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Evaluating Methods for Emergence Counts at Bat Roosts: A Pilot Study Comparing Drone-acquired Thermal Imagery, Acoustic Estimates, and Visual Observations

Karah Jaffe[1, 2, *], Richard Carter[2] and Aaron J. Corcoran[3]

Abstract - Evaluating methods for monitoring wildlife populations is essential for conservation. Here, we conduct an initial assessment of the precision of 3 methods deployed concurrently for surveying bats emerging from roosts: human visual observation, drone-acquired thermal imagery, and sound-pressure levels of ultrasonic recordings. We conducted 3 surveys at a roost of *Myotis grisescens* (Gray Bats) and 10 surveys at a roost of *Eptesicus fuscus* (Big Brown Bats). Fitting linear and exponential models to the data resulted in strong correlations between visual counts and both drone-acquired (r2 = 0.83–0.91) and acoustic (0.51–0.90) estimates. Nightly counts based on thermal imaging differed from visual counts by a median of 17.5% and 32% at our 2 study sites (range: 1.6–263%), while nightly acoustic estimates differed from visual counts by a median of 4.7% and 15.1% (3.5–144%). We discuss the limitations and advantages of each method and conclude that all can be useful for surveying bats, but each has its own inherent constraints. When possible, we recommend using multiple methods concurrently to increase the reliability of count estimates.

Introduction

Monitoring population sizes over time is integral for effective wildlife management. However, measuring wildlife populations with sufficient accuracy is often challenging (Vallecillo et al. 2021). Various methods have been developed to establish baseline population estimates and evaluate changes over time (Davis and Winstead 1980). With a range of methods available for monitoring populations, exploring the feasibility of integrating new technologies with traditional approaches is essential for enhancing and refining current methodologies.

Traditional approaches to counting colonies of bats include visual counts of bats in roosts, visual emergence counts, recorded imagery, and mark-recapture methods, all of which are then used to infer broader population-level trends (Kunz et al. 2009). Visual emergence counts have been used widely to estimate bat populations at tree cavities, caves, cave-like features, and rock crevices (Betke et al 2008, Speakman et al. 1992). This approach remains a primary method for surveying bats, such as with the North American Bat Monitoring Program (Loeb et al. 2015). In visual surveys, observers are stationed underneath an estimated flight pathway to view bats silhouetted against the sky. However, visual counts have some widely recognized limitations, such as being unreliable during high-density emergences (Betke et al. 2008, Hristov et al. 2008, McCracken 2003). Additionally, visual counts are considered labor-intensive, often requiring multiple observers at a single location.

The application of thermal imagery has become increasingly prevalent in wildlife detection and population estimation (Cilulko et al. 2013, Lethbridge et al. 2019, Sabol

[1]U.S. Fish and Wildlife Service, Kentucky Field Office, Frankfort, KY 40601. [2]Department of Biological Sciences, East Tennessee State University, Johnson City, TN 37614. [3]Department of Biology, University of Colorado, Colorado Springs, CO 80918.
[*]Corresponding author - jaffe.karah@gmail.com

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and Hudson 1995). In the past 3 decades, thermal imaging has been employed to record and count bats during emergence (Betke et al. 2008; Elliott et al. 2005, 2011; Frank et al. 2003; Sabol and Hudson 1995). Thermal imagery has typically been obtained with cameras positioned at fixed locations on the ground. Drones, also referred to as "unmanned aircraft systems," offer the advantage of capturing imagery from a top-down perspective and allowing real-time adjustments to capture bats that emerge unexpectedly from different locations. This approach ensures a more comprehensive recording of the entire emergence, accounting for unpredictability in individual flight paths, which ground observers (and ground-based cameras) may miss. The merits of employing thermal imaging with drones for wildlife surveys are considerable, providing biologists with a valuable tool for data collection (Christie et al. 2016, Hodgson et al. 2018, Israel 2011, McCarthy et al. 2023, Mulero-Pázmány et al. 2014, Sardà-Palomera et al. 2012, Seymour et al. 2017, Whitworth et al. 2022). Recent studies have also highlighted the utility of drones integrated with acoustic detectors for recording bat echolocation, further emphasizing the versatility of drones in wildlife research (Jespersen et al. 2022, Kloepper and Kinniry 2018, Michez et al. 2021).

Despite technological advancements, the use of thermal imaging with drones presents challenges. Drone operation requires trained personnel, and legal and safety considerations must be addressed, including complying with regulations and ensuring the safe operation of drones in various environments. Additionally, the analysis of thermal imagery can be time-consuming (Anderson and Gaston 2013, Linchant et al. 2015), although automatic counting software may streamline manual analysis (Bentley et al. 2023; Corcoran et al. 2021a, 2021b; Elliott et al. 2011).

Acoustic surveys are another option for surveying bats. Traditionally, acoustics have been used to assess bat activity rather than for counting bats exiting a roost (Kunz et al. 2009, Milchram et al. 2020). Acoustic methods are non-invasive, cost-efficient, and capable of covering large areas for extended periods with low maintenance (Milchram et al. 2020). More recent studies have developed methodologies for conducting bat counts using acoustic detectors. For example, Kloepper et al. (2016) counted *Tadarida brasiliensis* Geoffroy (Mexican Free-tailed Bats) leaving a cave by simultaneously recording emergences with thermal imaging and ultrasonic microphones placed outside the roost. The thermal-imaging videos were used to determine how many bats exited at each moment in time, and these data were used to establish a relationship between acoustic sound levels and number of bats that exited the cave. With this approach, the individual echolocation calls are not counted, but rather the sound level is measured in various ways, such as taking a root mean square (RMS) of the signal. This acoustic censusing method showed promising results with a colony of about 700,000–900,000 Mexican Free-tailed Bats (Kloepper et al. 2016) and 22,600–38,700 *Myotis grisescens* Howell (Gray Bats) (Eddington et al. 2023). However, this method has not been tested at smaller colonies with only hundreds or a few thousand bats.

We conducted an initial field test of 3 methods for counting bats during emergence events: visual counts by human observers, counts from thermal imagery acquired with drones, and acoustic-based estimates. Our first objective was to determine the similarity of the counts produced with these 3 approaches, both on short time scales (within 1-min intervals) and across an entire emergence. However, because no method can produce a perfect count, the differences in counts produced with different methods (i.e., error rates) result from the sum of errors from each method being compared. By using these methods concurrently, we aimed to provide a comparative analysis of their strengths and limitations, as well as provide practical knowledge for researchers and managers who may be interested in adopting these approaches. We emphasize that the aim of this study was not to conduct an exhaustive comparison of these methods under different conditions, but rather to conduct an initial test and comparison. The inclusion of drone-acquired thermal imagery and acoustic-based estimates was intended to broaden the toolkit available for bat monitoring and provide an exploratory review of method performance.

Field-site description

Bat emergence counts were conducted at 2 roosts using 3 concurrent methods (Fig. 1). The first site, located in Hawkins County, Tennessee, was surveyed for 10 nights from 15 to 25 June 2021. Across these 10 nights, we flew a drone at altitudes ranging from 45 to 65 m to determine the altitude at which the relative size of bats in the images became too small for accurate detection. After review, we discarded 7 nights during which the altitude was too high for accurate drone-based counts, retaining 3 nights for further analysis. The site consisted of an incomplete concrete building surrounded by open fields, industrial development, and forested parcels along the Holston River. A colony of Gray Bats inhabited the structure year-round. During emergence, bats exited the roost through a gap in the first-floor ceiling before traveling along wooded, edge habitat and crossing into an open field.

The second roost, located in Yancey County, North Carolina, was surveyed for 10 nights between 8 and 22 August 2021. This site features a concrete bridge that spans the South Toe River; the bridge is about 30-m long and located 10 m above the stream. A colony of *Eptesicus fuscus* Palisot de Beauvois (Big Brown Bats) inhabited the bridge. The bats emerged from the bridge's bearing spaces and traveled either east or west via the river corridor (Fig. 1). Bats leaving in opposite directions would not be visible or detected at the same time; therefore, eastward and westward counts were compared separately.

Methods

Visual observations

At each site, 2 people observed bats silhouetted against the sky during emergence. The observers had mixed experience with visual counts: 1 observer in North Carolina had prior experience, while the other 3 persons had training, but no direct experience counting bat emergences. Visual surveys commenced 30 min before sunset and continued throughout

Figure 1. Emergence pathway and data collection points at the (a) Tennessee and (b) North Carolina sites. The arrows represent the emergence pathways. Drone icons represents where the drone-acquired thermal imagery was recorded; microphone icons represent the location of the acoustic recorders; and person icons represent the location of the visual observers.

the emergence, concluding when bats were no longer visible due to decreased light. All observers used the smartphone application “Counter” (Tsukanov 2021). Bats were counted individually as they either exited (+1) or re-entered (-1) the roost. The application provided a precise time history of each bat that was counted, which could later be compared to concurrent counts from acoustics and thermal video.

In North Carolina, observers were stationed 35 m east and west of the bridge. Their counts were summed to generate the total count for each night to compare with counts from thermal imagery. Conversely, in Tennessee, 2 observers were stationed at a single emergence viewpoint; therefore, we had a replication of observer counts at this site.

Drone-acquired thermal imagery

A drone (Phantom 2 non-Vision, DJI, Shenzhen, China) was operated directly above the bat roost exit. In accordance with federal and state regulations, the pilot obtained a Part 107 Remote Pilot Certificate and scientific permits prior to deploying the drone. Regulatory compliance consisted of strict adherence to airspace restrictions, safety protocols, and use of anti-collision lighting visible from at least 3 statute miles. The drone used in the current study was acquired based on available resources, although there were known issues with its flight stability and limited duration of battery charges (see Discussion). Even with such limitations, we believe this model was suitable to test the ability of drones to produce accurate exit counts.

The drone was equipped with a thermal camera (Vue Pro R 640, Teledyne FLIR, Wilsonville, OR) positioned perpendicular to the ground. The camera had a thermal resolution of 640 by 512 pixels, a 13-mm lens, and a field of view of 45 by 37°. Thermal imagery was recorded at 30 frames/sec. The drone operator maintained a similar field of view between nights using visual landmarks. The clock of the drone was synchronized to the clock of the audio and visual observations, allowing us to compare counts during the intervals when the drone was collecting data. Recordings concluded either at the end of the emergence or when the batteries were depleted.

The drone was flown at an altitude of 45 m above ground level, providing a ground coverage of about 38 by 29 m. Operational procedures were implemented to maximize the drone’s limited battery life, with flight initiated at the first signs of emergence. The pilot maintained a visual line of sight on the drone throughout operation to prevent any potential interactions between the bats and the unit. However, the drone could fly and record thermal video for only 15 min before it had to land for replacement of batteries. Due to variable weather conditions, 1–3 flight periods were conducted per night.

For effective detection of target species in thermal imagery, sufficient thermal contrast is required to distinguish bats from the background. Gray Bats in Tennessee contrasted against the cooler concrete building, whereas Big Brown Bats in North Carolina contrasted against the cold river below the bridge (Fig. 2). Aspects of the thermal camera (i.e., lens focal length and sensor size), coupled with a flight altitude of 45 m, resulted in bats that appeared as 3–4 pixels in diameter within images. Thermal imagery was post-processed using video-editing software (Premiere Pro, version 15.0, Adobe, San Jose, CA), to stabilize drone-related motion and enhance contrast for target detection. Contrast values were adjusted depending on visual needs, typically being increased by 25–30°. The drone lacked GPS capabilities, leading to flight instability (drift) that resulted in unsteady imagery not suitable for automatic counting software (Corcoran et al. 2021a). Therefore, bats were manually counted in ImageJ (Schneider et al. 2012).

Acoustic-based estimates

Acoustic detectors equipped with omnidirectional microphones (models SM4BAT-FS and SMM-U2, Wildlife Acoustics, Maynard, MA) were positioned outside each roost's exit. The detectors were configured to record continuously throughout the emergence, with a sample rate of 256 kHz and minimum trigger frequency of 20 kHz; the schedule was set to "record always," and maximum length of recording was 60 min. Microphones were oriented to detect calls from the emergence pathway, minimizing interference from overhanging structures or vegetation. We placed detectors at the same locations with the same settings for 10 nights in both Tennessee and North Carolina.

In Tennessee, a single acoustic detector was deployed about 5 m from the center of the exit. To avoid recording bats swarming in the building, the microphone was aimed at a 45° angle away from the roost, in the direction that the bats traveled as they emerged. In North Carolina, 1 detector was placed about 15 m downstream (east), and another detector was placed about 15 m upstream (west) of the bridge to capture bats leaving in both directions. We determined that the exit rate from the detector on the west side of the bridge was likely too low to be used for acoustic analysis (<50 individuals per night), so we restricted our analysis to the detector and manual counts from the east side of the bridge, where exit rates were greater.

Acoustic files were processed using custom code written in MATLAB (v2022; Mathworks, Natick, MA; see Appendix 1 for sample code). At the quieter Tennessee site, drone noise was detected, whereas at the North Carolina site, insect, road, and other low-frequency ambient noises were more prominent. A 30-kHz high-pass filter was applied to

Figure 2. Example of drone-acquired thermal imagery during emergences. (a) Example image from North Carolina. (b) 3000% zoomed image of single bat from the red rectangle shown in (a). (c) Example image from Tennessee. (d) 3000% zoomed image of 3 bats from the red rectangle shown in (c).

remove low-frequency, non-targeted acoustic signals. We then split files into 1-min sections and calculated root mean square (RMS) sound pressure levels (SPL) and median SPL from each 1-min interval (Fig. 3). Because a calibration tone of known intensity was not used and is not necessary for this kind of analysis, we measured and reported results in decibels SPL relative to the maximum value that the acoustic detector can record, following the method described by Eddington et al. (2023). Specifically, RMS SPL in decibels (dB) was calculated as: SPL_RMS=20 log_10 ((RMS of signal)/(RMS of maximum signal)),

where signal was the recorded sequence of amplitude values that ranged from -1 to 1, and maximum signal was a repeating sequence of -1 and 1 values. With this approach, the maximum value that could be recorded on the detector was 0 dB, and all other values were negative relative to this maximum SPL. Median SPL in dB was calculated the same way, except the median signal level was used in the equation instead of RMS. We chose to calculate median SPL because we expected it not to be biased by short time periods with high sound levels, such as when individual bats flew close to the microphone.

The counts from visual observations were used to establish a correlation between visual emergence counts and either RMS or median SPL measured within 1-min intervals. For this analysis we modeled the relationship as an exponential equation, since a doubling of sound level results in an increase of 6 dB in SPL: N_bats=a SPL^x.

Here, Nbats is the number of bats exiting within a 1-min interval, SPL is either the RMS or median sound pressure within a 1-min interval, and a and x are solved for in the analysis.

Comparison of methods and statistical analysis

We calculated emergence counts from the data obtained from the observers, drone-acquired thermal imagery, and acoustic estimates. We split all counts into 1-min intervals and synchronized across methods for each site and night of surveys, based on timestamps established during data collection. We first compared human-to-human variability at the Tennessee site, where we had replication of observations at the same roost exit. We then

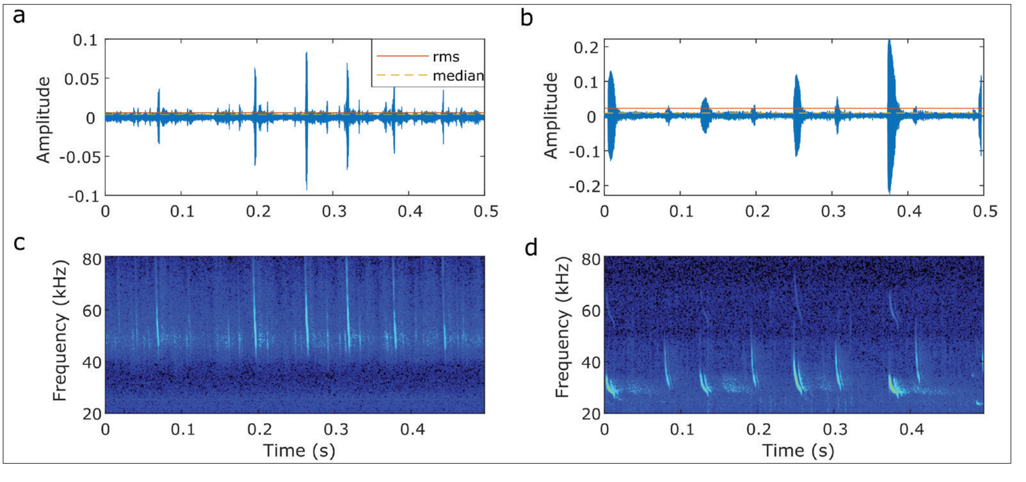

Figure 3. Example acoustic recordings used in analysis, including oscillograms and spectrograms from (a, c) Tennessee and (b, d) North Carolina. Measured levels for RMS and median amplitudes are shown. Note the nearly continuous background sound at approximately 50 kHz in (c) resulting from the large number of bats present at the Tennessee site, but the reduction of background signals at the lower-density North Carolina site (d).

compared video-based and acoustic-based counts to these human counts, although 2 nights of data from North Carolina (15 and 21 August 2021) were not used in the analysis because the sound files were corrupted by unknown causes. To determine the similarity of estimates, we compared total counts for each method and for each survey night, considering only the periods when data were available for the pairwise methods being compared. For these comparisons, 1 count was selected as a baseline (typically the human visual counts), and we calculated the absolute value of the difference between this count and the count measured with the other method. The median of this absolute difference was then taken as a summary of the overall similarity of the 2 counts. Because drones only operated for 15 in at a time, these counts were compared to human counts only within the same intervals.

In addition, we generated regression models, using visual observation count as the predictor variable and acoustic estimate or thermal count as the response variable, with counts binned into 1-min intervals. We used the coefficient of determination (r2), as an indicator of the similarity of the counts generated using the different methods. We note that there was likely temporal autocorrelation both on a minute-to-minute and night-to-night basis within our data, and therefore, statistically, there was some degree of pseudo-replication.

We used a cross-validation procedure to estimate nightly emergence counts from acoustic SPL levels from data collected on nights not including the night for which the prediction was made. For North Carolina, we used 8 nights of data. We split these 8 nights into 2 sets—the first 4 nights and the second 4 nights. From each set, we created an exponential regression model to predict nightly emergence counts for the other set of nights. For Tennessee, we had 3 nights of data; therefore, we created a model to predict each night's estimate using data from the other 2 nights.

Results

Comparison of counts by multiple human observers

Across 3 nights in Tennessee, the 2 observers had highly correlated overall counts, as displayed in the time-courses of the counts for each night (Fig. 4a) and the linear regression of counts conducted broken down for all 3 nights in 1-min intervals (Fig. 4b; r2 = 0.85, F1, 142 = 690, $P < 0.0001$). However, there was a 5-min period during the third night (gray-filled symbols in Fig. 4a–b) when the 2 observers differed considerably, with observer 2 producing counts approximately 200% that of observer 1. This event occurred near the peak emergence of the bats. An assessment of counts across methods (discussed below) suggested that observer 1 had a more accurate count. Therefore, the counts from observer 2 during these 5 min were excluded as outliers from further comparisons. Observer 1 also had slightly lower counts than observer 2 overall, as indicated by a slope of the regression line equal to 0.85 (95% CI: 0.82–0.88), even with the outliers removed.

Comparison of visual observation and thermal counts

Overall, visual counts were closely correlated with thermal-imagery counts for each night at both locations (Fig. 5a–c). The linear regression of the 1-min interval counts for the 2 methods revealed a high correlation in both Tennessee (r2 = 0.91, F1, 150 = 906, $P < 0.0001$) and North Carolina (r2 = 0.83, F1, 578 = 1164, $P < 0.0001$). The slope of the regression equation did not differ from 1 (95% CI 0.99–1.13) for Tennessee; however, the slope equaled 0.90 (95% CI: 0.85–0.95) for North Carolina, indicating that visual counts were about 10% lower than thermal counts on average.

When we compared counts aggregated across study nights during the times when drones were operational, thermal-imaging counts differed from visual counts by a median of 32% in Tennessee and 17.5% in North Carolina. However, some nights differed by as much as 263% between methods in North Carolina (Table 1).

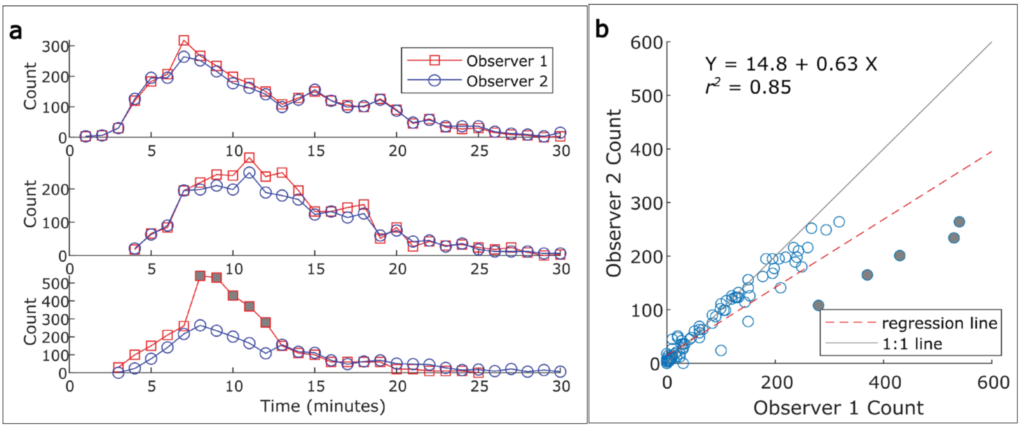

Figure 4. Temporal activity of bats during emergence censused by 2 observers in Tennessee. (a) Counts of bats over time for the 3 nights censused. (b) Comparison of counts recorded by the 2 observers over 1-min intervals. Gray-shaded symbols indicate 5 intervals during which the 2 individuals differed substantially in their counts. Time in (a) indicates minutes after the beginning of the surveys.

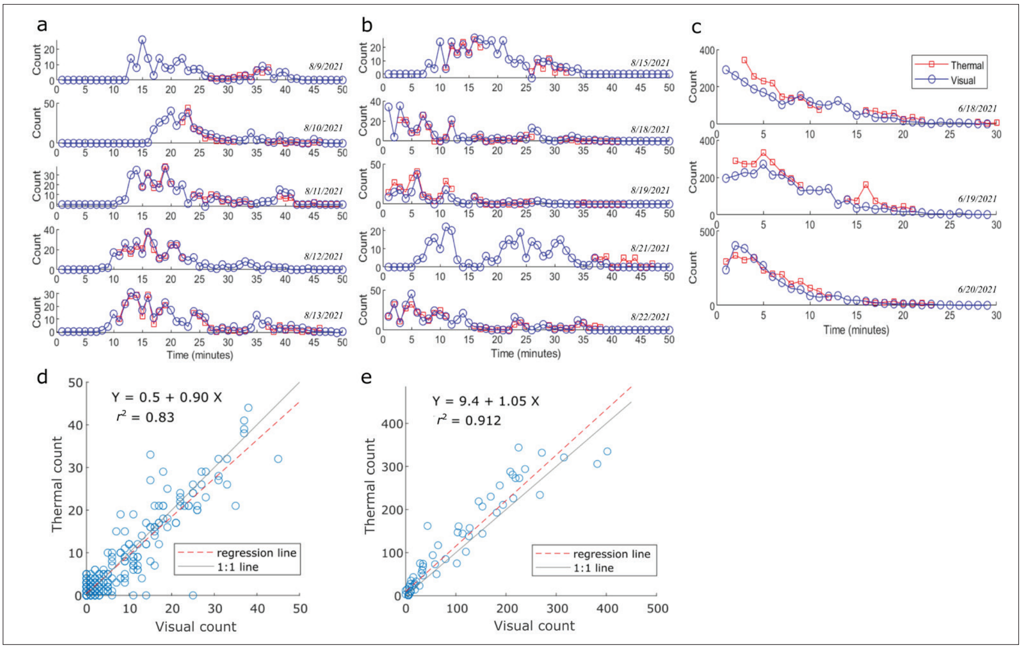

Figure 5. Temporal activity and comparison of observer counts and drone-acquired thermal imagery counts at our 2 field locations. (a, b) Temporal activity of Big Brown bats over 10 survey nights in North Carolina. (c) Temporal activity of Gray Bats during 3 nights in Tennessee. (d, e) Comparison of visual and thermal counts within 1-min intervals (d) North Carolina and (e) Tennessee. Note that data were only available for thermal counts in 15-min intervals because of limited drone battery life, which required the drone to be landed periodically to replace batteries.

Table 1. Comparison of counts from visual observations and from thermal imagery at the Tennessee (TN) and North Carolina (NC) sites in 2021.

TN date	TN visual*	TN thermal	% Difference	NC date	NC visual*	NC thermal	% Difference
18 June	1524	2012	+32%	9 August	28	39	+39%
19 June	1937	2570	+33%	10 August	226	140	-38.1%
20 June	2449	2598	+6%	11 August	236	227	-3.9%
-	-	-	-	12 August	252	241	-4.4%
-	-	-	-	13 August	261	235	-10%
-	-	-	-	15 August	176	162	-8%
-	-	-	-	18 August	197	148	-24.9%
-	-	-	-	19 August	168	262	+56%
-	-	-	-	21 August	8	29	+263%
-	-	-	-	22 August	315	310	-1.6%
Absolute median difference	**-**	**-**	**32%**	**-**	**-**	**-**	**17.5%**

Comparison of visual observation and acoustic counts

Using an exponential model, we found a strong positive correlation between counts obtained by human observers and counts estimated from acoustic recordings (Fig. 6). In North Carolina, RMS SPL had a higher correlation with visual counts ($r2 = 0.51$, $F1, 359 = 398$, $P < 0.0001$) than did median SPL ($r2 = 0.33$, $F1, 359 = 484$, $P < 0.0001$). In contrast, at the Tennessee site, median SPL was better predicted by observer count ($r2 = 0.91$, $F1, 78 = 2220$, $P < 0.0001$) than was RMS SPL ($r2 = 0.54$, $F1, 78 = 180$, $P < 0.0001$). Our use of cross-validated exponential models to predict nightly counts produced median error rates between visual observation and acoustic estimates of 4.7% (range 3.5–21.8%) in Tennessee and 15.1% (range 6.4–144%) in North Carolina (Table 2).

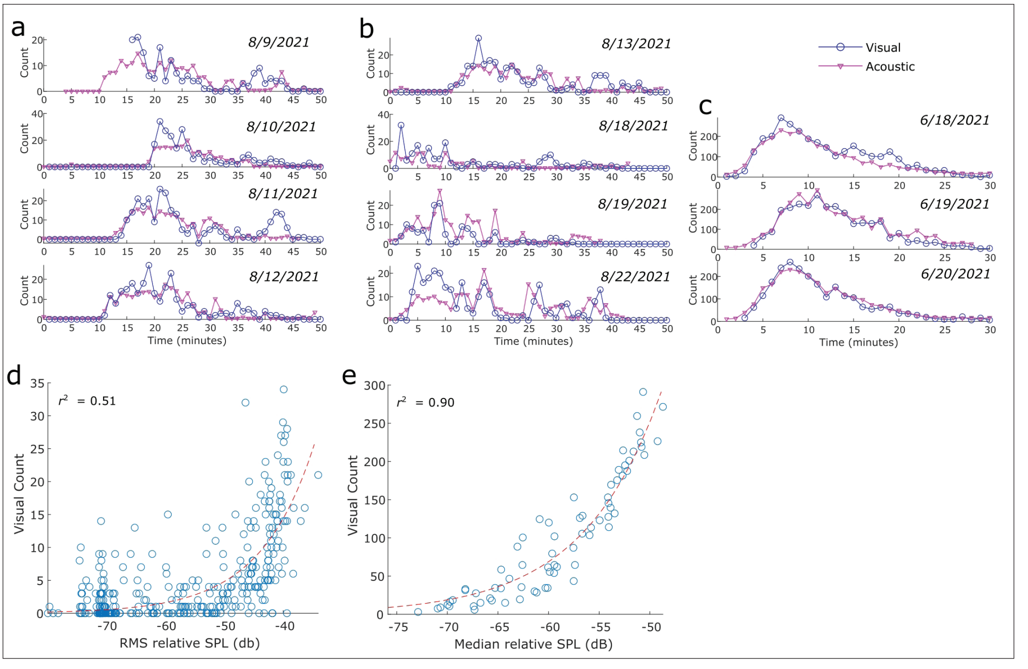

Figure 6. Comparison of acoustic sound pressure levels and visual observation counts. Time courses of activity are shown for (a, b) North Carolina and (c) Tennessee. The relationship between root mean square (RMS) or median Sound Pressure Levels (SPL) is modeled as an exponential equation for (d) North Carolina and (e) Tennessee, respectively. Acoustic levels are shown in decibels (dB) relative to the highest level the acoustic recorders were capable of recording. Red dashed lines indicate the fitted exponential models. Acoustic count values in (a–c) show estimated counts based on the exponential models shown in (d) and (e).

Discussion

We compared the similarity of 3 methods for counting bat emergences at 2 sites with varying conditions. Overall, the 3 methods provided similar counts, with some notable exceptions that we discuss below. Human-human variability at the Tennessee site, for which we had replicated observers, showed an r2 of 0.85 (Fig. 4), with much of the difference coming from a 5-min period on 1 night when the 2 observers differed in their counts by approximately 200%. This discrepancy occurred during a period of high-density emergence (about 200–300 bats/min or 3.3–5.0 bats/sec). The emergence rate when counts differed between observers may approximate an upper limit, above which human observers can no longer count bats with high accuracy (Betke et al. 2008); however, more research is needed in this area, and observer experience may be an important factor in determining accuracy under challenging circumstances. We recommend the use of counter software for visual observation counts in the research and management of bat populations. This approach is simple, inexpensive, and provides valuable additional data in the form of time-course histories for bat emergences.

This was the first study to test drone-acquired thermal imagery for counting emerging bats. Previous researchers used drones with thermal cameras and acoustic detectors to study bat behavior (Fu et al. 2018, Jespersen et al. 2022, Kloepper and Kinniry 2018, Michez et al. 2021) or drones with thermal cameras to count stationary bats roosting in trees (McCarthy et al. 2023, Whitworth et al. 2022). We found high r2 values for predicting drone-acquired thermal counts from concurrent ground-based human counts (r2 = 0.83 and 0.91), based on comparisons of data collected within 1-min intervals (Fig. 5). However, while nightly counts of visual observers and thermal imagery typically differed by 10–30%, counts differed by as much as 263% on individual nights (Table 1). With the approach taken here, we cannot determine to what degree each method contributed to this discrepancy. Nonetheless, in at least some cases, these differences appear to be the result of humans under-counting bats during dark conditions. Overall, our findings indicate that drone-acquired thermal imaging is a promising method for surveying bats.

We have several recommendations based on this initial test of drone-acquired thermal imaging. We used a drone with known limitations, based on the equipment available for this project. For future surveys, a drone with sufficient battery life to fly for an entire emergence is essential; in our case that would be 30–60 min. Our drone had poor flight stability, resulting in shaky thermal images. Unsteady drone flight is commonly referred to as "drift" and can be attributed to lack of a GPS signal, poor calibration, or other mechanical issues. To reduce the likelihood of drift, drones should be regularly inspected for damage, have a balanced payload, and maintain an unobstructed GPS satellite connection. Excessive drift

Table 2. Comparison of visual observation counts and population estimates from acoustic methods at the Tennessee (TN) and North Carolina (NC) sites in 2021.

TN date	TN visual	TN acoustic	% Difference	NC date	NC visual	NC acoustic	% Difference
18 June	2987	2335	-21.8%	9 August	167	156	-6.4%
19 June	2769	2867	+3.5%	10 August	246	212	-13.9%
20 June	2366	2254	-4.7%	11 August	263	241	-8.5%
-	-	-	-	12 August	262	245	-6.6%
-	-	-	-	13 August	255	296	+16.3%
-	-	-	-	18 August	193	236	+22.4%
-	-	-	-	19 August	127	310	+144%
-	-	-	-	22 August	283	392	+38.4%
Absolute median difference	**-**	**-**	**4.7%**	**-**	**-**	**-**	**15.1%**

prevented us from using automated software, such as ThruTracker (Corcoran et al. 2021a) or BatCount (Bentley et al. 2023) for counting bats from thermal images, but an automated counting approach would significantly reduce the time required for video processing. Sufficient thermal contrast with the background is crucial for successfully detecting bats in thermal imagery. Some of the error in our counts may have resulted from bats flying under vegetation, where they could not be detected by the cameras (Jumail et al. 2020, Wang et al. 2019). Bats can also be missed by thermal cameras if the background is at a temperature similar to that of the bats, so care should be taken in selecting sites and testing acquisition of thermal video under different conditions.

Another consideration for thermal imagery is selection of camera resolution, focal length, and altitude of the drone, which collectively determine the pixel size of bats in imagery (Christiansen et al. 2014). In Tennessee, several nights of data had to be discarded when the flight altitude of the drone was increased from 45 to 65 m, indicating that we operated near the detection limit for bats under our conditions. Although concerns arose at 45 m that the drone might affect bat behavior, an examination of the emergence data (Fig. 5c) revealed no unusual patterns in the counts during periods of drone flight compared to non-flight periods. Although we did note noise from drones in audio recordings in Tennessee, this was not the case at the louder North Carolina location. Ambient noise levels should be considered when conducting drone surveys and evaluating their impact on wildlife in the future. Several studies have examined the disturbance caused by drones to bats and other wildlife, highlighting the importance of minimizing such disturbances (Ednie et al. 2021, Mulero-Pázmány et al. 2017, Schad and Fischer 2022). Additionally, we recommend using thermal cameras with a lens of greater focal length and narrower field of view than the one we used, which would allow drones to be flown at higher altitudes (preferably ≥65 m) and still obtain sufficient pixel resolution to image bats.

In our study, using a camera with a 13-mm lens and a resolution of 640 by 512 pixels, we detected bats with a wingspan of 25–35 cm in imagery taken at an altitude of 45 m. Bats in images were typically 9–16 pixels in area or 3–4 pixels in diameter (Fig. 2). A geometric conversion indicates that, under these conditions, 1 pixel equals 5 cm, and therefore, the bats we studied should have appeared as 5–7-pixels wide, if the full wingspan were visible. Although our observed diameter was only 3–4 pixels, a lower value is expected, since the wings of bats are colder than the body (Rummel et al. 2019) and will not always be visible throughout the wingbeat cycle. For future studies, we suggest using half the wingspan for estimating detection range and appropriate flight altitude for thermal imaging of bats. An online tool (Drone Observing Tool) is available for determining the altitude required for detecting animals of a certain size for a given camera resolution and viewing angle (Burke et al. 2019).

In addition to these technical considerations, researchers should adhere to regulations and guidelines concerning drone flights to ensure responsible and legal drone operation. Depending on location, specific regulations may govern flight altitude, airspace restrictions, and the permits required for conducting research with drones. Regulations often vary between national and local authorities and may change over time. Therefore, researchers should stay informed about current rules and ensure compliance before conducting drone operations.

To our knowledge, this was only the third study to test the use of acoustic sound levels for censusing bats at emergence (Eddington et al. 2023, Kloepper et al. 2016). On a minute-by-minute basis, the acoustic results from the Tennessee site were more consistent with visual estimates (Fig. 6; r2 = 0.91) than at the North Carolina site (r2 = 0.51), where there were fewer bats (mean visual observation estimate of 2707 bats in TN, compared to 224 in NC). However, cross-validated nightly estimates had a median error of 4.7% in Tennessee and 15.1% in North Carolina, which suggested that aggregated nightly estimates can be precise even when there is a higher variance in acoustic SPL data, as in North Carolina. The 2 previous tests of this method were conducted at roosts containing 700,000–900,000 bats (Kloepper et al. 2016) and 22,600–38,700 individuals (Eddington et al. 2023). Therefore, our study demonstrated that counts based on sound level yield results similar to those produced with other methods on colonies that were 2 orders of magnitude smaller than previously tested. We did observe some nightly counts with large discrepancies compared to visual counts, such as a 144% difference on 19 August in North Carolina (Table 2). We cannot determine whether this was caused by errors in the human or acoustic counts; however, we recommend counts at smaller emergences be based on multiple nights of data to yield more accurate results.

The RMS SPL method was initially shown to be applicable to emergence rates of 10 bats/sec or higher (Kloepper et al. 2016), but notably, neither the site in North Carolina or Tennessee reached this rate limitation. To account for low emergence rates, we developed a variation of the acoustic-based method that used the median sound level from acoustic files. This technique also accounted for the varying distance of emerging bats from the microphone, which results in animals closer to the microphone being recorded at a higher level than those farther away. The median SPL had a higher correlation with visual counts in Tennessee than in North Carolina, and this difference could be attributed to the differential placement of the acoustic recorders. Positioning the microphone near the roost exit in Tennessee resulted in recordings of many bats flying nearby, a scenario effectively accounted for by the median SPL method. In comparison, the deployment of acoustic detectors farther from the exit in North Carolina resulted in a more uniform sound level of recorded calls. The median SPL method provides an additional tool for conducting SPL-based acoustic bat counts, but the technique should be compared to the RMS method on a case-by-case basis.

Acoustic-based surveys offer notable advantages; they are cost-effective, require little labor, and allow extended deployment periods, ranging from weeks to months. In addition, acoustic methods can be simultaneously used at multiple roost exits, minimizing the need for extensive personnel involvement, compared to traditional count methods. However, some additional labor is required to establish baseline visual or thermal counts. Once an acoustic-based estimate is established for an emergence site, it can enable continuous monitoring of bat roosts throughout the occupancy period, providing an opportunity to record nightly fluctuations in colony size (Barclay et al. 2024). Besides counting bats, acoustics can concurrently serve the traditional purpose of identifying bats to species. We recommend that these baseline counts with other methods be conducted at intervals throughout the survey period, if feasible, to account for potential changes to the acoustic scene or factors that could affect the relationship between number of bats present and acoustic SPL. Ideally, conservation managers should possess some training in acoustics and thoroughly examine files for extraneous noise from insects, different bat species, and other acoustic artifacts that may not be filtered during processing.

In future studies aimed at refining counting methods, several considerations can be explored. Subsequent research could focus on monitoring bat behaviors more closely during drone operations to ascertain potential stress or alterations in emergence patterns. Advancements in both drone and thermal-imaging technologies could provide enhanced flight stability, extended battery life, and increased thermal resolution. These technological improvements could significantly enhance the accuracy of population monitoring while ensuring minimal disturbance. Additionally, acoustic-based estimates could be improved with the implementation of machine learning. Machine-learning models might be better suited than the methods employed here for filtering background noise and handling variation in sound levels of bats while minimizing human error (Mac Aodha et al. 2018).

We conducted an initial comparison of 3 methods for counting bat emergences at 2 sites with varying conditions. Our study should be viewed as preliminary and needs further replication. Nevertheless, our findings support the validity of drone-acquired thermal imagery and acoustic-based estimates for external emergence counts under the conditions that were tested. Each method has unique benefits and limitations, and we support utilization of multiple methods in combination, to provide more confidence in the results than those provided by any single method used in isolation.

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Literature Cited

Anderson, K., and K.J. Gaston. 2013. Lightweight unmanned aerial vehicles will revolutionize spatial ecology. Frontiers in Ecology and the Environment 11:138–146.

Barclay, R.M.R., T.A. Monteiro, E.N. Miller, and L.E. Hiles. 2024. Fluctuations in bat-house colony size may hamper estimation of population changes. Journal of North American Bat Research. Special Issue 1:1–10.

Bentley, I., V. Kuczynska, V.M. Eddington, M. Armstrong, and L.N. Kloepper. 2023. BatCount: A software program to count moving animals. PLoS ONE 18(3):e0278012.

Betke, M., D.E. Hirsh, N.C. Makris, G.F. McCracken, M. Procopio, N.I. Hristov, S. Tang, A. Bagchi, J.D. Reichard, J.W. Horn, S. Crampton, C.J. Cleveland, and T.H. Kunz. 2008. Thermal imaging reveals significantly smaller Brazilian Free-tailed Bat colonies than previously estimated. Journal of Mammalogy 89:18–24.

Burke, C., M. Rashman, S. Wich, A. Symons, C. Theron, and S. Longmore. 2019. Optimizing observing strategies for monitoring animals using drone-mounted thermal infrared cameras. International Journal of Remote Sensing 40:439–467.

Christiansen, P., K.A. Steen, R.N. Jørgensen, and H. Karstoft. 2014. Automated detection and recognition of wildlife using thermal cameras. Sensors 14:13778–13793.

Christie, K.S., S.L. Gilbert, C.L. Brown, M. Hatfield, and L. Hanson. 2016. Unmanned aircraft systems in wildlife research: Current and future applications of a transformative technology. Frontiers in Ecology and the Environment 14:241–251.

Cilulko, J., P. Janiszewski, M. Bogdaszewski, and E. Szczygielska. 2013. Infrared thermal imaging in studies of wild animals. European Journal of Wildlife Research 59:17–23.

Corcoran, A.J., M.R. Schirmacher, E. Black, and T.L. Hedrick. 2021a. ThruTracker: Open-source software for 2-D and 3-D animal video tracking. BioRxiv Preprint. 2021.05.12.443854 https://doi.org/10.1101/2021.05.12.443854

Corcoran, E., M. Winsen, A. Sudholz, and G. Hamilton. 2021b. Automated detection of wildlife using drones: Synthesis, opportunities and constraints. Methods in Ecology and Evolution 12:1103–1114.

Davis, D.E., and R.L. Winstead. 1980. Estimating the numbers of wildlife populations. Pp. 221–245, In S.D. Schemnitz (Ed.). Wildlife Management Techniques Manual. The Wildlife Society, Bethesda, MD. 686 pp.

Ednie, G., D.M Bird, and K.H. Elliott. 2021. Fewer bat passes are detected during small, commercial drone flights. Scientific Reports 11:11529

Elliott, W.R., S.T. Samoray, and S.E. Gardner. 2005. The MDC method: Counting bats with infrared video. Pp. 147–53, In S. Stokowski and G.T. Rea (Eds.). Proceedings of the 2005 National Cave and Karst Management Symposium. The NCKMS Steering Committee, Albany, NY. 258 pp.

Elliott, W.R., D. Shiels, S. Colatskie, and C. Dzurick. 2011. Gray Bat (Myotis grisescens) thermal infrared monitoring in Missouri, 2008–2011. Missouri Department of Conservation, Jefferson City, MO. 17 pp.

Frank, J.D., T.H. Kunz, J. Horn, C. Cleveland, and S. Petronio. 2003. Advanced infrared detection and image processing for automated bat censusing. Pp. 261–271, In B.F. Andresen and G.F. Fulop (Eds.). Infrared Technology and Applications. Volume 5074. Available online at https://doi.org/10.1117/12.499422. Accessed 14 August 2024

Fu, Y., M. Kinniry, and L.N. Kloepper. 2018. The Chirocopter: A UAV for recording sound and video of bats at altitude. Methods in Ecology and Evolution 9:1531–1535.

Hodgson, J.C., R. Mott, S.M. Baylis, T.T. Pham, S. Wotherspoon, A.D. Kilpatrick, R.R. Segaran, I. Reid, A. Terauds, and L.P. Koh. 2018. Drones count wildlife more accurately and precisely than humans. Methods in Ecology and Evolution 9:1160–1167.

Hristov, N.I., M. Betke, and T.H. Kunz. 2008. Applications of thermal infrared imaging for research in aeroecology. Integrative and Comparative Biology 48:50–59.

Israel, M. 2011. A UAV-based Roe Deer fawn detection system. Pp. 51–55, In H. Eisenbeiss, M. Kunz, and H. Ingensand (Eds.). ISPRS Archives. Volume XXXVIII-1/C22. Zurich, Switzerland. 314 pp.

Jespersen, C., D. Docherty, J. Hallam, C. Albertsen, and L. Jakobsen. 2022. Drone exploration of bat echolocation: A UAV□borne multimicrophone array to study bat echolocation. Ecology and Evolution 12:e9577.

Jumail, A., L. Thor-Seng., M. Salgado-Lynn, K.M. Fornace, and D.J. Stark. 2020. A comparative evaluation of thermal camera and visual counting methods for primate census in a riparian forest at the Lower Kinabatangan Wildlife Sanctuary (LKWS), Malaysian Borneo. Primates 62:143–151.

Kloepper, L.N., and M. Kinniry. 2018. Recording animal vocalizations from a UAV: Bat echolocation during roost re-entry. Scientific Reports 8:7779.

Kloepper, L.N., M. Linnenschmidt, Z. Blowers, B. Branstetter, J. Ralston, and J.A. Simmons. 2016. Estimating colony sizes of emerging bats using acoustic recordings. Royal Society Open Science 3:160022.

Kunz, T.H., M. Betke, N.I. Hristov, and M.J. Vonhof. 2009. Methods for assessing colony size, population size, and relative abundance of bats. Pp. 133–157, In T.H. Kunz and S. Parsons (Eds.). Ecological and Behavioral Methods for the Study of Bats. 2nd Edition. Johns Hopkins University Press, Baltimore, MD. 990 pp.

Lethbridge, M., M. Stead, and C. Wells. 2019. Estimating kangaroo density by aerial survey: A comparison of thermal cameras with human observers. Wildlife Research 46:639–648.

Linchant, J., J. Lisein, J. Semeki, P. Lejeune, and C. Vermeulen. 2015. Are unmanned aircraft systems (UASs) the future of wildlife monitoring? A review of accomplishments and challenges. Mammal Review 45:239–252.

Loeb, S.C., T.J. Rodhouse, L.E. Ellison, C.L. Lausen, J.D. Reichard, K.M. Irvine, T.E. Ingersoll, J.T.H. Coleman, W.E. Thogmartin, J.R. Sauer, C.M. Francis, M.L. Bayless, T.R. Stanley, and D.H. Johnson. 2015. A plan for the North American Bat Monitoring Program (NABat). U.S. Department of Agriculture Forest Service, Southern Research Station, General Technical Report SRS-208:1–100.

Mac Aodha, O., R. Gibb, K.E. Barlow, E. Browning, M. Firman, R. Freeman, B. Harder, L. Kinsey, G.R. Mead, S.E. Newson, I. Pandourski, S. Parsons, J. Russ, A. Szodoray-Paradi, F. Szodoray-Paradi, E. Tilova, M. Girolami, G. Brostow, and K.E. Jones. 2018. Bat detective—deep learning tools for bat acoustic signal detection. PLOS Computational Biology 14(3):e1005995.

McCarthy, E.D., J.M. Martin, M.M. Boer, and J.A. Welbergen. 2023. Ground-based counting methods underestimate true numbers of a threatened colonial mammal: An evaluation using drone-based thermal surveys as a reference. Wildlife Research 50:484–493.

McCracken, G.F. 2003. Estimates of population sizes in summer colonies of Brazilian Free-tailed Bats. Pp. 21–30, In T.J. O'Shea and M.A. Bogan (Eds.). Monitoring Trends in Bat Populations of the Unites States and Territories: Problems and Prospects. Information and Technology Report USGS/BRD/ITR-2003-0003. 287 pp.

Michez, A., S. Broset, and P. Lejeune. 2021. Ears in the sky: Potential of drones for the bioacoustic monitoring of birds and bats. Drones 5:9.

Milchram, M., M. Suarez-Rubio, A. Schröder, and A. Bruckner. 2020. Estimating population density of insectivorous bats based on stationary acoustic detectors: A case study. Ecology and Evolution 10:1135–1144.

Mulero-Pázmány, M., S. Jenni-Eiermann, N. Strebel, T. Sattler, J.J. Negro, and Z. Tablado. 2017. Unmanned aircraft systems as a new source of disturbance for wildlife: A systematic review. PloS One 12(6):e0178448.

Mulero-Pázmány, M., R. Stolper, L.D. van Essen, J.J. Negro, and T. Sassen. 2014. Remotely piloted aircraft systems as a rhinoceros anti-poaching tool in Africa. PLoS ONE 9:e83873.

Rummel, A.D., S.M. Swartz, and R.L. Marsh. 2019. Warm bodies, cool wings: Regional heterothermy in flying bats. Biology Letters 15(9):20190530.

Sabol, B.M., and M.K. Hudson. 1995. Technique using thermal infrared-imaging for estimating populations of Gray Bats. Journal of Mammalogy 76:1242–1248.

Sardà-Palomera, F., G. Bota, C. Viñolo, O. Pallarés, V. Sazatornil, L. Brotons, S. Gomáriz, and F. Sardà. 2012. Fine-scale bird monitoring from light unmanned aircraft systems. Ibis 154:177–183.

Schad, L., and J. Fischer. 2022. Opportunities and risks in the use of drones for studying animal behaviour. Methods in Ecology and Evolution 14:1864–1872.

Schneider, C.A., W.S. Rasband, and K.W. Eliceiri. 2012. NIH Image to ImageJ: 25 years of image analysis. Nature Methods 9:671–675.

Seymour, A.C., J. Dale, M. Hammill, P.N. Halpin, and D.W. Johnston. 2017. Automated detection and enumeration of marine wildlife using unmanned aircraft systems (UAS) and thermal imagery. Scientific Reports 7:45127.

Speakman, J.R, D.J. Bullock, L.A. Eales, and P.A. Racey. 1992. A problem defining temporal pattern in animal behaviour: Clustering in the emergence behaviour of bats from maternity roosts. Animal Behaviour 43:491–500.

Tsukanov, R. 2021. Counter. Version 22, May 2020. Available online at https://play.google.com/store/apps/details?id=me.tsukanov.counter. Accessed 15 May 2021.

Vallecillo, D., M. Gauthier□Clerc, M. Guillemain, M. Vittecoq, P. Vandewalle, B. Roche, and J. Champagnon. 2021. Reliability of animal counts and implications for the interpretation of trends. Ecology and Evolution 11:2249–2260.

Wang, D., Q. Shao, and H. Yue. 2019. Surveying wild animals from satellites, manned aircraft and unmanned aerial systems (UASs): A review. Remote Sensing 11(11):1308.

Whitworth, A., C. Pinto, J. Ortiz, E. Flatt, and M. Silman. 2022. Flight speed and time of day heavily influence rainforest canopy wildlife counts from drone-mounted thermal camera surveys. Biodiversity and Conservation 31:3179–3195.