension from the input file to create **FILE** column with formula: =LEFT(H2,LOOKUP(2^15,FIND(".",H2,ROW(INDIRECT("1:"&LEN(H2)))))-1)
6. Convert hexadecimal file to decimal and create **TIMESTAMP** column with formula: =HEX2DEC(N2)
7. Convert timestamp to date/time with OFFSET and create **GMT_DATE** column with format with Date type: *3/14/2012 and formula: =(((O2/60)/60)/24)+DATE(1970,1,1)+(((J2/60)/60)/24)
8. Convert date/time from Greenwich Mean Time (GMT) to Eastern Standard Time (ET) and populate **DATE** column with format with Date type: *3/14/2012 and formula: =P2- (4/24)
9. Format cells to create three additional columns splitting ET result field (DATE) into:
    a. **TIME**, with Custom Time type: h:mm:ss
    b. Create a Date_Time_Concatenated column and store date and time in one field with formula: =TEXT(R2,"mm/dd/yyyy")&" "&TEXT(S2,"hh:mm:ss"). ***This can also be done by using formula: =P2- (4/24) and custom Time type: mm/dd/yyyy, h:mm:ss

SM4BAT-ZC File Processing

In Excel:

1. Create all additional columns to match the AudioMoth file table structure.
2. Create a Date_Time_Concatenated column and store date and time in one field with formula: =TEXT(R2,"mm/dd/yyyy")&" "&TEXT(S2,"hh:mm:ss").

Combined File Processing for Confident Calls and Recorder Night

1. Combine all processed files into a single Excel worksheet with all formulas removed.
2. Filter results:
    a. AUTO ID Column: Remove “NoID” and “Noise” records
    b. PULSES Column: Filter to remove files with less than nine (9) pulses
    c. MATCH RATIO Column: Filter to remove files that have a match ratio greater than (>) 0.66
3. Determine recording night, i.e. continuous duration recorder was logging data:
    a. Add additional columns: AM or PM and Nightly Call Date
    b. Parse AM or PM from Time column and populate AM or PM column
    c. Populate Nightly Call Date with the date shown in Recorded Date column if PM or the Recorded Date (-)1 day if AM

Appendix B

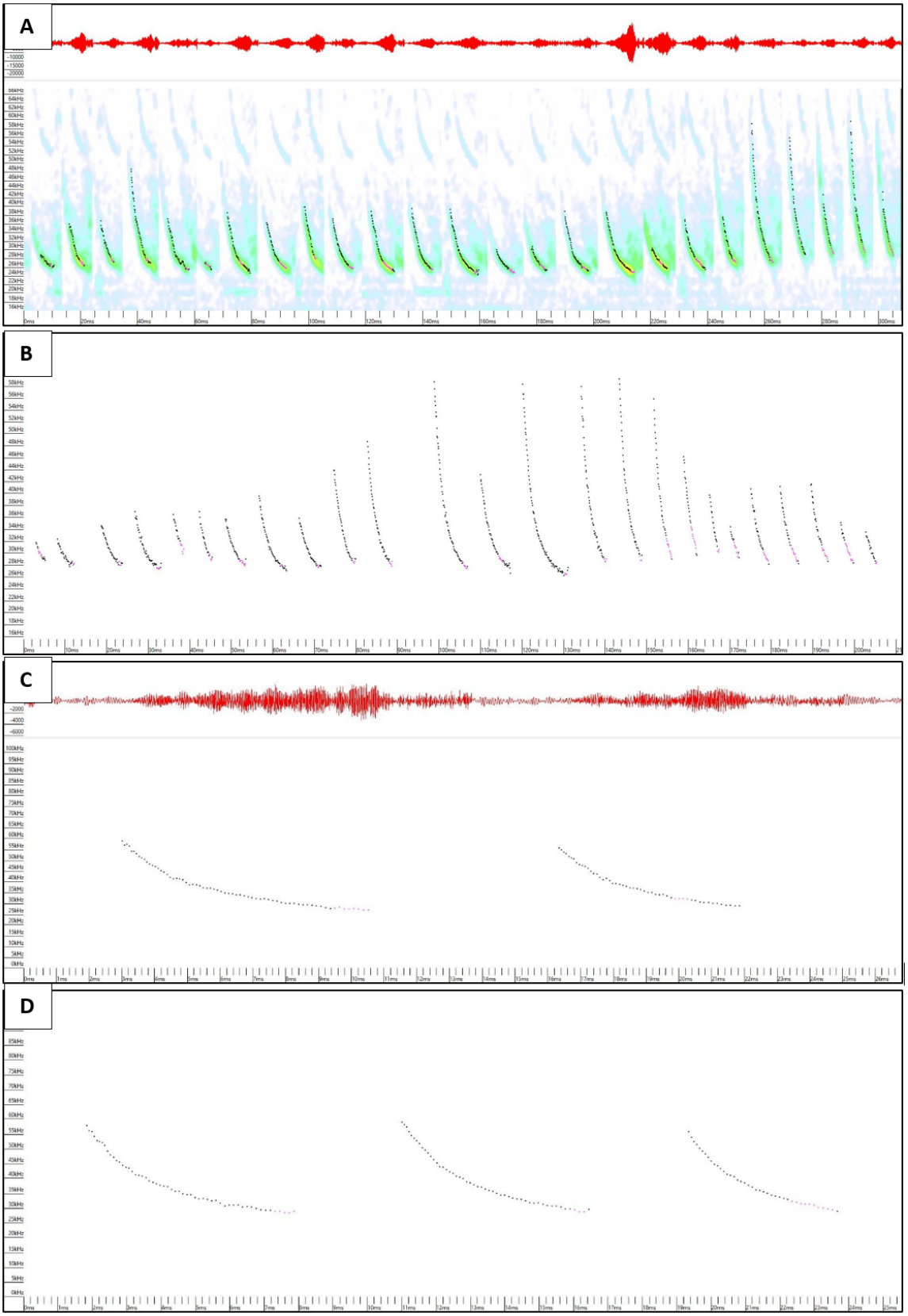

Figure 6. Panel A shows an image recorded by the AudioMoth and auto-identified as a Big brown bat at CONW-0001 at 10:12:32 PM on July 26, 2018. Panel B shows an image recorded by the SM4BAT-ZC and auto-identified as a Big brown bat at CONW-0001 at 10:30:18 PM on August 6, 2018. Panel C (AudioMoth) and panel D (SM4BAT-ZC) are zoomed in to highlight two pulses on the AudioMoth call and three on the call SM4BAT-ZC call.

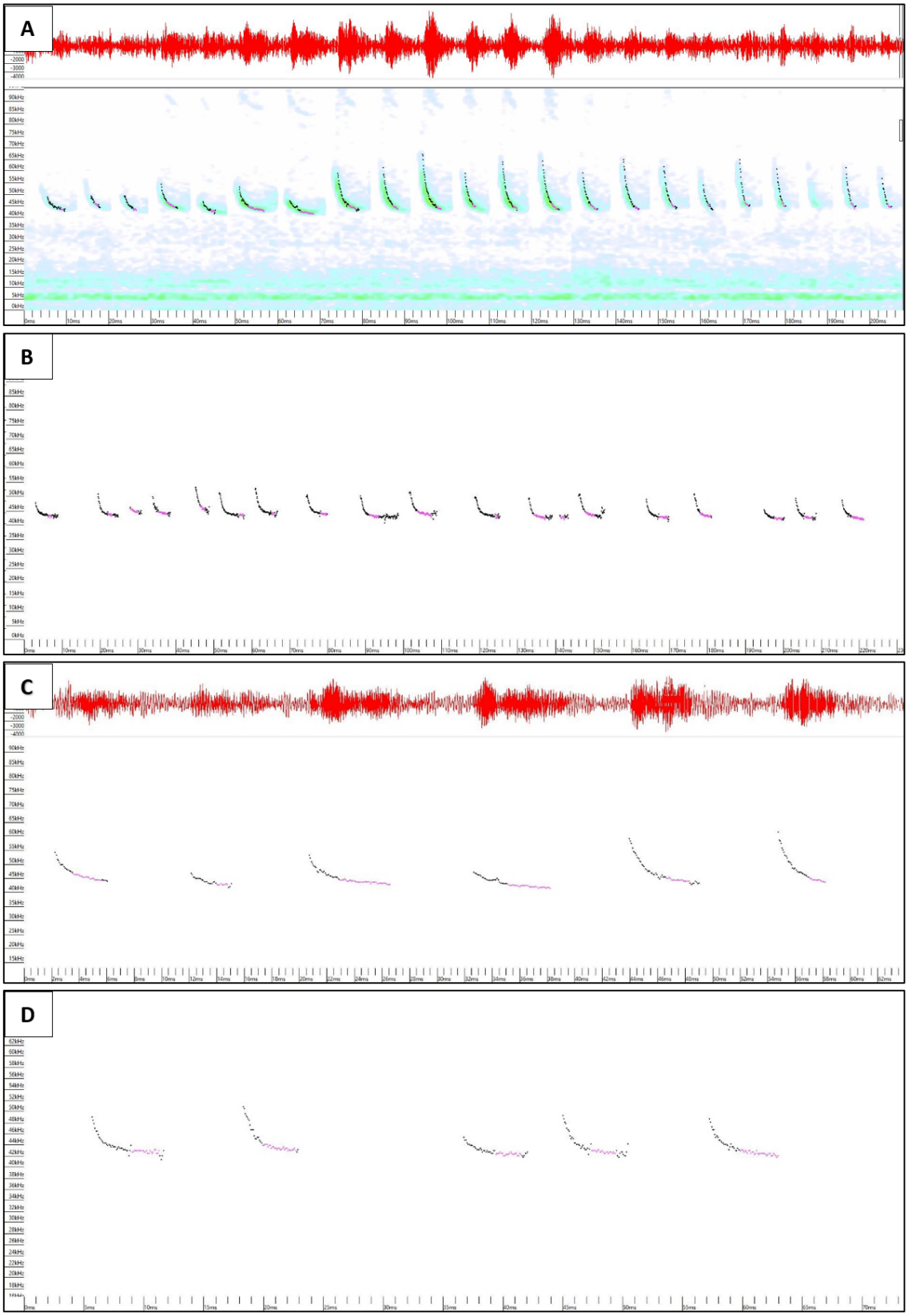

Figure 7. Panel A is an image recorded by the AudioMoth and auto-identified as a Tri colored bat at CONW-0001 at 02:22:32 AM on August 18, 2018. Panel B is an image recorded by the SM4BAT-ZC and auto-identified as a Tri colored bat at NATW-0001 at 05:38:15 AM on August 24, 2018. Note the hockey stick shaped calls in panel B with only several similar shaped pulses in panel A. Note when zoomed in the individual pulses the AudioMoth (panel C) spectrogram is not as clear as the SM4BAT-ZC (panel D).

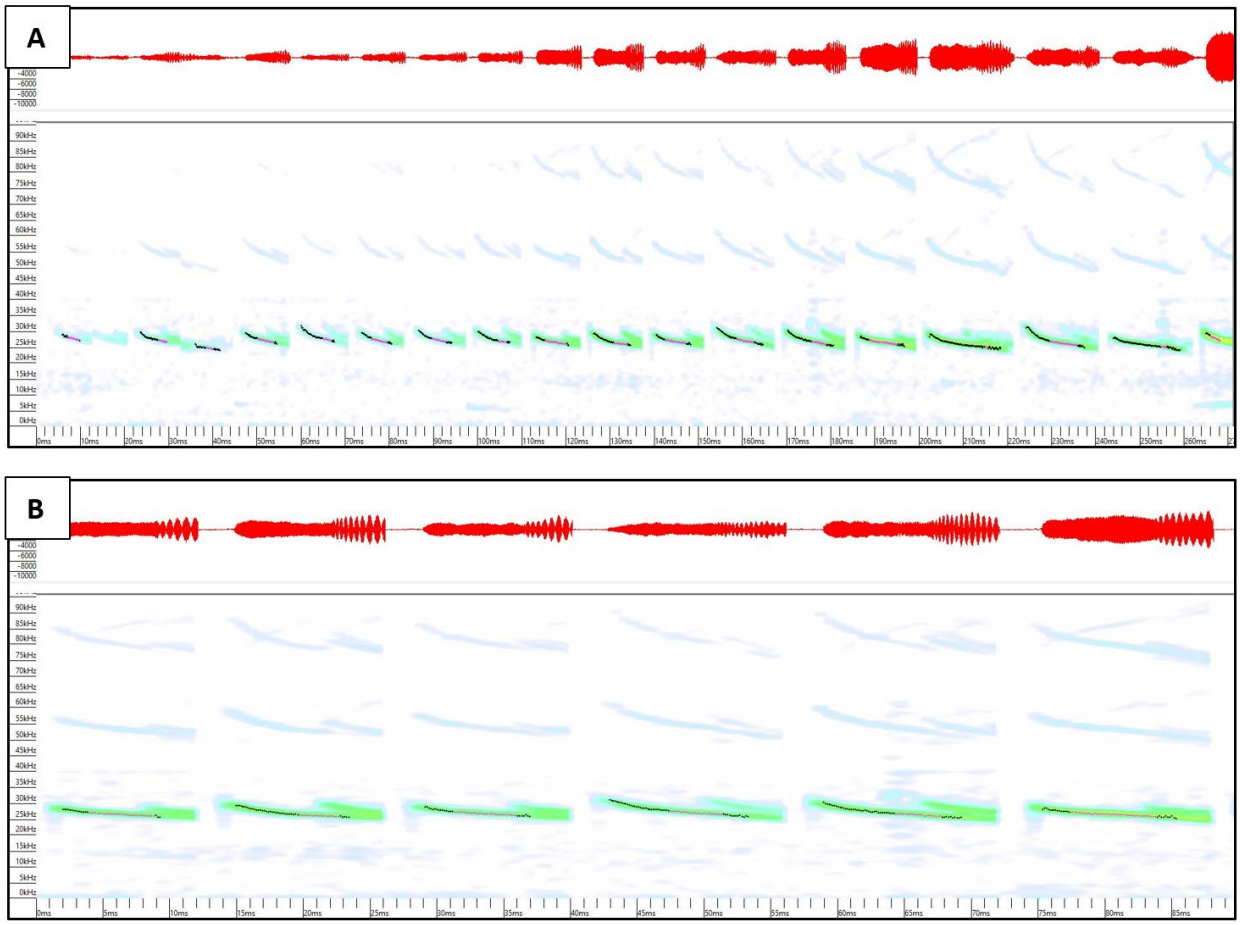

Figure 8. Panels A and B show an image recorded by the AudioMoth and auto-identified as a Silver haired bat at NATW-0001 at 05:24:15 AM on August 27, 2018. Panel B is zoomed in to see six pulses. Note what appear to be echoes above 50kHz. This file was one of the clearest files recorded by the AudioMoth during the deployment. No file was auto-identified as a Silver haired bat by the SM4BAT-ZC throughout the deployment.

Appendix C

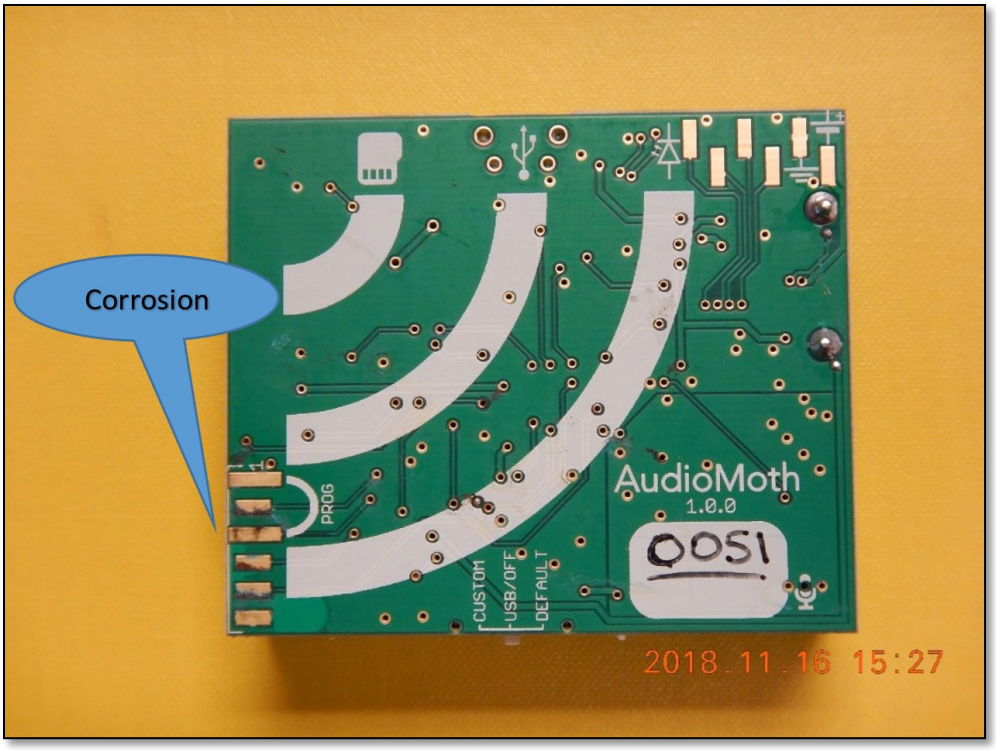

Figure 9. AudioMoth deployed at CONW-0002 for duration of deployment. Note corrosion on lower left corner of the recorder. Credit: Brian Dailey, OSM.

Figure 10. Open water pond CONW-0002. Note the iron flocculation/precipitation in the treatment pond and coating the plastic pipe along the water's edge. Credit: Brian Dailey, OSM.

Figure 11. AudioMoth 1.0.0 deployed two (2) meters off the ground at NATW-0001. Microphone is oriented toward the camera. Credit: Brian Dailey, OSM.

Figure 12. AudioMoth 1.0.0 deployed at NATW-0001. View is of the open water pond from behind the detector. Microphone is oriented toward the open water. Credit: Brian Dailey, OSM.