FINAL REPORT

South Carolina Competitive State Wildlife Grant

SC-U2-F14AP00958

South Carolina Department of Natural Resources

October 1, 2015 – September 30, 2016*

* *This grant was recently extended to December 28, 2017 for reporting purposes.*

Project Title: **Carolinas Acoustic Bat Survey**

Objective 1: We proposed to employ the North American Bat Monitoring Program (NABat) (Loeb *et al*. 2015) in North and South Carolina. The NA Bat guidelines had not been published when the grant was awarded but the national grids had been established by USGS. Then, NABat guidelines were published in 2015. The NABat sampling frame consists of a GIS-generated sampling grid across North America of 10x10 km grid cells. The states' Wildlife Action Plans have determined that many of our bat species are a high priority (SGCN), and this project will provide bat population information that is comparable to data that are collected nationwide.

Accomplishments: We employed the NABat program in North and South Carolina in 2015 and 2016, sampling a total of 95 grid cells.

South Carolina Summary

In 2015 and 2016, we established surveys in 38 grid cells: mobile transect surveys were established in 30 cells, and stationary point surveys were established in 25 cells. In the stationary cells, a total of 44 unique locations were created. In 2015, 35 total cells were surveyed; 15 with mobile transects only, six with stationary points only, and 14 with both methods. During this time, stationary points were surveyed on 147 occasions. In 2016, 38 total cells were surveyed; 13 with mobile transects only, eight with stationary points only, and 17 with both methods. During this time, stationary points were surveyed on 200 occasions. Using acoustic data collected from these efforts, over 400 calls were identified through bat call software and manual vetting, resulting in 10 species and 3 species groups identified. Four species were found outside their known range (including the federally threatened northern long-eared bat), with only one species known to occur in SC not detected during our surveys. Detection probabilities were also determined for both survey methods, with stationary surveys showing higher detection probability than mobile surveys. Lastly, calls identified through Echoclass 3.1 and Kaleidoscope 3.1.5 programs were compared to manual vetting, and false-positive and false-negative errors were shown to exist in both programs, varying depending on species identification, survey duration, and survey method. Thus, manual vetting of call identifications must be done regardless of the program used for call classifications. For further information and complete results, see Ben Neece's thesis in Appendix A.

North Carolina Summary

In 2015 and 2016, we visited 57 highly ranked NABat grid cells and developed mobile transect surveys for 41 grid cells and stationary point surveys for 40 grid cells. Through over 300,000 acoustic files, we generated NABat grid cell specific presence/no detection information. Fourteen species of bats were detected, and both mobile and stationary surveys revealed similar general patterns of bat distributions in NC. We also generated species specific activity density maps via ArcGIS mapping. For federally listed species, MYSO were concentrated mainly in the Cherokee National Forest near the NC/TN border; MYGR were mainly distributed along the Appalachian Mountains between Asheville and Boone; and more MYSE

were found in the coastal plain region than the rest of NC. We suggest that the future implementation of NABat should involve multiple partners with a centralized coordinator to facilitate participation and integrate data. We also have issues of concern that need to be addressed in the future. For further information and complete results, see the final report created by the University of North Carolina at Greensboro for the North Carolina Wildlife Resources Commission in Appendix B.

Significant deviations:

There were no significant deviations.

Objective 2: Development of an identification model for bat recordings of three underrepresented South Carolina species is another objective essential to this project. Current models often do not correctly identify the Southeastern bat (*Myotis austroriparius*), the Northern yellow bat (*Lasiurus intermedius*), and Brazilian (Mexican) free-tailed bat (*Tadarida brasiliensis*); the former two are species of special concern and are a high priority within both states' Action Plans. Current models in wide use will label these species as unknown or will sometimes misidentify them as other species such as big brown bats.

Accomplishments:

Ben Neece gathered additional recordings of these underrepresented bats. Both Kaleidoscope and Echoclass produce some errors in classifications of bat calls. Echoclass produced more false-positives of southeastern bats than Kaleidoscope (and they both over-report relative to manual vetting). Through manual vetting, yellow bats were detected in 3 of the cells sampled in 2015. Free-tailed bats were detected in twenty-eight, and southeastern bats were detected in eight of 36 cells surveyed in 2015. For further information and complete results, see Ben Neece's thesis in Appendix A.

Significant deviations:

Though additional recordings of underrepresented bats were collected, and an assessment of Echoclass and Kaleidoscope accuracy with southeastern bat calls was conducted, we were unable to create identification models. Also, Palmetto Bluff Conservancy was unable to record northern yellow bat calls to share with SCDNR and Ben Neece.

Literature Cited: See appendices.

Estimated Federal Cost: SC: $75,480.00; NC: $190,000.00

Recommendations: Close the grant. We need to emphasize the limitations of automated classifications of bat calls. It has become very clear through this project that for accurate bat call identification, there is no substitute for manual vetting.

Submitted by Jennifer Kindel, SCDNR on 12/8/2017

Appendix A: (North American Bat Monitoring Program (Nabat) In South Carolina: Acoustic Detection and Landscape Occupancy Of Bats)

Appendix B: Final Report for Carolinas Regional Acoustic Bat Survey, NABat in North Carolina

NORTH AMERICAN BAT MONITORING PROGRAM (NABAT) IN SOUTH CAROLINA: ACOUSTIC DETECTION AND LANDSCAPE OCCUPANCY OF BATS

A Thesis
Presented to
the Graduate School of
Clemson University

In Partial Fulfillment
of the Requirements for the Degree
Master of Science
Wildlife and Fisheries Biology

---

by
Benjamin D. Neece
December 2017

Accepted by:
David S. Jachowski, Committee Chair
Catherine M. Bodinof Jachowski
Yoichiro Kanno
Susan C. Loeb

ABSTRACT

Bats are under threat from habitat loss, energy development, and the disease white-nose syndrome. The North American Bat Monitoring Program (NABat) suggests standardized, large scale monitoring to benefit ecologists and managers. Our first objective was to determine the efficacy of NABat in South Carolina. Detection probabilities differ within and among species and among survey conditions. Thus, our second objective was to determine factors affecting detection probabilities. Finally, effective management strategies addressing large scale threats require landscape scale analyses. Thus, our third objective was to conduct state-wide assessments of environmental factors influencing landscape occupancy and generate predicted distributions.

We conducted NABat acoustic surveys across South Carolina from mid-May through July 2015 and 2016. To determine the efficacy of NABat, we compared species detections to known distributions based on historical records, and to predicted distributions based on environmental occupancy models. We detected some species throughout their ranges and others in $\leq 50\%$ of cells within their ranges, and detected some species outside their ranges. Thus, NABat monitoring may be suitable for many species but may not be suitable for species with echolocation calls that are difficult to detect or identify, and may also reveal new information about species distributions.

To determine factors that affected detection, we evaluated support for detection models. We found that detection covariates greatly varied among species, but most species had higher detection probabilities at stationary points than mobile transects. Our

results suggested that effects of factors on detection probabilities were based on biological and behavioral characteristics of species, which indicated the importance of monitoring survey variables and accounting for them in analyses.

To assess effects of environmental factors on occupancy, we evaluated temporally dynamic occupancy models. Occupancy probability differed among ecoregions for northern yellow bats (*Dasypterus intermedius*) and *Myotis* species. Hoary bats (*Lasiurus cinereus*) were negatively associated with forest edge density. We found no significant effects of habitat conditions for five species. Thus, for some species, site-use analyses of NABat data may be more appropriate than grid-based occupancy analyses. However, predicted distributions closely matched species habitat associations. Our findings can improve future monitoring efforts and inform conservation priorities.

ACKNOWLEDGMENTS

First, I would like to thank my advisors, Dr. Susan Loeb and Dr. David Jachowski, for offering the opportunity to further my career in wildlife ecology. They have been excellent mentors, essential to my professional development, and supportive of my career goals. I would like to acknowledge my committee members, Dr. Catherine Bodonif Jachowski and Dr. Yoichiro Kanno, who provided great feedback and substantially improved my analytical abilities. I also want to thank Mary Bunch for her collaborative efforts which helped initialize and support this research. This research would not have been possible without funding from a U.S. Fish and Wildlife Service Competitive State Wildlife Grant (SC-U2-F14AP00958).

I am thankful for the support of the South Carolina Department of Natural Resources, U.S. Forest Service, U.S. Fish and Wildlife Service, U.S. Army Corps of Engineers, South Carolina Forestry Commission, South Carolina Department of Parks, Recreation, and Tourism, Folk Land Management, Naturaland Trust, Greenville Water, and other agencies, organizations, and volunteers. I would especially like to thank Johnny Stowe, Rob Harrison, Donald Cockman, Daniel Belken, Daniel Jones, and Ashley Watford for devoting their time collecting data. I would also like to thank my fellow graduate students and friends for their feedback and camaraderie. Finally, I want to thank my parents and family for always supporting my interests and helping develop my love of nature.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER ONE

AN ASSESSMENT OF THE NORTH AMERICAN BAT MONITORING PROGRAM IN SOUTH CAROLINA

Bat populations in North America are currently under stress from a number of major threats (Loeb et al. 2015, Pauli et al. 2015). White-nose syndrome (WNS) has caused severe declines in hibernating bat populations since 2007 in the northeastern United States (Turner and Reeder 2009). The epidemic has continued to spread across the East and Midwest regions, and was recently discovered in the western state of Washington (Lorch et al. 2016). Additionally, bat populations are being impacted by energy development (Kunz et al. 2007, Horn et al. 2008), habitat alteration, and climate change (Jones et al. 2009, Rebelo et al. 2010).

To develop effective conservation strategies for bat populations, an appropriate method of monitoring must be established. Acoustic monitoring has become a common method of monitoring bat populations, due to the ease of the equipment setup and low personnel requirement. Relative to more traditional methods such as mist netting, acoustic monitoring requires no bat handling and few or no permits. Therefore, it is relatively easy to conduct acoustic surveys of bats in a variety of habitat types (Murray et al. 1999, Britzke et al. 2013).

The North American Bat Monitoring Program (NABat) was developed to provide standardized methods to monitor bat populations across the continent (Loeb et al. 2015). Surveys can be implemented from local to range-wide scales and researchers can analyze their data to make inferences about local populations and develop suitable management

strategies. Data can be submitted to a national database and, with NABat's standardized site selection and sampling methods, large scale analyses of changes in bat relative abundance and distributions are possible (Loeb et al. 2015). NABat guidelines were released in 2015, when we began our study. Thus, our first objective was to determine the efficacy of NABat methods by implementing the suggested protocols within South Carolina and comparing species detection locations to their known distributions based on historical survey records, and to their predicted distributions based on landscape occupancy models.

Additionally, the probability of detecting bats with acoustic surveys varies within and among bat species (Yates and Muzika 2006, Hein et al. 2009, Bender et al. 2015) and may be affected by factors that vary among survey occasions such as weather as well as factors that vary across sites such as habitat condition. Variation in detection probability can affect the level of sampling effort needed (i.e., high variation may require more sampling effort) to detect some species (Law et al. 2015). Also, a failure to detect a species when it is present (i.e., a false negative) can misinform management of critical habitat (MacKenzie 2005). Therefore, because detection probability should be accounted for in analyses of NABat acoustic data, our second objective was to determine which factors significantly affect detection probabilities for each species of bat in South Carolina. We expect results of this study to help improve implementation of NABat acoustic surveys by showing which factors affect detection probabilities so they may be taken into account in future studies and monitoring efforts.

METHODS

Study Location

We collected data throughout the state of South Carolina, which consists of five major physiographic regions in a gradient from northwest to southeast: Blue Ridge, Piedmont, Southeastern Plains, Middle Atlantic Coastal Plain, and Southern Coastal Plain (U.S. Environmental Protection Agency 2011). We also collected a small amount of data in bordering areas of Georgia and North Carolina. Land usage in South Carolina includes various intensities of urban development, silviculture, agriculture, livestock, and undeveloped land. Land cover in the Blue Ridge is dominated by deciduous, evergreen, and mixed forests; the Piedmont by deciduous and evergreen forests and hay or pasture; the Southeastern Plains and Middle Atlantic Coastal Plains by woody wetlands, evergreen forests, shrub lands, and cultivated crops; and the Southern Coastal Plain by emergent herbaceous wetlands, woody wetlands, evergreen forests, and open water (Homer et al. 2015). Mountainous landscapes in the northwestern part of the state have the highest elevations, up to 1085 m, which quickly fade into lower elevation plains for much of the central region and become low-lying wetlands near the coast.

Fourteen bat species are known to occur in South Carolina, 12 of which are considered species of greatest conservation need by the State Wildlife Action Plan (South Carolina Department of Natural Resources 2015). Some species are found throughout the state, while others have more restricted ranges (Menzel et al. 2003). The northern long-eared myotis (*Myotis septentrionalis*; MYSE), was recently listed as a threatened species under the U.S. Endangered Species Act due to declines from WNS (Federal Register

2015). WNS is also severely impacting little brown bat (*M. lucifugus*; MYLU) and tricolored bat (*Perimyotis subflavus*; PESU) in South Carolina, and can infect eastern small footed bat (*M. lebeii*; MYLE) and big brown bat (*Eptesicus fuscus*; EPFU) (United States Fish and Wildlife Service 2014); however, the latter two species are not experiencing significant declines due to WNS (Langwig et al. 2012).

Sampling Design

We utilized the NABat continent-wide grid of 10 x 10 km cells to identify priority cells for acoustic surveys within South Carolina. The sampling design for NABat is the generalized random tessellation stratified (GRTS) algorithm, which assigns priority numbers to cells to maintain a spatially balanced, random sample (Loeb et al. 2015). NABat guidelines suggest each cell should have one mobile transect and two to four stationary points and recommend sampling at least 30 cells within each state. We followed this recommendation and selected the top 30 priority cells from the NABat master sample for South Carolina (https://www.sciencebase.gov/catalog/item/569e64b4e4b0961cf27ec85d).

Mobile Transects

We followed NABat guidelines requirements for mobile transect routes to develop routes 25-48 km in length that were primarily contained within the cells, passed through common habitat types of the area, did not come within 100 m of another section of themselves, were safe to drive at 32 kph at night, required minimal stopping, passed through no stoplights, and did not include roads with heavy traffic, gates to open and close, or sections where driving at 32 kph was dangerous (Loeb et al. 2015). When

selecting roads for transect routes, we utilized the National Transportation Dataset (NTD) RoadSegment data (USGS, National Geospatial Technical Operations Center 2014) and filtered it to secondary and tertiary road classes because these typically meet the transect criteria. We also examined the National Forest System Roads (U.S. Forest Service 2015*a*) because some of these road segments are not included in the NTD and may be suitable for transects. We categorized habitat types within the cells using the National Land Cover Database (NLCD; U.S. Geological Survey 2014) and made certain transects passed through or adjacent to common habitat types in each cell. Finally, we cross-checked the GIS data by examining roads in Google Maps (Google n.d.) and used Google Street View to make sure routes did not pass through stoplights. If cells were not suitable for mobile transects, we dropped them from the sample or surveyed them with stationary detectors only, and selected replacement cells sequentially from the GRTS list until a total of 30 transects could be developed.

Stationary Points

We followed the NABat criteria for stationary sampling points and attempted to find sites which maximized the quality of recordings as well as the diversity of species detected. We sought 2-4 points per cell and ideally, one point in each quadrant of a cell, or in different habitats in cells that had heterogeneous habitat types. To select sites for stationary point surveys, we used the NLCD to examine habitat types within each cell and the U.S. Forest Service BasicOwnership database (U.S. Forest Service 2015*b*) to identify public lands. We also viewed aerial imagery from Google Maps (Google n.d.) to examine land cover and vegetation structure so the most appropriate survey locations

could be determined based on two criteria. First, during summer, bat species in South Carolina commonly roost in trees and shrubs or in human structures and fly along edges to reach water and foraging areas (Menzel et al. 2003). Therefore, we typically sought out forest edges and water sources. Second, for long-term monitoring, access to the same sites is needed for many years. Therefore, we prioritized sites on public land. However, few cells contained public lands, and we found it necessary to also secure permission to survey private lands.

Survey Equipment

For both mobile transect and stationary point acoustic surveys we used Anabat SD2 bat detectors with directional, stainless steel microphones (Titley Scientific, Columbia, MO, USA) and 2.5 m microphone cables. We used 10 detectors for stationary surveys and four for mobile surveys. Before each survey season, we calibrated detector sensitivities using the Anabat Equalizer (Titley Scientific, Columbia, MO, USA). We set the internal sensitivity to 30% and kept detectors, microphones, and cables together throughout the season to retain calibrations.

For mobile surveys, we powered the detector with the vehicle's power outlet and kept it inside the cabin during operation to monitor its functionality throughout the surveys. The microphone was attached via suction mount to the center of the roof near the front and was oriented straight up from the roof of the vehicle, with no housing or weatherproofing; the surface of the microphone was 18.5 cm above the vehicle's roof. We attached a Garmin 18x PC GPS Navigator Unit (Garmin 2017) to the vehicle's roof adjacent to the microphone to log coordinates each second of the mobile survey.

For stationary point surveys, the detector was housed in a steel, waterproof ammunition case along with a 12 V battery to supply power. To waterproof the microphone, we placed it at one end of a 3.8 cm PVC tube with a 45° slip coupling elbow. The microphone and housing were attached to the top of a 1.8 m camera tripod, with the opening of the PVC horizontal and set 3 – 5 m from clutter and oriented away from it. We took 360° panoramic photographs of the area surrounding each stationary point and recorded the microphone's bearing to ensure this remained constant between survey years.

Survey Timing

Surveys were conducted mid-May through July in 2015 and 2016. To efficiently utilize and distribute survey equipment and complete surveys within the sampling season, we grouped two to six neighboring cells into nine weekly survey areas. Parturition dates in temperate bats are related to temperature (Racey 1982). Because NABat surveys should be completed before young become volant (Loeb et al. 2015), we began surveys in the southeastern-most cells and proceeded north and west through the state, with the final surveys occurring in the Blue Ridge region in the northwest.

All stationary points were surveyed for four consecutive nights from 30 minutes prior to sunset to 30 minutes after sunrise. Each mobile transect was surveyed twice during this period, beginning 45 minutes after sunset, with the same start and end locations. The same points and transects were surveyed in both 2015 and 2016, where possible. If it rained or wind speed was > 10 kph during a mobile survey, we paused for

up to 15 minutes to allow conditions to improve. If they did not, we ended the survey at that location and made another attempt to survey the entire transect later in the week.

Data Processing

We removed acoustic files that contained no bat calls using a custom noise filter in AnalookW version 4.2.7 (AnalookW 2016), and then manually removed files which contained fewer than three bat call pulses. For 2015 data, we classified the remaining files using EchoClass version 3.1 and Kaleidoscope Pro version 3.1.5, and then manually vetted all classifications based on reference calls of each species. We often observed misclassifications and low agreement between the two automated classifiers. Thus, for 2016 data, we did not use classification software and instead manually classified all high quality calls. We used reference calls that were recorded from identified captured bats which were light-tagged (Britzke et al. 2011).

Data Analysis

Efficacy of NABat

We used two methods to evaluate the efficacy of NABat. If NABat acoustic surveys are a good approach to monitoring bat species in South Carolina, we expected to detect species in each cell within their known distributions. Thus, we compared our detections with previously known species ranges throughout the state (Menzel et al. 2003). However, if species distributions have shifted in South Carolina due to habitat changes and the presence of WNS, or if historic surveys were insufficient, distribution maps from 2003 may not be accurate. Thus, we also compared detections with predicted distribution maps that we developed with landscape occupancy models (see Chapter 2),

where we treated cells with ≥ 50% predicted occupancy as within the distribution of a species. For both 2003 known ranges and predicted distributions, we determined the percentage of the cells surveyed within each species range in which it was detected. We considered NABat acoustic surveys an effective method for monitoring a species if the percentage was ≥ 50% for either 2003 known range or predicted distribution. Detections of species outside of their 2003 known ranges provides new information for effective bat conservation and habitat management and is another measure of the efficacy of NABat; therefore, we also counted the number of cells outside of the 2003 known range in which each species was detected.

Factors Affecting Probability of Detection

We used a Bayesian occupancy modeling approach to evaluate the relative importance of hypothesized environmental and survey factors on the detection probability for each species. We first created presence/non-detection tables for each species on each survey occasion within each cell. We treated one night at a stationary point as a survey occasion and one mobile transect survey as a separate survey occasion, even if they occurred on the same night. This allowed us to compare the effects of survey method on detection probabilities.

The type of acoustic survey, mobile transect or stationary point, is known to influence detection probability of bat species. Some studies comparing the two methods have found higher probabilities of detection at stationary points (Tonos et al. 2014, Whitby et al. 2014) and others have found higher probabilities of detection on mobile transects (Fisher-Phelps et al. 2017). Because NABat stationary point surveys last all

night and mobile transect surveys are approximately one hour, we hypothesized that detection probabilities would be higher at stationary points compared to mobile transects for all species in our study (Table 1.1). We used two approaches to test this hypothesis. One approach utilized a categorical covariate designating either mobile transect or stationary point for each occasion. The second approach used the duration of each survey occasion in minutes. Because of the differences in duration of mobile transects and stationary point surveys, this variable was another comparison of the two methods, but also considered variation within each survey method.

Vegetation clutter, such as dense forest and shrub stands, can also cause differences in detection probabilities within and among bat species. Bat morphological and call structure adaptations to clutter differ among species (Menzel et al. 2005), so species abundances may vary by the amount of clutter, which could result in variation in probabilities of detection among species based on clutter amount. Additionally, sound transmission is affected by the amount of clutter, and the effects vary by echolocation call frequency (Patriquin et al. 2003). Therefore, even if the abundance of a species is not affected by vegetation clutter, the probability of detecting it could still vary by clutter amount. Both open-adapted and clutter-adapted bat species occur in South Carolina and the areas we surveyed varied in vegetation clutter amount. Thus, we hypothesized increasing vegetation clutter around stationary points would decrease detection probabilities for open-adapted species, but would not affect detection of clutter-adapted species. To test this hypothesis, we created a categorical covariate based on the vegetation cover surrounding each detector as viewed in the panoramic photos. Because

vegetation clutter varies along mobile transects, we used those as the reference value (0). We considered points near large open areas as low clutter (1), points with some clutter and some open areas as medium clutter (2), and points in dense vegetation as high clutter (3).

Reproductive phenology and seasonal activity patterns affect bat activity (Hayes 1997), and therefore, survey date could affect detection probability. Because our surveys were conducted within a few months and before young became volant, we hypothesized that survey date would not affect detection probabilities of species distributed statewide. However, because we moved across the state as we surveyed, we hypothesized that detection probability of species with limited distributions would be positively affected by survey date if areas surveyed later in the season were in their known range, and negatively affected by survey date if areas surveyed earlier in the season were in their known range.

Equipment malfunction and weather conditions could also influence detection probabilities. We hypothesized that an equipment malfunction (e.g., a stationary detector was knocked over, not functioning properly upon retrieval), or an incomplete mobile transect survey would result in lower probability of detection. Bat activity tends to increase with increasing temperature (O'Donnell 2000, Broders et al. 2006, Kitzes and Merenlender 2014, Wolbert et al. 2014). Therefore, we hypothesized that the probability of detection for all species would increase as temperature increased. Relative humidity affects the attenuation of sound waves (Bass et al. 1990) and may both positively and negatively affect the detection of bats (Starbuck 2013). Thus, we tested whether it had an

effect for any species in our surveys. Increasing wind speed decreases the probability of detection (O'Farrell et al. 1967, Rydell 1989), and the occurrence of rain can reduce bat activity (Loeb et al. 2015, Appel et al. 2017). Therefore, we also hypothesized that wind and rain would negatively affect detection probabilities of all species (Table 1.1). To test our hypotheses, we obtained data from the nearest Meteorological Terminal Aviation Routine Weather Reports (METARs) stations to each cell. We used the mean temperature, relative humidity, and wind speed over each survey period, and created a categorical covariate for whether or not it rained during the survey.

Table 1.1: Predicted effects of each covariate on the probability of detection for each species. Symbols indicate positive (+), negative (-), no (0), and unknown (?) effect. *Type* is mobile survey (0) or stationary point survey (1). *Duration* is the length of the survey period in minutes. *Clutter* is categorical with mobile transect (0) and low (1), medium (2), or high (3) vegetation clutter stationary point. *Date* is Julian day of the survey occasion. *Temp* (temperature), *RH* (relative humidity), and *Wind* (wind speed) are mean values during the survey period. *Rain* is categorical with either no rain (0) or rain (1) during the survey period. Species codes are as follows: *Corynorhinus rafinesquii* (CORA), *Dasypterus intermedius* (DAIN), *Eptesicus fuscus* or *Lasionycteris noctivagans* (EPFULANO), *Lasiurus borealis* or *L. seminolus* (LABOLASE), *L. cinereus* (LACI), *Myotis austroriparius* (MYAUS), *M. leibii* (MYLE), *M. lucifugus* (MYLU), *M. septentrionalis* (MYSE), *Nycticeius humeralis* (NYHU), *Perimyotis subflavus* (PESU), and *Tadarida brasiliensis* (TABR).

Species	Type	Duration	Clutter	Date	Temp	RH	Wind	Rain
CORA	+	+	-	0	+	?	-	-
DAIN	+	+	-	-	+	?	-	-
EPFULANO	+	+	-	0	+	?	-	-
LABOLASE	+	+	0	0	+	?	-	-
LACI	+	+	-	+	+	?	-	-
MYAUS	+	+	0	-	+	?	-	-
MYLE	+	+	0	+	+	?	-	-
MYLU	+	+	0	+	+	?	-	-
MYSE	+	+	0	+	+	?	-	-
NYHU	+	+	0	0	+	?	-	-
PESU	+	+	0	0	+	?	-	-

TABR	+	+	-	0	+	?	-	-

We used a Bayesian approach to fit detection models for each species independently while holding occupancy constant. We used non-informative priors and treated all of the terms as fixed. We used three independent Markov chains, each with 25,000 iterations after discarding the first 5,000 iterations as burn-in, and retained every fourth iteration for a total of 18,750 iterations per model. We fit models by calling JAGS version 4.1.0 (http://mcmc-jags.sourceforge.net/) with the package 'rjags' (Plummer 2016) in program R version 3.3.3 (https://www.r-project.org/).

Prior to analysis, we standardized all continuous covariates to have a mean of 0 and standard deviation of 1. We used Pearson's correlation to test for correlations among covariates and considered those with a Pearson's $|r| > 0.7$ as correlated and did not include them in the same model. *Type*, *Duration*, and *Clutter* were correlated with one another, so we did not include them in the same models (Table A-1).

We evaluated support of a null model and single-term models for each of the nine covariates (*Type, Duration, Clutter, Issue, Date, Temp, RH, Wind, Rain*). We expected survey method and factors negatively affecting acoustic detectors would strongly affect probabilities of detection, so we also tested a model with the best performing survey method covariate with *Issue* and *Rain*. We tested a global model composed of the best performing survey method covariate and the six other covariates. Finally, we tested all combinations of covariates from the three best performing single-term models.

We monitored model convergence using the potential scale reduction factor (i.e., the Brooks-Gelman-Rubin diagnostic) and assumed convergence when the R-hat of each

parameter was < 1.1. To rank models, we calculated the Widely Applicable Information Criterion (WAIC) for each model using the package 'loo' version 1.1.0 (Vehtari et al. 2016). For each species, we calculated ΔWAIC from the top ranked model and each model's relative likelihood and weight. We calculated 95% credible intervals for covariate estimates and considered their effects significant if the intervals did not include zero. For the top ranked model for each species, we evaluated model performance with k-fold cross-validation. We created five random partitions of the data, with 66% of each partition as a training dataset and the remainder as a testing dataset. We reviewed each training partition to be sure at least one cell from each of the five ecoregions was in each dataset, and used the same partitions to evaluate models for each species. For each model, we used the package 'ROCR' version 1.0.7 (Sing et al. 2005) to calculate area under the receiver-operating curve (AUC). AUC values range from 0 to 1, with 0.5 indicating no predictive power (i.e., random) and 1.0 indicating perfect predictive performance (Cumming 2000).

RESULTS

Cell Selection

When selecting cells from the NABat master sample, we found that six of the top 30 priority cells were primarily in neighboring states. Two cells primarily in North Carolina were surveyed by researchers in that state, so we replaced those cells with the next two cells in the master sample. NABat surveys were not conducted in Georgia during 2015 and 2016, so we surveyed all top priority cells overlapping this border. Three top priority cells were primarily in the Atlantic Ocean. Since they contained very little

land and we were unable to secure permission to conduct stationary surveys, we did not sample them in 2015. In 2016, we were able to secure permission to conduct stationary surveys within two of these cells. In total, we surveyed 35 cells in 2015 and 38 cells in 2016 (Figure 1.1).

Mobile Transects

In 2015, we surveyed 29 cells with mobile transects; 15 cells with mobile transects only, and 14 cells with both mobile transects and stationary points. In 2016, we surveyed 30 cells with mobile transects; 13 cells with mobile transects only, and 17 with both methods (Figure 1.1). We were unable to develop routes in nine of the top priority NABat cells. Issues encountered when we developed transects included cells which did not contain enough suitable roads, gates and stoplights restricting use of roads which were otherwise suitable, and road segments which were not connected within the cells which would require too much time spent driving outside the cells (e.g., in coastal cells where waterways limited road intersections). One transect had to be modified in 2016 due to a road closure on a section of the route. Transect length ranged from 25.5 – 49.5 km with a mean of 33.5 km. Mobile surveys were conducted on 65 occasions each season and ranged in duration from 1 – 99 minutes, with a mean of 62.4 minutes, not including time paused for weather or other issues.

Stationary Points

We completed stationary point surveys in 20 cells in 2015, six of which were surveyed with stationary points only, and 25 cells in 2016, eight of which were surveyed with stationary points only (Figure 1.1). In 2015, we surveyed eight cells with one

stationary point, nine cells with two stationary points, and three cells with three stationary points. In 2016, we surveyed 10 cells with one stationary point, 11 cells with two stationary points, and four cells with three stationary points. We were able to establish stationary point surveys in all cells which were unsuitable for mobile transect surveys, with the exception of one cell that was primarily in the Atlantic Ocean and contained very little accessible land. All three stationary point survey locations within one cell were moved to new locations in 2016 due to concerns with long term access. Stationary point surveys were conducted on 147 occasions in 2015 and 200 occasions in 2016 and ranged in duration from 601 to 640 minutes per night, with a mean of 615.7 minutes.

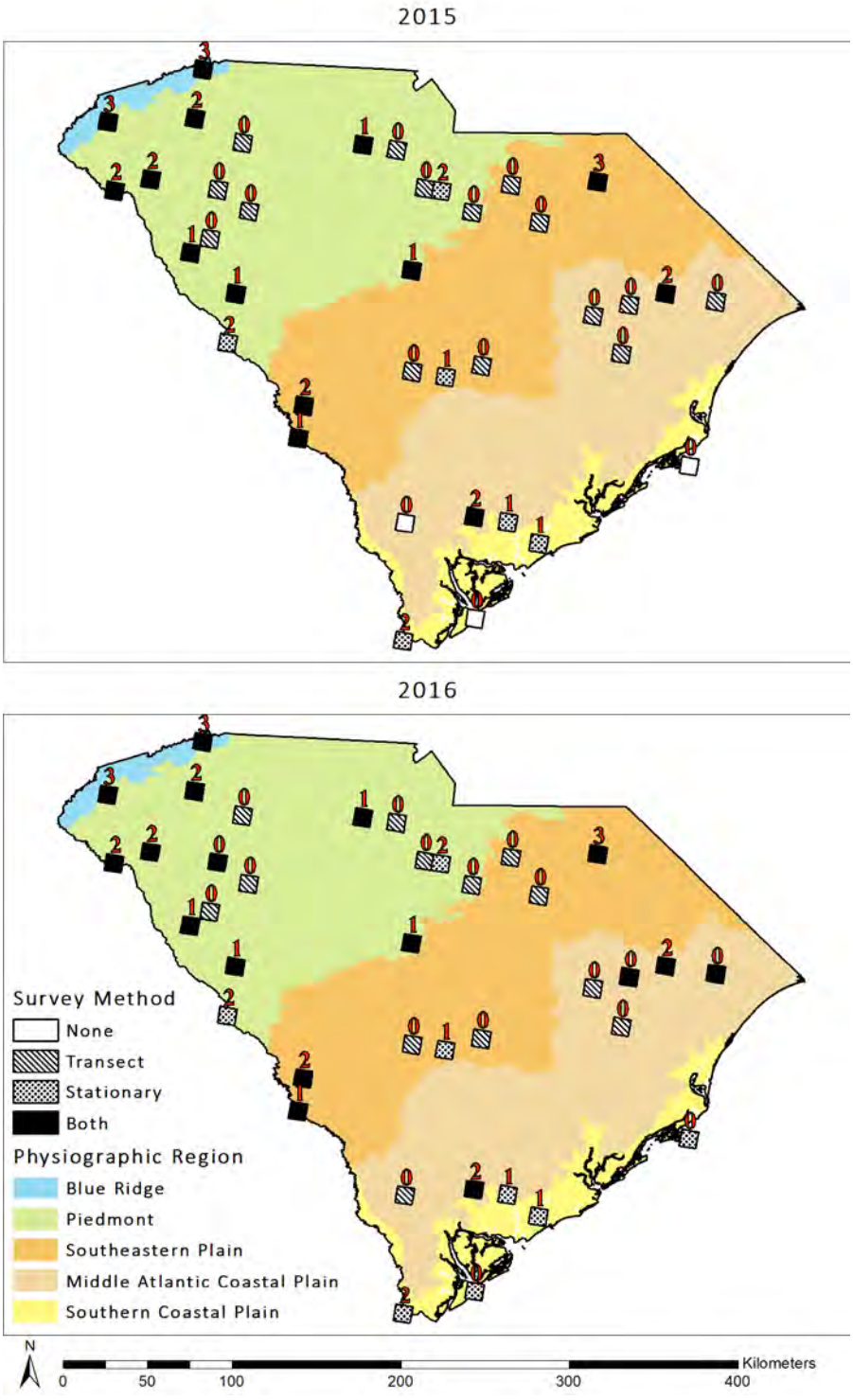


Figure 1.1: Survey methods and number of stationary points within each cell in 2015 (top) and 2016 (bottom) and cell distributions throughout the physiographic regions of South Carolina (U.S. Environmental Protection Agency 2011).

Species Distributions

We recorded 61,397 and 65,727 call files in 2015 and 2016, respectively; 21,972 call files from 2015 and 42,960 call files from 2016 passed our custom noise filter. After manually removing remaining noise files and poor quality and non-search phase calls, 15,292 identifiable bat call files from 2015 remained. We manually classified 27,380 of the 2016 call files to species and labeled the rest as unknown species or as containing no bat calls. Because some species have very similar call characteristics and cannot always be discriminated, we grouped calls of EPFU and silver-haired bat (*Lasionycteris noctivagans*; LANO) as EPFULANO, and eastern red bat (*Lasiurus borealis*; LABO) and Seminole bat (*L. seminolus*; LASE) as LABOLASE. Because we had very few MYLE, MYLU, and MYSE detections (Table A-2) and it is sometimes difficult to discriminate among their calls, we combined their detection histories into one group (MYLELUSE) for more robust modeling of these species. We also included unknown *Myotis* calls from the Blue Ridge and Piedmont regions in this group. We did not include unknown *Myotis* calls from the other regions because those may have been calls of MYAUS, which has different habitat associations than the other three species.

Species distributions based on our detections varied in how well they matched 2003 known distributions and predicted distributions, and differed by year (Table A-2; Figure 1.2; Figure 1.3). In 2015, we detected EPFULANO, LABOLASE, hoary bat (*Lasiurus cinereus*; LACI), MYLU, NYHU, PESU, and Mexican free-tailed bat (*Tadarida brasiliensis*; TABR) in ≥ 50% of the cells throughout their 2003 known

ranges, while we detected CORA, northern yellow bat (*Dasypterus intermedius*; DAIN), MYAUS, MYLE, and MYSE in < 50% of the cells within their 2003 known ranges (Table A-2, Figure 1.2). In 2016, we detected DAIN, EPFULANO, LABOLASE, LACI, MYLU, NYHU, PESU, and TABR in ≥ 50% of cells within their 2003 known ranges, while we detected CORA, MYAUS, MYLE, and MYSE in < 50% of the cells within their 2003 known ranges (Table A-2, Figure 1.2). We detected the LABOLASE group in every cell within its 2003 known range (i.e., every cell we surveyed) in both years, LACI in every cell within its 2003 known range in 2015, and MYLU and TABR in every cell within their 2003 known ranges in 2016. CORA was the only species known to occur in the state that we never detected during our surveys. We were able to generate predicted range maps for some species and not others in the occupancy modeling step (see Chapter 2), and all species were detected in higher percentages of their predicted range than their 2003 known ranges, except PESU in 2016 and LACI in both years (Table A-2; Figure 1.3). All species except LACI were detected in ≥ 50% of the cells within their predicted distributions (Table A-2). We also detected species outside of their 2003 known ranges. We detected LACI both years in a cell 28 km outside its known range, and in nine other cells, one year each, up to 353 km outside its known range (Table A-2; Figure 1.2). We detected MYAUS in 2015 in a cell 17 km outside its known range, and in 2016 in a cell 72 km outside its known range (Table A-2; Figure 1.2). In 2015, we detected MYLU in a cell 128 km outside its known range (Table A-2; Figure 1.2). We detected MYSE in 2015 in a cell 114 km outside its known range, and in 2016 in a cell 305 km outside its known range (Table A-2; Figure 1.2).

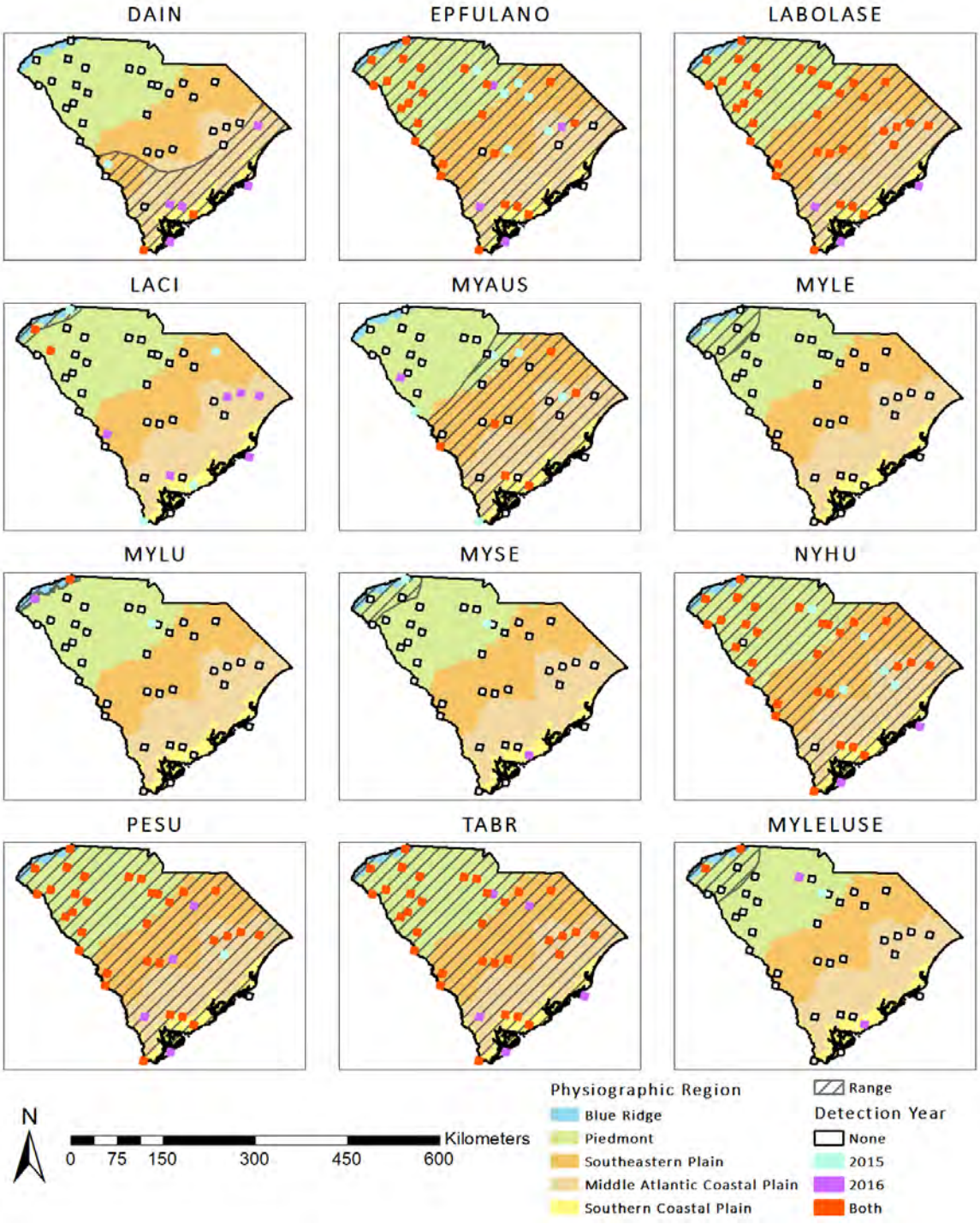


Figure 1.2: Known summer ranges within South Carolina for all species and groupings (Menzel et al. 2003), and their detection/non-detection histories during our acoustic surveys. Refer to Table 1.1 for species code definitions.

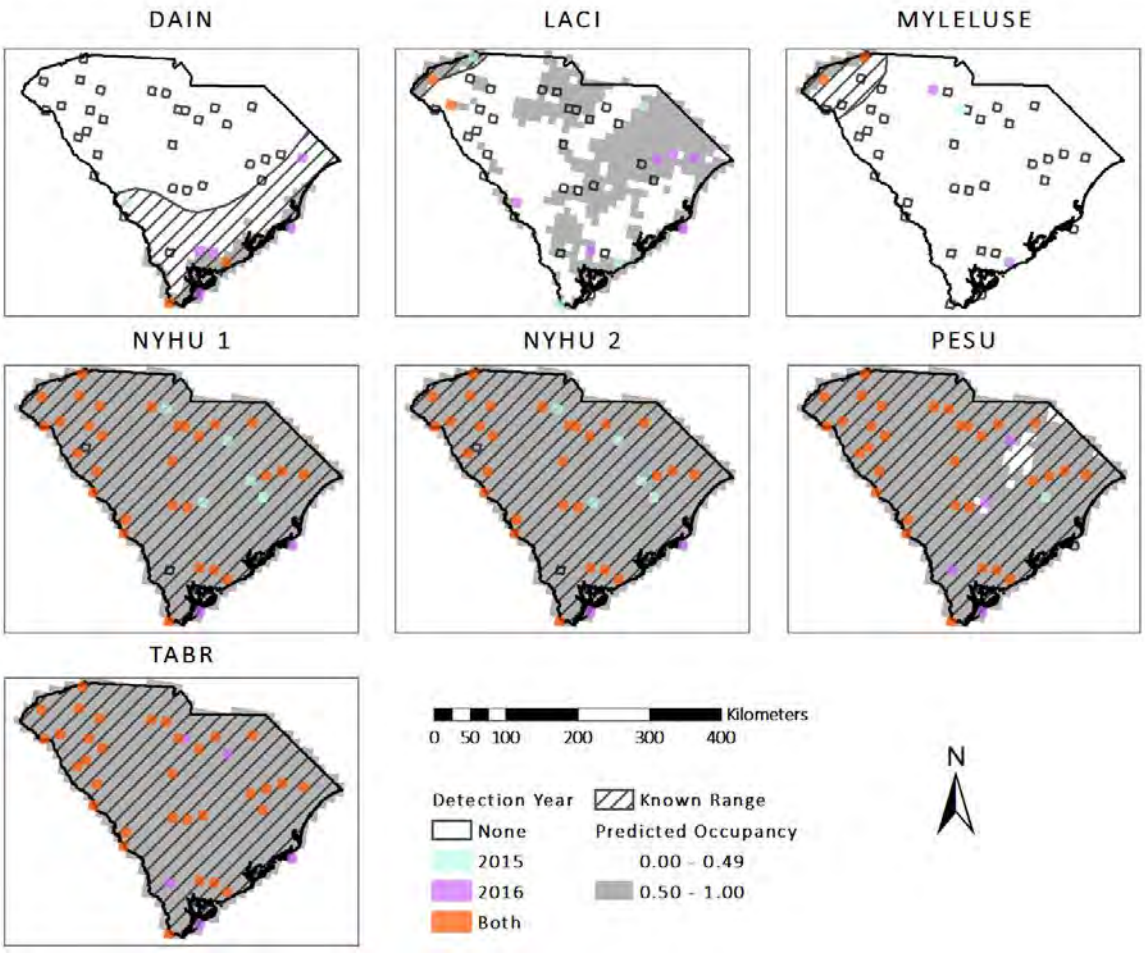


Figure 1.3: Predicted distribution maps and detection histories for species with non-null top ranked occupancy models. Gray shaded areas represent 10 x 10 km NABat cells where models predicted ≥ 50% probability of occupancy. See Table 1.1 for species code definitions and Table A-6 for covariate effects used to generate distribution maps.

Detection Probabilities

The top ranked detection model differed substantially among species, but predictive performance was high for most species (Table 1.2). Only one top model, *Clutter+Issue*, was shared by multiple species (DAIN, LABOLASE, and MYAUS; Table 1.2). Some covariates were retained in the top ranked model for multiple species such as *Issue* (six species), *Clutter* (five species), and *Duration* (three species). We did not

observe support for *Type* in top ranked models for any species; however, for all species except NYHU we observed support for either *Clutter* or *Duration* (Table 1.2), which were highly correlated with *Type* (Table A-1). Top ranked models for all species had predictive performance above 0.70 except PESU (AUC = 0.68; Table 1.2).

Table 1.2: Detection probability models which performed better than the null model, ordered from highest to lowest performance based on WAIC. A "." indicates the null model (i.e., intercept only) and "+" indicates additive effects. Model weights based on WAIC scores are shown. Refer to Table 1.1 for species code definitions.

Species	Model	WAIC	ΔWAIC	Rel. Like.	Weight	AUC
DAIN	Clutter+Issue	**124.6**	0.0	1.00	0.33	0.99
	Clutter+Issue+Temp	125.3	0.7	0.70	0.23	
	Clutter+Issue+Rain	125.8	1.2	0.55	0.18	
	Issue	126.1	1.5	0.47	0.15	
	Issue+Temp	127.9	3.3	0.19	0.06	
	Clutter+Temp	129.2	4.6	0.10	0.03	
	Clutter	130.9	6.3	0.04	0.01	
	Clutter+Issue+Date+Temp+RH +Wind+Rain	136.6	12.0	0.00	0.00	
	Temp	137.4	12.8	0.00	0.00	
	Type	137.8	13.2	0.00	0.00	
	.	138.3	13.7	0.00	0.00	
EPFULANO	Clutter+Issue+Date+Temp+RH +Wind+Rain	**440.0**	0.0	1.00	1.00	0.88
	Date+Temp+RH	461.6	21.6	0.00	0.00	
	Date+Temp	461.8	21.8	0.00	0.00	
	Date	464.8	24.8	0.00	0.00	
	Date+RH	467.0	27.0	0.00	0.00	
	Temp	487.1	47.1	0.00	0.00	
	Temp+RH	488.3	48.3	0.00	0.00	
	Temp+RH+Wind+Rain	490.4	50.4	0.00	0.00	
	RH	514.1	74.1	0.00	0.00	
	.	521.6	81.6	0.00	0.00	
LABOLASE	Clutter+Issue	**127.4**	0.0	1.00	0.45	0.70
	Clutter+Issue+Wind	127.7	0.3	0.86	0.38	
	Clutter+Issue+Rain	129.7	2.3	0.32	0.14	
	Clutter+Issue+Date+Temp+RH +Wind+Rain	133.5	6.1	0.05	0.02	

	Clutter	138.1	10.7	0.00	0.00	
	Clutter+Wind	138.3	10.9	0.00	0.00	
	Issue+Wind	140.6	13.2	0.00	0.00	
	Issue	141.1	13.7	0.00	0.00	
	Type	144.4	17.0	0.00	0.00	
	Duration	147.3	19.9	0.00	0.00	
	Wind	148.0	20.6	0.00	0.00	
	.	148.7	21.3	0.00	0.00	
LACI	Duration+Temp	**172.1**	0.0	1.00	0.38	0.93
	Duration	173.3	1.2	0.55	0.21	
	Type	174.2	2.1	0.35	0.13	
	Duration+Temp+RH	175.0	2.9	0.23	0.09	
	Duration+RH	175.1	3.0	0.22	0.08	
	Clutter	175.9	3.8	0.15	0.06	
	Duration+Issue+Rain	177.1	5.0	0.08	0.03	
	Duration+Issue+Date+Temp+RH +Wind+Rain	180.2	8.1	0.02	0.01	
	Temp	188.8	16.7	0.00	0.00	
	RH	191.1	19.0	0.00	0.00	
	.	191.4	19.3	0.00	0.00	
MYAUS	Duration+Issue	**214.7**	0.0	1.00	0.45	0.90
	Duration+Issue+Temp	215.7	1.0	0.61	0.27	
	Duration+Issue+Rain	216.9	2.2	0.33	0.15	
	Issue+Temp	220.1	5.4	0.07	0.03	
	Issue	220.2	5.5	0.06	0.03	
	Duration+Temp	221.1	6.4	0.04	0.02	
	Duration	221.6	6.9	0.03	0.01	
	Type	222.0	7.3	0.03	0.01	
	Temp	223.6	8.9	0.01	0.01	
	RH	225.0	10.3	0.01	0.00	
	.	225.4	10.7	0.00	0.00	
MYLELUSE	Clutter+Issue	**83.2**	0.0	1.00	0.66	0.97
	Clutter+Issue+Rain	85.0	1.8	0.41	0.27	
	Clutter	88.7	5.5	0.06	0.04	
	Clutter+Rain	90.6	7.4	0.02	0.02	
	Clutter+Issue+Date+Temp+RH +Wind+Rain	92.0	8.8	0.01	0.01	
	Issue	94.1	10.9	0.00	0.00	
	Issue+Rain	95.0	11.8	0.00	0.00	
	.	99.4	16.2	0.00	0.00	
NYHU	Issue+Date+Wind	**457.8**	0.0	1.00	0.44	0.73

	Issue+Date	458.7	0.9	0.64	0.28	
	Issue+Wind	459.7	1.9	0.39	0.17	
	Issue	462.3	4.5	0.11	0.05	
	Date+Wind	463.9	6.1	0.05	0.02	
	Type+Issue+Date+Temp+RH +Wind+Rain	464.8	7.0	0.03	0.01	
	Date	464.9	7.1	0.03	0.01	
	Wind	465.9	8.1	0.02	0.01	
	Type+Issue+Rain	468.5	10.7	0.00	0.00	
	.	468.8	11.0	0.00	0.00	
PESU	Duration+Date	**472.4**	0.0	1.00	0.51	0.68
	Duration+Date+Rain	473.5	1.1	0.58	0.29	
	Duration+Issue+Date+Temp+RH +Wind+Rain	477.4	5.0	0.08	0.04	
	Duration	477.5	5.1	0.08	0.04	
	Clutter	477.7	5.3	0.07	0.04	
	Type	478.6	6.2	0.05	0.02	
	Duration+Rain	479.1	6.7	0.04	0.02	
	Duration+Issue+Rain	479.1	6.7	0.04	0.02	
	Date+Rain	484.9	12.5	0.00	0.00	
	Date	485.3	12.9	0.00	0.00	
	.	487.0	14.6	0.00	0.00	
TABR	Clutter+RH+Issue	**392.4**	0.0	1.00	0.61	0.88
	Clutter+Issue	394.8	2.4	0.30	0.18	
	Clutter+Issue+Rain	395.8	3.4	0.18	0.11	
	Clutter+Issue+Date+Temp+RH +Wind+Rain	396.2	3.8	0.15	0.09	
	Clutter+RH	401.5	9.1	0.01	0.01	
	Clutter	403.7	11.3	0.00	0.00	
	RH+Issue	410.4	18.0	0.00	0.00	
	RH	412.8	20.4	0.00	0.00	
	Issue	415.0	22.6	0.00	0.00	
	Temp+RH+Wind+Rain	417.4	25.0	0.00	0.00	
	.	417.3	24.9	0.00	0.00	

We observed support for *Duration* in the top ranked models for three species (LACI, MYAUS, and PESU; Table 1.2). As we predicted, the effects of *Duration* on detection probabilities were positive (Table 1.3). Over the range of survey duration (1 to 640 minutes), detection probability increased from 0.19% to 23% for LACI, which were never detected on mobile surveys, 7.9% to 44% for MYAUS, and 60% to 85% for PESU (Figure 1.4). Additionally, as we predicted, these three species had higher probabilities of detection at stationary points than on mobile transect surveys.

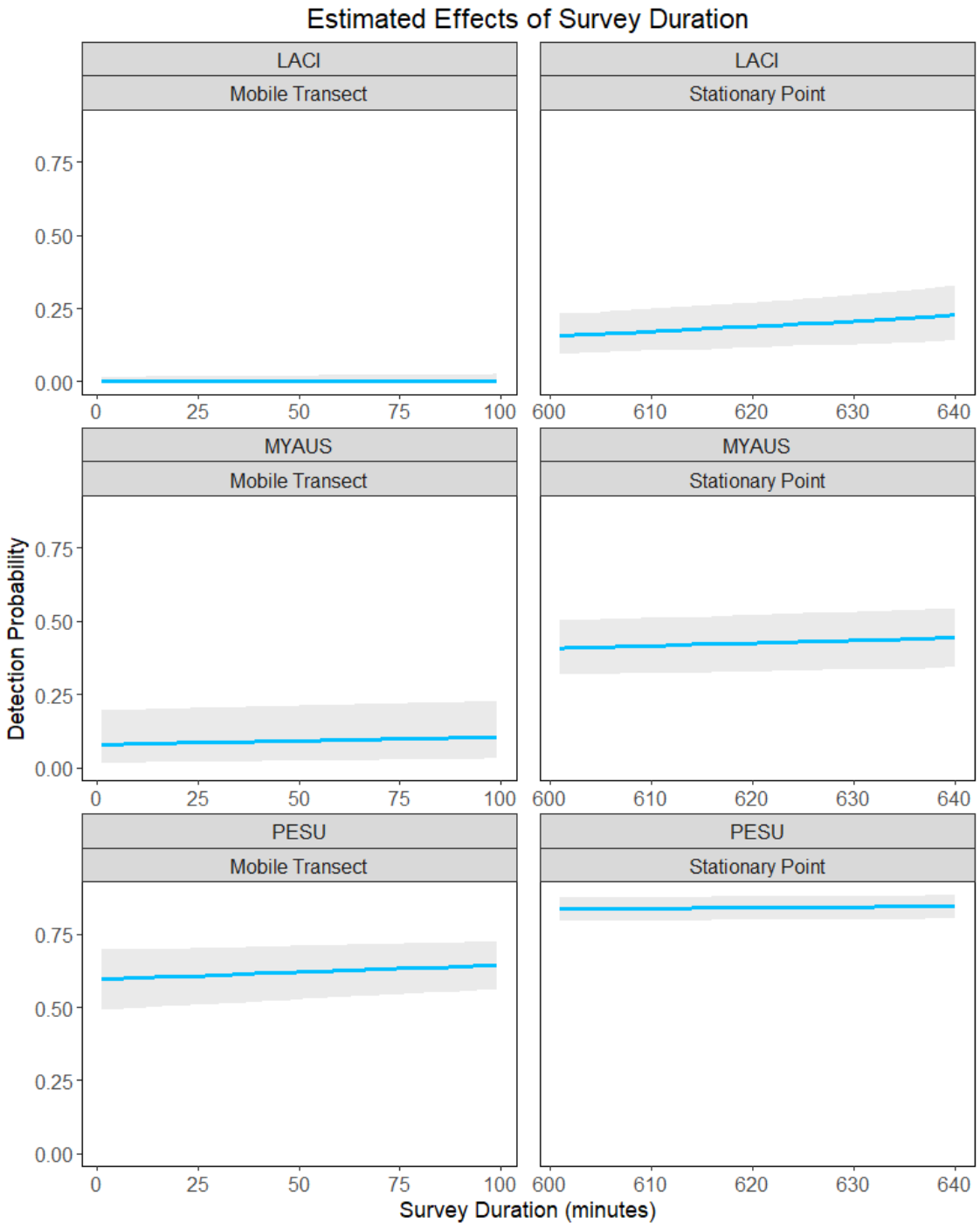


Figure 1.4: Estimated effects of survey duration (minutes) on the probability of detection for species with *Duration* retained in their top ranked model. Duration ranged from 1 to 99 minutes on mobile transect surveys and 601 to 640 minutes on stationary point surveys. Gray shading indicates the 95% credible interval. Refer to Table 1.1 for species code definitions.

We observed support for an effect of *Clutter* in top ranked models for five (DAIN, EPFULANO, LABOLASE, MYLELUSE, and TABR) of the nine species (Table 1.2). As we predicted, DAIN, EPFULANO, LABOLASE, and TABR detection probabilities declined with increasing clutter (Figure 1.5; Table 1.3). Additionally, detection probabilities were significantly higher in at least one stationary point clutter class than in mobile transects for DAIN, EPFULANO, and MYLELUSE (Figure 1.5). Therefore, these results also supported our prediction that stationary points would yield higher probabilities of detection than mobile transects for these species. Contrary to what we predicted, detection probability was significantly greater at high clutter points than along mobile transects or low clutter points for the MYLELUSE group, and did not differ between mobile transects and stationary points for TABR (Figure 1.5; Table 1.4).

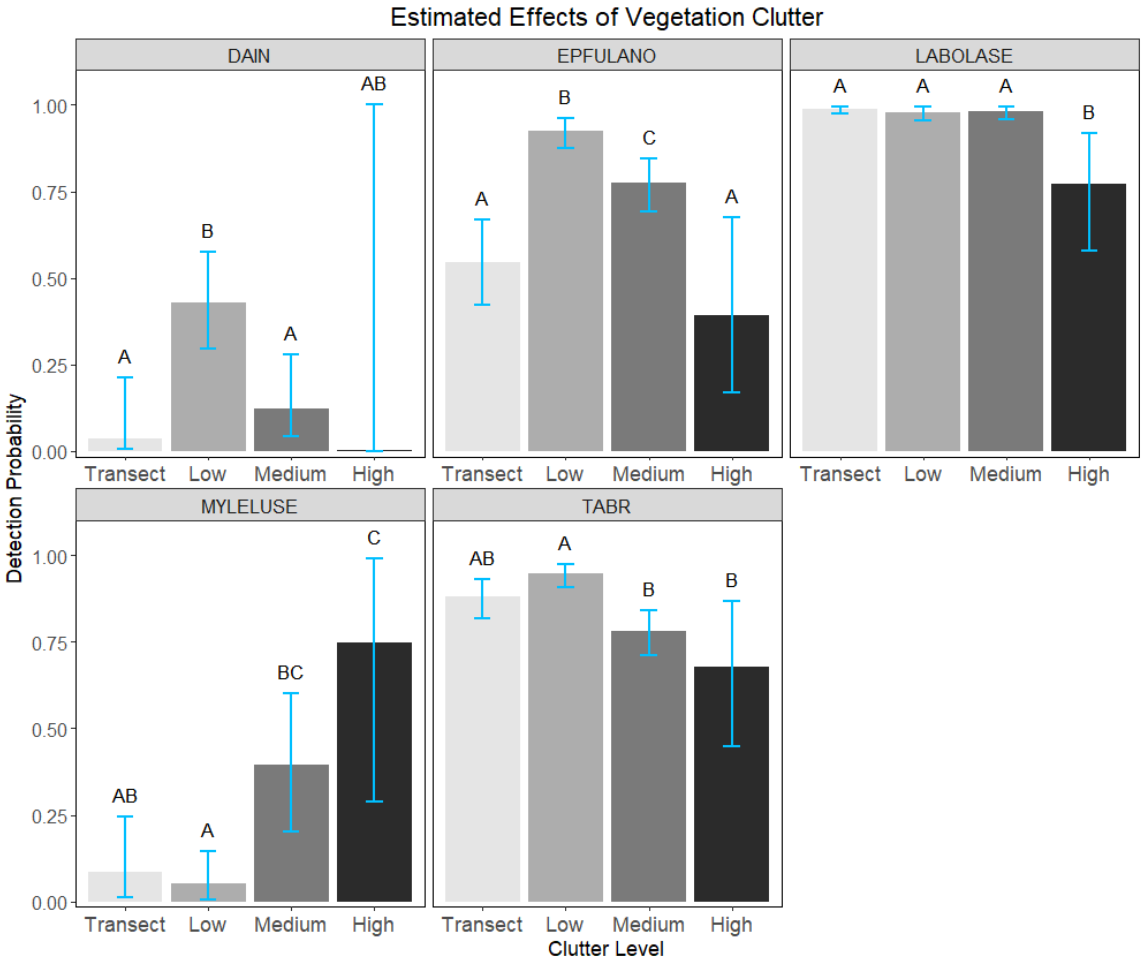


Figure 1.5: Mean estimated detection probabilities at each vegetation clutter level for species with *Clutter* retained in their top ranked models. Transect is the reference value, and Low, Medium, and High are categorical levels of clutter at stationary points. Blue bars indicate 95% credible intervals. Within species, clutter levels which share a letter above their intervals are not significantly different from one another. Refer to Table 1.1 for species code definitions.

We found support for an effect of *Date* for three species: EPFULANO, NYHU, and PESU (Table 1.2). *Date* had a significant positive effect on detection probabilities of EPFULANO and PESU, contrary to what we predicted, and a negative but non-significant effect on detection probability of NYHU (Table 1.3; Table A-3). Detection probability from the first day (Julian day 133) to the final day (Julian day 198) increased

from 18% to 89% for EPFULANO and 70% to 87% for PESU. As we predicted, detection probabilities of LABOLASE, NYHU, and TABR were not significantly affected by *Date* (Table 1.3; Table A-3).

For seven (DAIN, EPFULANO, LABOLASE, MYAUS, MYLELUSE, NYHU, and TABR) of the nine species, we observed support for an effect of *Issue* (Table 1.2). Detection probabilities significantly declined with the occurrence of *Issue* for all species except DAIN, where the effect was significantly positive, and EPFULANO, where the effect was negative but non-significant (Table 1.3; Table A-3).

Although we predicted detection probabilities of all species would be affected by weather covariates, we only found significant effects in three cases (Table 1.3). We predicted positive effects of increasing temperature, but we only found support for this hypothesis for EPFULANO, and found significant negative effects on detection probability of LACI (Table 1.3). Over the range of temperatures (12 °C to 32 °C), detection probability of EPFULANO increased from 13% to 86% and detection probability of LACI decreased from 16% to 1.76%. We observed positive effects of *Humidity* on detection probabilities of EPFULANO and TABR, but it was only statistically significant for the latter (Table 1.3; Table A-3), where detection probability increased from 78% to 92% over the range of *Humidity* (43.5% to 100%). We hypothesized a negative effect of wind speed on detection probability, but *Wind* was only retained in top ranked models for EPFULANO and NYHU, where the effects were negative but non-significant (Table 1.3; Table A-3). We also predicted a negative effect of the occurrence of rain on detection probability, but *Rain* was only retained in the top

ranked model for EPFULANO, where the effect was negative but non-significant (Table 1.3; Table A-3).

Table 1.3: Estimated effects of coefficients from top ranked detection models for each species. "+" indicates a positive effect, "-" indicates a negative effect, and "0" indicates a coefficient not retained in a top ranked model. "*" indicates the effect was statistically significant. We never detected CORA and could not run models for this species. We rarely detected MYLE, MYLU, and MYSE, therefore we combined them into one group: MYLELUSE. Refer to Table 1.1 for species code definitions.

Species	Type	Duration	Clutter	Date	Issue	Temp	RH	Wind	Rain
DAIN	0	0	-*	0	+*	0	0	0	0
EPFULANO	0	0	-*	+*	-	+*	+	-	-
LABOLASE	0	0	-*	0	-*	0	0	0	0
LACI	0	+*	0	0	0	-*	0	0	0
MYAUS	0	+*	0	0	-*	0	0	0	0
MYLELUSE	0	0	+*	0	-*	0	0	0	0
NYHU	0	0	0	-	-*	0	0	-	0
PESU	0	+*	0	+*	0	0	0	0	0
TABR	0	0	-*	0	-*	0	+*	0	0

Based on the top ranked detection models, we found great variability in the average detection probabilities among species. Mean estimated detection probabilities ranged from 0.04 to 0.98 (Table 1.4). All detection models we tested converged well, with no R-hat values exceeding 1.1.

Table 1.4: Mean estimated detection probabilities (Mean *p*) and 95% credible intervals (Lower CI and Upper CI) based on the top ranked detection model for each species and species group. Refer to Table 1.1 for species code definitions.

Species	Mean *p*	Lower CI	Upper CI
DAIN	0.16	0.01	0.97
EPFULANO	0.77	0.17	0.99
LABOLASE	0.98	0.76	0.99
LACI	0.04	8E-6	0.39
MYAUS	0.26	0.03	0.51
MYLELUSE	0.10	8E-4	0.77
NYHU	0.81	0.56	0.91
PESU	0.81	0.66	0.91
TABR	0.86	0.52	0.97

DISCUSSION

With strong coordination and participation of volunteers and personnel from state and federal agencies, we implemented NABat acoustic surveys throughout South Carolina, which provided valuable, large scale information about species distributions and detection probabilities in the state. We found that species detections appear to more closely match predicted distributions from our surveys than they match known range maps from 2003. We also found that it is important to control for variation in detection probabilities among species and survey occasions.

With one lead coordinator, we were able to follow the NABat guidelines to establish our goal of 30 mobile transects and at least one stationary point survey within 25 cells. Public land managers and private landowners we contacted were willing to grant permission to conduct stationary surveys on their property. However, primarily due to a lack of public lands within many cells, 40% of the cells that we surveyed each year had only one stationary point, less than recommended by the NABat plan. We established

three stationary point surveys in each cell in northwestern South Carolina, where public land is prevalent, and if more time can be dedicated to identifying and contacting private landowners, it would likely be possible to establish two to four points within each cell where public land is not prevalent.

We determined NABat acoustic surveys were effective at monitoring all species except CORA, MYAUS, MYLE, and MYSE, and that most species detections more closely matched predicted distributions than their 2003 known ranges, which may indicate that distributions have changed since 2003 or that we more thoroughly surveyed throughout the state than historical efforts. Species range maps may therefore need to be updated. However, there were still detections outside predicted ranges for DAIN, LACI, MYLELUSE, and PESU and with continued surveying, and perhaps surveying more cells in these extra-range detection areas, species' distributions may be more accurately mapped. The four species we detected outside their 2003 known ranges, including three *Myotis* species, are all considered species of greatest conservation need within the state by the SCDNR (South Carolina Department of Natural Resources 2015). However, even though we were confident with our identification of acoustic calls, misclassifications are a possibility and extra-range presence should be verified by physical identification through methods such as mist netting. For example, although not prompted by our results, mist netting efforts in coastal areas of South Carolina resulted in the capture of MYSE in two locations in 2016 and 2017 (White et al. 2017, in review; http://www.dnr.sc.gov/news/2017/july/jul7_longearbats.html), approximately 93 km northeast and 72 km southwest from our detection. These new capture records reinforce

our acoustic detection and demonstrate the potential for NABat acoustic monitoring to improve the effectiveness of mist net surveys by providing suggested locations to target. Compared to species with statewide distributions, we tended to detect species with more limited distributions, especially the *Myotis* species, in a lower percentages of cells within their 2003 known ranges. We tended to select sites for stationary point surveys along forest edges and in less cluttered areas to decrease distortion of bat echolocation calls and increase detection range, but because *Myotis* species are clutter-adapted species (Patriquin and Barclay 2003, Starbuck 2013), this may have decreased the probability of detecting them. Additionally, all *Myotis* species 2003 known ranges in South Carolina except that of MYAUS overlap the area impacted by WNS. WNS has contributed to declines of MYLU and MYSE in South Carolina (Loeb et al. 2016), and may have led to low acoustic detections.

We observed great variability in the top ranked detection model among species, but predictive performance of most models was very high, suggesting that it is important to control for detection, and that multiple factors should be considered when accounting for detection probability in NABat surveys. *Issue* was the most commonly retained covariate in top ranked models among species, likely because it negatively impacts the acoustic detector itself and should reduce detection probability of all species. This result emphasizes the importance of fully completing mobile transect surveys and taking measures to ensure stationary point detectors do not fall over (e.g., anchoring, staking, or attaching guy-lines to tripods or poles). However, the significant positive association of DAIN detection probability with the occurrence of a survey issue seems counterintuitive.

We believe this result could be due to a majority of DAIN detections in 2015 occurring at two stationary points where the detectors had fallen over but were still functioning and recording bat calls. Additionally, it was not possible to confidently determine the date and time these detectors fell over, so detections may have occurred while they were still upright.

Clutter was another commonly supported predictive covariate, but the effects varied by species and appeared to be related to the clutter adaption of each species. DAIN, EPFULANO, and TABR are considered open-adapted species (Menzel et al. 2005, Loeb and O'Keefe 2006) and we accordingly found negative effects of increasing vegetation clutter for these species. *Myotis* species are considered clutter-adapted species (Patriquin and Barclay 2003, Starbuck 2013), and accordingly, we found the probability of detecting MYLELUSE significantly increased with increasing vegetation clutter and was very high at high clutter stationary points. Additionally, we found low probabilities of detection of MYLELUSE at low and medium clutter stationary points, which did not significantly differ from mobile transects. LABOLASE are considered open- or semi-clutter-adapted species (Menzel et al. 2005, Loeb and O'Keefe 2006, Starbuck et al. 2015), and we found detection probabilities did not significantly differ among mobile transect and stationary point surveys, except at high clutter points where detection probability was significantly lower. The effect of high vegetation clutter may be due to the ability of LABOLASE to modify their calls in cluttered environments, making the calls too difficult to classify or appear to be calls of PESU or *Myotis* species. Our results suggest researchers conducting NABat stationary point surveys should consider selecting

locations from a range of vegetation clutter amounts, not just open areas, especially to increase the probability of detecting *Myotis* and other clutter adapted species.

Duration was retained in top ranked models for LACI, MYAUS, and PESU. It is possible that longer duration surveys increased the chance of bats encountering our detectors during a survey occasion. We never detected LACI on mobile surveys, unlike another study (Whitby et al. 2014), which may be due to the migratory behavior of LACI (Cryan 2003). The majority of LACI individuals may have been moving north at the beginning of our survey season. We may have detected transient individuals at stationary points, but not at mobile transects because stationary point surveys were conducted during twice as many nights and had longer durations than mobile transect surveys. We primarily detected LACI in cells early and late in the season, when the length of night was greatest and, thus, the duration of stationary point surveys was greatest, leading to a significant effect of *Duration* on the probability of detecting LACI.

Although survey method (i.e., *Type*) was not retained in the top model for any species, six out of nine bat species had significantly higher probabilities of detection on stationary point than mobile transect surveys, which is consistent with the findings of some studies (Tonos et al. 2014, Whitby et al. 2014). Detection probabilities of LABOLASE at medium and low clutter stationary points, and TABR at all stationary points did not significantly differ from mobile transects (Figure 1.5; Table 1.4), which is similar to the findings of a study dominated by TABR in Texas (Fisher-Phelps et al. 2017). Overall, these results suggest that stationary points may be more effective than mobile transects for detecting some species. However, mobile transects may still be

suitable in cases where it is not possible or feasible to conduct stationary point surveys, even for species with low probabilities of detection on mobile transects, and they may be used to estimate relative abundance of species, whereas stationary points cannot. Additionally, positive effects of increasing survey duration suggest higher probabilities of detection could be achieved with longer mobile transects, but further research is needed (e.g., a comparison of a range of transect lengths within each cell, or multiple passes within one night).

In general, it appears NABat survey guidelines appropriately control for reproductive phenology, seasonal activity patterns, and weather effects in South Carolina. We hypothesized *Date* would be a significant factor for detection of species with limited distributions, but we found only significant positive effects for EPFULANO and PESU, both of which had statewide distributions (Table 1.4). This may be due to increasing levels of activity as the summer progressed, or perhaps higher abundance (MacKenzie 2005) in cells sampled later in the season. PESU are experiencing significant declines in areas we sampled later in the season (S. Loeb, pers. comm. 2017). But, the cells we surveyed later in the season in northwestern South Carolina were dominated by forests, whereas those we sampled earlier in the season were dominated by agriculture and forested wetlands. PESU are positively associated with forest cover (Farrow and Broders 2011) and, thus may be more evenly distributed in cells in the northwestern part of the state, resulting in higher probabilities of detection relative to earlier in the season. LACI detection probability was negatively associated with increasing temperatures, possibly because they are a migratory species (Cryan 2003) and were moving into the mountains

(i.e., the northwestern part of our study area) as temperatures increased. EPFULANO detection probability was positively associated with increasing temperatures, which follows findings of other studies that have found increasing bat activity with higher temperatures (O'Donnell 2000, Broders et al. 2006, Kitzes and Merenlender 2014, Wolbert et al. 2014). Although survey date and weather variables were rarely retained in top ranked detection models, likely due to the seasonal timing and weather restrictions of NABat surveys, these effects should still be considered when determining the timing of surveys, in statistical analyses, and when conducting NABat surveys in other areas.

We found very high mean estimated probabilities of detection for EPFULANO, LABOLASE, NYHU, PESU, and TABR. All of these species are known to occur throughout our study region and none of them are affected by WNS in South Carolina except for PESU, which is declining due to WNS in the northwestern part of the state (Loeb et al. 2016). We found low probabilities of detection for other species, and we never detected CORA throughout our study, but recent mist netting and cavity surveys conducted by other researchers have detected CORA in South Carolina (Lucas et al. 2015, Loeb 2017). The presence of CORA in the state but lack of detection on acoustic surveys is consistent with other research that found CORA are less likely to be detected with acoustic surveys than other methods due to their relatively quiet echolocation calls (Clement and Castleberry 2011; although, see Comer et al. 2014). Even though we combined MYLE, MYLU, and MYSE into one group, the mean estimated detection probability remained very low (0.10). In addition to their echolocation calls being easily mistaken for one another, these *Myotis* species have relatively high frequency, short

duration echolocation pulses, which attenuate more rapidly than lower frequency calls and often resemble feeding buzzes of other species, so some of their calls may be dismissed during classification. Also, MYLU and MYSE populations have declined in South Carolina due to WNS, and mist netting efforts have captured fewer MYLU and MYSE than in the past (Loeb et al. 2016). Thus, lower rates of positive identification in combination with relatively low abundance may be driving low probabilities of detection. Our mean estimated detection probability for MYAUS was also relatively low. MYAUS is another species with echolocation pulses which can be confused with feeding buzzes. They have somewhat specific habitat requirements, preferring low lying forested wetlands and typically roost in large tree cavities (Gooding and Langford 2004, Carver and Ashley 2008, Bender et al. 2015). Thus, due to the random distribution of NABat priority survey cells, suitable habitat can be missed, decreasing the probability of detection. We found the lowest mean estimated probability of detection (0.04) for LACI, which have a small known summer distribution in South Carolina and exhibit migratory behavior (Cryan 2003), so they may only occupy an area for a short time, which would effectively reduce the probability of detection. Additionally, a study of the Hawaiian hoary bat (*Lasiurus cinereus semotus*), found that the bats were often present but not acoustically detected during flight (Gorresen et al. 2017), which may also be true for *L. c. cinereus* (the subspecies found in South Carolina) which would further reduce their probability of detection.

From our results, it appears that NABat acoustic survey methods may be suitable for monitoring most species in South Carolina, but not appropriate for others. Further, we

found that survey method affects the probability of detecting many species, and that mobile transect surveys may be effective for some species, but not suitable for others. In addition to NABat acoustic surveys, it may therefore be necessary to conduct hibernacula or summer roost surveys, mist netting, and possibly active acoustic surveys for monitoring CORA, DAIN, and *Myotis* species, since we had no or very low acoustic detections of these species. We also found that biological and behavioral differences among species can influence whether survey variables affect their probabilities of detection as well as whether the effects are positive or negative. To effectively utilize the results of acoustic surveys when determining management actions, mapping species distributions, and evaluating bat activity and habitat use, researchers should monitor survey variables and determine how they may affect the probability of detecting bat species.

MANAGEMENT RECOMMENDATIONS

We recommend continuing the NABat surveys we established throughout South Carolina to monitor the status of bat populations. We suggest dedicating time toward establishing at least 2 stationary point surveys in each of the top priority cells due to the higher detection probabilities at stationary points than mobile transects for most species. Stationary points should be in a variety of habitats within each cell, even in areas with vegetation clutter, because we found that detection of clutter adapted *Myotis* species was significantly higher in more cluttered areas. Mobile transect surveys should also be conducted, however, because these data may be used to calculate relative abundance of species (Roche et al. 2012, Loeb et al. 2015). We also recommend taking detailed,

accurate measures of survey level variables, particularly whether a survey issue occurred (i.e., detector malfunction or incomplete mobile transect) and the vegetation clutter around stationary points, to account for variation in detection probabilities. We realize biologists may not have the resources to manually classify tens of thousands of echolocation calls to species each year, and it may therefore be necessary to use automated classification software. If automated software is used, we recommend vetting calls and determining false positive rates (i.e., detection when a species is not present) of the software to account for this in analyses. When extra-range acoustic detections occur, we recommend conducting further studies in these areas (e.g., mist-netting) to verify the acoustic detections and to learn more about populations in these areas.

CHAPTER TWO

EFFECTS OF ENVIRONMENTAL FACTORS ON LANDSCAPE SCALE BAT SPECIES OCCUPANCY

Bats, a diverse and widespread order of mammals that provide important ecosystem services, have been experiencing significant regional declines due to the introduction and spread of disease, energy development, and habitat loss (Kunz et al. 2007, Jones et al. 2009, Boyles et al. 2011, Kunz et al. 2011, Hammerson et al. 2017). To understand the impacts of these threats at a population scale, landscape scale monitoring is needed (Jones et al. 2009, Loeb et al. 2015). With monitoring at large temporal and spatial scales, the status and trends of bat populations at the landscape scale can be assessed, aiding the development of effective management practices (Roche et al. 2012, Loeb et al. 2015, Rodhouse et al. 2015).

Land use and land cover change are substantial threats to the sustainability of bat populations (Duchamp and Swihart 2008, Cryan and Barclay 2009). Landscape attributes (e.g., amount of forest cover, fragmentation, edge density) may influence the presence of bat species, due to the importance of roosting and foraging site requirements for habitat selection (Kunz 1982). For instance, the presence and activity of many bat species are positively associated with forest cover (Ford et al. 2005, Broders et al. 2006, Reid 2006), and the loss and alteration of forest cover influences occupancy rates (Yates and Muzika 2006, Farrow and Broders 2011). However, foraging habitat preferences of species are related to their foraging behavior (i.e., gleaning vs. hawking their prey), size, and wing morphology, where some species prefer to forage in open areas and along forest edges

(Patriquin and Barclay 2003, Loeb and O'Keefe 2006, Hein et al. 2009), and others prefer forest interiors or have no strong habitat preference (Starbuck et al. 2015). Roads often create edges on the landscape and, thus, may influence the presence of bat species, but findings are inconsistent because roads can act also as barriers to movement (Loeb and O'Keefe 2006, Hein et al. 2009, Zurcher et al. 2010, Bennett and Zurcher 2013, Kitzes and Merenlender 2014, Bender et al. 2015, Pauli et al. 2015). Across the landscape, habitat associations of each species are related to their roosting preferences and clutter adaptions (Ford et al. 2005, Duchamp and Swihart 2008). Additionally, climate change is expected to cause widespread changes in land cover and habitat, and shrinking habitat for terrestrial vertebrates has been predicted for the southeastern United States, especially in scenarios with expansion of urban and agricultural areas (Martinuzzi et al. 2015), which may therefore impact bat species distributions (Rebelo et al. 2010).

Effective conservation strategies that address the threats to bat populations in the southeastern United States require broad scale monitoring and analyses (Jones et al. 2009, Rodhouse et al. 2012). Thus, our objective was to conduct the first state-wide assessment of factors influencing bat species habitat use in South Carolina and produce statewide maps of predicted ranges. The North American Bat Monitoring Program provides the means to establish large scale monitoring. Data from this program can be used to evaluate support for the hypothesized effects of environmental variables on bat species summer occupancy at large scales (Loeb et al. 2015). With temporally dynamic (i.e., multi-season) analyses, changes in species occupancy rates over time and annual colonization and extinction rates (i.e., turnover) can be estimated, and predicted distribution maps can

be generated, which may reveal changes in species distributions in response to changing habitat (Rodhouse et al. 2015). Species-specific findings could be used to inform landscape management decisions that may affect bat populations. Additionally, these data can be used to guide subsequent, finer-scale investigations in species-specific patterns in habitat use and serve as a baseline for future comparative studies examining changes in bat habitat usage and species distributions over time.

METHODS

Study Location

We conducted our study throughout South Carolina and within 10 km of the state border in Georgia and North Carolina. There are five physiographic regions in South Carolina, which occur in a gradient from northwest to southeast: Blue Ridge, Piedmont, Southeastern Plains, Middle Atlantic Coastal Plain, and Southern Coastal Plain (U.S. Environmental Protection Agency 2011). Land use throughout South Carolina includes developed urban areas, silviculture, agriculture, livestock, and undeveloped land (Homer et al. 2015), but the dominant land cover varies among regions. Forest was the dominant land cover in the Blue Ridge, forest and hay or pasture were the dominant land cover types in the Piedmont, woody wetlands, forest, shrublands, and cultivated crops were the dominant land cover types in the Southeastern Plains and Middle Atlantic Coastal Plain, and herbaceous wetlands, woody wetlands, forest, and open water were the dominant land cover types in the Southern Coastal Plain. Topographic relief and elevation in South Carolina are greatest in the Blue Ridge, with peaks up to 1085 m, and sharply decrease in the central regions, finally becoming low elevation plains and wetlands near the coast.

We included all fourteen bat species that are known to occur within South Carolina in our study. During summer, big brown bat (*Eptesicus fuscus*; EPFU), eastern red bat (*Lasiurus borealis*; LABO), Seminole bat (*L. seminolus*; LASE), evening bat (*Nycticeius humeralis*; NYHU), tri-colored bat (*Perimyotis subflavus*; PESU), and Mexican free-tailed bat (*Tadarida brasiliensis*; TABR) occur throughout South Carolina, while Rafinesque's big-eared bat (*Corynorhinus rafinesquii*; CORA), northern yellow bat (*Dasypterus intermedius*; DAIN), hoary bat (*L. cinereus*; LACI), silver-haired bat (*Lasionycteris noctivagans*; LANO), southeastern myotis (*Myotis austroriparius*; MYAUS), eastern small-footed bat (*M. leibii*; MYLE), little brown bat (*M. lucifugus*; MYLU), and northern long-eared bat (*M. septentrionalis*; MYSE) have more limited distributions within the state (Cryan 2003, Menzel et al. 2003).

Sampling Design

We used the North American Bat Monitoring Program (NABat) framework to acoustically survey bat species across South Carolina (Loeb et al. 2015). The sampling frame for NABat, which was selected through a review of other large-scale monitoring programs, consists of a continuous grid of 10 x 10 km cells across North America. We identified priority survey cells within South Carolina based on the NABat master sample, which utilizes the generalized random tessellation stratified algorithm to assign priority numbers to cells to maintain a spatially balanced and randomly distributed sample. Within each cell, we conducted stationary point surveys for four consecutive nights and mobile transect surveys on two of the four nights. In 2015, we surveyed 35 cells: 15 with mobile transects only, six with stationary point surveys only, and 14 with both survey

methods. In 2016, we surveyed the same 35 cells from 2015 and three additional cells (one with mobile transects only, and two with stationary points only) for a total of 38 cells surveyed: 13 with mobile transects only, eight with stationary point surveys only, and 17 with both survey methods. We surveyed three cells with stationary point and mobile transect surveys in 2016 which were surveyed with mobile transects only in 2015 (Figure 2.1). Each stationary point survey began 30 minutes prior to sunset and ended 30 minutes after sunrise, while each mobile transect survey began 45 minutes after sunset and was driven at 32 kph, with duration dependent on the length of the transect (25 – 48 km). During each year, we surveyed the same stationary point locations and mobile transect routes within each cell where possible.

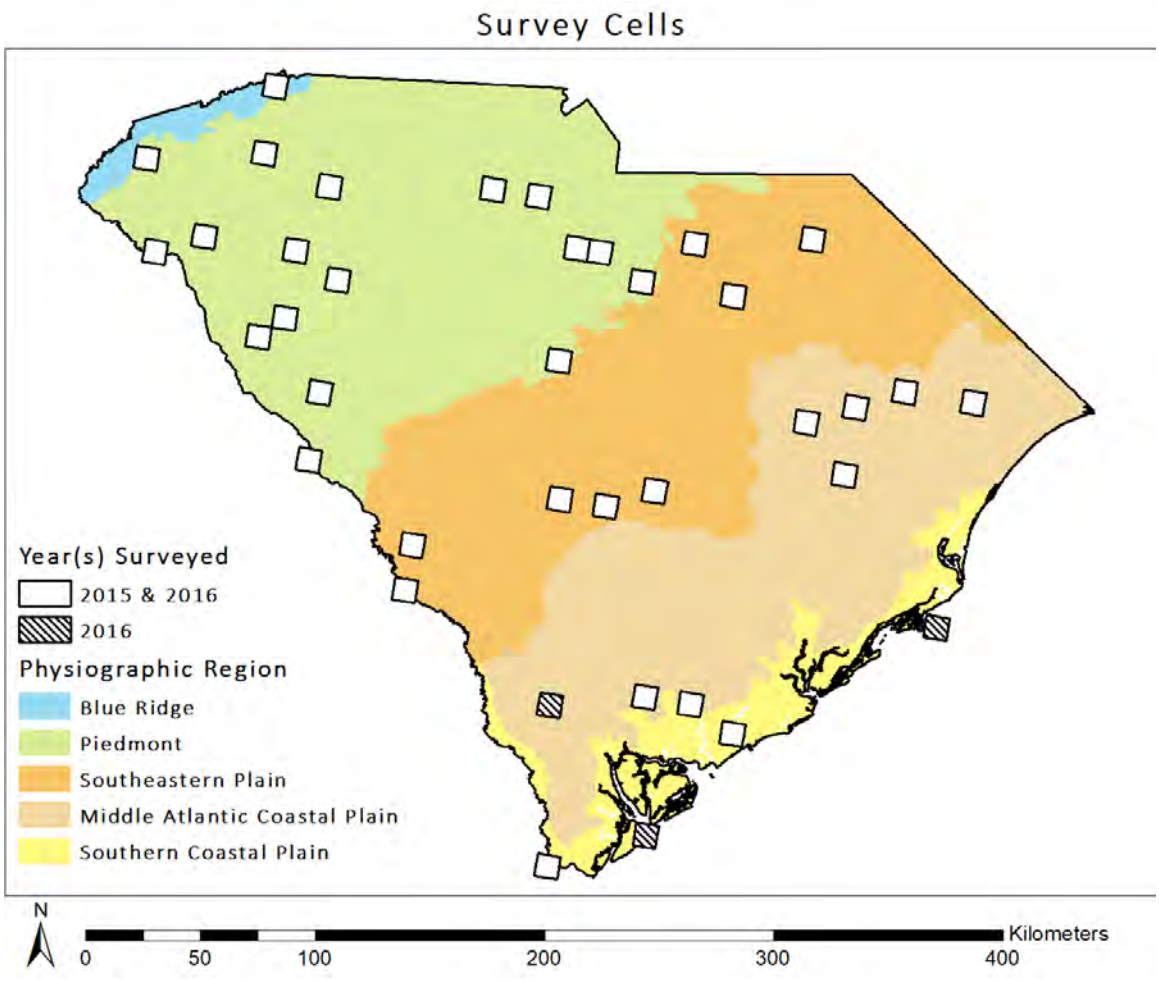


Figure 2.1: Distribution of NABat priority cells across South Carolina which we surveyed, by year. Physiographic regions of South Carolina are displayed (U.S. Environmental Protection Agency 2011).

For both survey methods, we used Anabat SD2 bat detectors with directional, stainless steel microphones (Titley Scientific, Columbia, MO, USA) and 2.5 m microphone cables. For stationary point surveys we mounted the microphone inside a water resistant PVC housing and attached it to the top of a 1.8 m high tripod. For mobile

transect surveys we placed the microphone at the center of a vehicle's roof with no waterproof housing.

Data Processing

To determine which species we detected during surveys, we first removed call files containing no bat calls or calls with fewer than three search-phase pulses using a custom noise filter in AnalookW version 4.2.7 (AnalookW 2016) and through manual review of each file. We classified the remaining call files collected during 2015 to species using EchoClass version 3.1 and Kaleidoscope Pro version 3.1.5, and manually vetted all classifications based on reference calls of each species. We observed low classification agreement between automated classifiers, so we manually classified all high quality search phase calls from 2016. Our reference calls were recorded from captured bats which were identified and light-tagged (Britzke et al. 2011). We aggregated EPFU and LANO calls as EPFULANO and LABO and LASE calls as LABOLASE because these species have very similar echolocation call structures. We also grouped calls of MYLE, MYLU and MYSE as MYLELUSE for analyses because we detected them in very few cells, their echolocation calls can be difficult to distinguish, and their foraging habitat preferences are very similar (Reid 2006, Duchamp and Swihart 2008). We included unknown *Myotis* calls from the Blue Ridge and Piedmont regions in the MYLELUSE group because they were most likely calls of one of those species.

Data Analysis

We hypothesized that probability of occupancy would vary by ecoregion for species with limited ranges within our study area, but that it would not vary by ecoregion

for species with statewide ranges (Table 2.1; Menzel et al. 2003). Thus, we included a categorical covariate (*Region*) based on the primary U.S. Level III Ecoregion (U.S. Environmental Protection Agency 2011) within each cell.

We hypothesized that the effects of land cover types and forest fragmentation on probability of occupancy varied among species, based on species summer roosting and foraging site preferences (Table 2.1). Many species are known to forage along forest edges and riparian areas (CORA, DAIN, LABO, LACI, LANO, MYAUS, MYLU, and PESU; Reid 2006, Hein et al. 2009), and other species are known to forage in openings or over agricultural areas (LABO, NYHU, and TABR; Reid 2006). MYAUS, MYLE, MYLU, and MYSE are associated with relatively contiguous tracts of forest cover and MYLE and MYLU are often found in higher elevation forests (Reid 2006, Duchamp and Swihart 2008). NYHU are typically found in lower elevation forests, DAIN and MYAUS are found in lower elevation forested wetlands, and EPFU are generalists that utilize a variety of habitat types (Reid 2006). For measures of land cover, we calculated percent land coverage within each cell from the National Land Cover Database (NLCD 2011; U.S. Geological Survey 2014) and aggregated "Pasture/Hay" and "Cultivated Crops" as *Ag*, all classes of development as *Dev*, and upland forest types as *Forest*. For bottomland forest associated bat species, we also used "Woody Wetlands" (*F.Wet*). We used our reclassified NLCD 2011 data as input in Fragstats version 4.2.1 (http://www.umass.edu/landeco/research/fragstats/fragstats.html). Within each cell, we calculated *Contagion*, a landscape measure which increases as land cover type

interspersion decreases and dispersion increases, and *F.ED* and *F.Wet.ED*, measures of forest and forested wetland edge density, respectively.

Multiple species in our study prefer foraging near streams and riparian areas (Grindal et al. 1999, Reid 2006), so we hypothesized positive effects of increasing stream length on their probability of occupancy. Additionally, streams can act as forest edges and may be important sources of water, so we also hypothesized positive effects of increasing stream length on probability of occupancy for species that commonly forage along edges, but may not be explicitly associated with riparian areas (Table 2.1). We calculated total stream length within each cell using 'NHDFlowline' data from the National Hydrology Dataset (NHD; U.S. Geological Survey, National Geospatial Program 2017).

Due to effects of roads on species presence (Loeb and O'Keefe 2006, Hein et al. 2009, Zurcher et al. 2010, Bennett and Zurcher 2013, Kitzes and Merenlender 2014, Bender et al. 2015, Pauli et al. 2015), and because different road classes are often associated with different landscapes (Hawbaker et al. 2005), we hypothesized effects of roads on bat species occupancy would vary based on road type (Table 2.1). We used National Transport Dataset (NTD) RoadSegment data (USGS, National Geospatial Technical Operations Center 2014) and U.S. Forest Service Roads (U.S. Geological Survey, National Geospatial Technical Operations Center 2016). We classified roads into four categories, primarily based on Master Address File/Topologically Integrated Geographic Encoding and Referencing Feature Class Code Definitions (https://www.census.gov/geo/reference/mtfcc.html): *Pri* included divided highways with

access ramps, *Sec* included highways with intersections, *Ter* included single lane rural and city roads, and *Qua* included forest access roads. We then calculated the length of all four road classes within each cell.

Table 2.1: Predicted (left of "|") and observed (right of "|") effects of environmental variables on the probability of occupancy for each species. *Region* is the physiographic region, *Ag*, *Dev*, *Forest*, and *F.Wet* are percent coverage of agriculture, urban development, forest, and forested wetland, respectively. *Contagion* is a landscape measure that increases with fewer, more aggregated cover types, and decreases with a greater number and more dispersed cover types. *F.ED* and *F.Wet.ED* are forest and forested wetland edge density. *Stream* is stream length. *Pri, Sec,* and *Qua* are measures of primary, secondary, and quaternary road length, respectively. Predicted and observed effects on probability of occupancy are indicated by "Y" as an effect of a categorical covariate, and "+" as a positive effect, "0" as no effect, or "-" as a negative effect for continuous variables. "NA" indicates an effect we did not test for a species, based on habitat preferences. Effects that were statistically significant are highlighted with black background. Refer to Table 1.1 for species code definitions.

Species	Region	Ag	Dev	Forest	F.Wet	Contagion	F.ED	F.Wet.ED	Stream	Pri	Sec	Qua
DAIN	**Y | Y**	- | 0	- | 0	NA	+ | 0	0 | 0	NA	+ | 0	+ | 0	- | 0	- | 0	0 | 0
EPFULANO	0 | 0	0 | 0	0 | 0	0 | 0	NA	0 | 0	+ | 0	NA	0 | 0	- | 0	0 | 0	+ | 0
LACI	Y | 0	0 | 0	- | 0	+ | 0	NA	0 | 0	**0 | -**	NA	+ | +	- | 0	- | 0	0 | 0
MYAUS	Y | 0	- | 0	- | 0	NA	+ | 0	+ | 0	NA	+ | 0	+ | 0	- | 0	- | 0	0 | 0
MYLELUSE	**Y | Y**	- | 0	- | 0	+ | 0	NA	+ | 0	- | 0	NA	+ | 0	- | 0	- | 0	0 | 0

NYHU	0 | 0	+ | 0	0 | 0	+ | 0	NA		0 | 0	+ | -	NA	0 | +	- | 0	0 | 0	+ | 0
PESU	0 | 0	- | -	- | +	+ | +	NA		0 | 0	+ | 0	NA	+ | 0	- | 0	- | 0	+ | +
TABR	0 | 0	+ | 0	+ | 0	- | 0	NA		0 | 0	0 | 0	NA	0 | 0	- | +	0 | +	0 | +

Due to limited access for stationary point surveys and constraints in establishing mobile transects (see Chapter 1), some stationary point locations were near cell edges and short segments of some mobile transects were up to 2.2 km outside cells. Additionally, we may have detected bats within cells that commuted from areas outside cells. Thus, we buffered cell boundaries by 2.2 km before we measured covariates to include relevant landscape effects similar to other studies (Yates and Muzika 2006, Duchamp and Swihart 2008, Farrow and Broders 2011, Bender et al. 2015)

Annual turnover rates (i.e., colonization and extinction) at the landscape scale could indicate changes in species range due to land cover changes or regional threats to bat populations, such as WNS, and could also be higher for migratory species. Therefore, we hypothesized higher turnover rates for MYELUSE and PESU, since they are affected by WNS in the northwestern part of our study area (Loeb et al. 2016), and LACI, since they exhibit migratory behavior (Cryan 2003). We did not predict high rates of turnover for species with statewide distributions that are not affected by WNS.

We used a multi-season Bayesian occupancy modeling approach to evaluate the influence of hypothesized environmental factors on the probability of occupancy for each species and to calculate turnover rates. This approach models the probability of

occupancy for each sample unit in each sampling period as a temporally autoregressive function of intercept and sample unit-level covariate effects with parameterization for sample unit-level colonization and survival, and models probability of detection, which is dependent on presence of the species, as a function of intercept and survey-level covariate effects (Rodhouse et al. 2015). We treated cells as our sample unit, considered each night at each point or transect as a separate survey occasion, and created presence/non-detection tables for each species on each survey occasion. Because we surveyed each cell within one week each year, we treated populations as closed within years, and open between years and calculated turnover rates for each species between the two years as the probability that an unoccupied cell became occupied (i.e., colonization), and an occupied cell became unoccupied (i.e., extinction). We used non-informative priors and treated all terms as fixed effects. To fit models, we used JAGS version 4.1.0 (http://mcmc-jags.sourceforge.net/) through package 'rjags' (Plummer 2016) in program R version 3.3.3 (https://www.r-project.org/). We ran three independent chains of 25,000 iterations, discarded an initial 5,000 iterations as burn-in, and retained every fourth iteration for a total of 18,750 iterations per model. We assumed model convergence when the potential scale reduction factor (i.e., the Brooks-Gelman-Rubin diagnostic; R-hat) of each parameter was < 1.1. We ranked models using the Widely Applicable Information Criterion (WAIC), which we calculated for each model using the package 'loo' version 1.1.0 (Vehtari et al. 2016), and considered models closely competing when they were within 2.0 WAIC from the top ranked model (Burnham and Anderson 2003). We calculated ΔWAIC from the top ranked model, then calculated each model's relative

likelihood, and finally calculated model weights to evaluate relative support of the models. We considered a covariate effect significant if the estimated 95% credible intervals did not include zero. To generate predicted range maps, we measured environmental covariates in all 893 10 x 10 km NABat cells throughout South Carolina and calculated estimated occupancy rates for each cell based on effect estimates in the top ranked occupancy model for each species, if the model was non-null (i.e., included environmental covariate effects).

To account for imperfect detection, we modeled detection probabilities for each species independently and modeled occupancy as a function of intercept only. We used nine survey variables (*Type*, *Duration*, *Clutter*, *Date*, *Issue*, *Temp*, *RH*, *Wind*, and *Rain*) to test hypothesized effects of single-term models and additive effects models (Chapter 1). We included the covariates from the top ranked detection model for each species in the occupancy modeling process (Table A-5).

We used single term models and models with additive effects of some covariates based on a priori predictions to model occupancy of each species (Table 2.2). We also tested null and global models. Prior to model fitting, we standardized all continuous covariates to have a mean of 0 and standard deviation of 1. We used Pearson's correlation to test for correlations among covariates and considered those with a Pearson's $|r| > 0.7$ as correlated and did not include them in the same model. *Pri*, *Sec*, and *Ter* road classes were correlated with *Dev*, *Sec* was correlated with *Ter*, and *F.Wet* was correlated with *F.Wet.ED* (Table A-4). Since DAIN and MYAUS are associated with forested wetlands (Reid 2006, Carver and Ashley 2008), we substituted *Forest* and *F.ED* with *F.Wet* and

F.Wet.ED, respectively, and omitted *F.Wet.ED* from the global model for these species because it was significantly correlated with *F.Wet*.

Table 2.2: A priori reasoning for 11 occupancy models that we tested for each species, independently. We also tested null and global models. Refer to Table 2.1 for descriptions of each covariate.

Model	Reasoning
Region	May be significant for species with limited distributions
Ag + Dev + Forest/F.Wet	Land cover measures may be good predictors of habitat quality
Region + Ag + Dev + Forest/F.Wet	Land cover composition can vary within regions
Contagion	Some species require continuous tracts of preferred habitat
F.ED/F.Wet.ED	Many species forage along edges
Forest/F.Wet + Contagion	Some species are associated with contiguous tracts of forest cover
Stream	Streams often occur at habitat edges, and they may be important sources of drinking water and foraging areas
Stream + F.ED/F.Wet.ED	Streams along forest edges may be more important than those in forest interiors or urban and agricultural areas
Stream + Ag + Dev + Forest/F.Wet	May describe important foraging and roosting habitat
Pri + Sec + Qua	Roads may act as edges for foraging and commuting
Ag + Dev + Forest/F.Wet + Qua	May predict habitat quality and areas for foraging and commuting

We evaluated model performance with k-fold cross-validation for the top detection model, and each model with WAIC ≤ the null occupancy model. We randomly partitioned the data five times, with 66% as training datasets and the remainder of the data as testing datasets. We reviewed partitions to be sure each training dataset included at least one cell from each of the five ecoregions, and used the same partitions to evaluate models for each species. We used the package 'ROCR' version 1.0.7 (Sing et al. 2005) to calculate area under the receiver-operating curve (AUC) for each model. AUC values range from 0 to 1, where 0.5 indicates no predictive power and 1.0 indicates perfect predictive performance (Cumming 2000).

RESULTS

Survey Results

We recorded 61,397 call files in 2015, 21,972 of which passed our noise filter, and 65,727 call files in 2016, 42,960 of which passed our noise filter. We classified 15,292 and 27,380 call files to species in 2015 and 2016, respectively. We never detected CORA and we detected LABOLASE in every cell each year; therefore, we were unable to model occupancy for these species.

Occupancy Modeling

We found minimal support of top ranked occupancy models among species based on low model weights, and most top ranked models had low predictive performance (Table 2.3). The null model ranked highest for EPFULANO and MYAUS. NYHU had two top ranked models, and DAIN, LACI, MYLELUSE, and NYHU had at least three models that closely competed with the top ranked model (i.e., within 2.0 WAIC; Table 2.3). Additionally, the predictive performance (AUC) of the top ranked models varied, from 0.46 for LACI to 0.76 for NYHU (Table 2.3). EPFULANO and LACI had top ranked models with predictive performance < 0.5 (Table 2.3). DAIN, LACI, MYLELUSE, NYHU, and TABR had top ranked models with equivalent or lesser predictive performance than competing models (Table 2.3). All covariates in all occupancy models reached convergence, except the Piedmont region in two models for EPFULANO.

Table 2.3: Occupancy probability models for each species which performed better than the null model, ordered from highest to lowest performance based on WAIC. A model with only "." indicates the null model (i.e., intercept only) and "+" indicates additive effects models. Model weights based on WAIC scores, and predictive performance based on area under the receiver operator curve (AUC) are shown. Refer to Table 1.1 for species codes, Table 2.1 for covariate descriptions, Table A-6 for covariate beta estimates, and Table A-5 for detection covariates used in modeling.

Species	Occupancy Model	WAIC	Weight	AUC
DAIN	*Region*	122.9	0.15	0.65
	Stream + Ag + Dev + F.Wet	123.3	0.12	0.58
	Stream	123.4	0.12	0.66
	Pri + Sec + Qua	123.6	0.10	0.52
	Region + Ag + Dev + F.Wet + Contagion + Stream + Qua	124.1	0.08	0.65
	Ag + Dev + F.Wet + Qua	124.1	0.08	0.52
	Ag + Dev + F.Wet	124.6	0.06	0.51
	.	124.6	0.06	0.53
EPFULANO	.	440.0	0.12	0.48
LACI	*Stream + F.ED*	170.8	0.39	0.46
	F.ED	171.8	0.24	0.53
	.	172.1	0.21	0.38
MYAUS	.	214.7	0.24	0.62
MYLELUSE	*Region*	81.7	0.21	0.62
	F.ED	82.7	0.13	0.35
	.	83.2	0.10	0.67
NYHU	*F.ED*	457.4	0.12	0.76
	Stream + F.ED	457.4	0.12	0.76
	Pri + Sec + Qua	457.7	0.10	0.68
	.	457.8	0.10	0.34
PESU	*Ag + Dev + Forest + Qua*	467.8	0.28	0.71
	Ag + Dev + Forest	468.9	0.16	0.53
	Stream + Ag + Dev + Forest	469.1	0.15	0.52
	Region + Ag + Dev + Forest	469.6	0.11	0.53
	Region + Ag + Dev + Forest + Contagion + F.ED + Stream + Qua	469.9	0.10	0.52
	Stream + F.ED	471.6	0.04	0.35
	Forest + Contagion	472.2	0.03	0.52
	F.ED	472.2	0.03	0.41

	Pri + Sec + Qua	472.3	0.03	0.49
	.	472.4	0.03	0.48
TABR	*Pri + Sec + Qua*	390.9	0.29	0.60
	.	392.4	0.14	0.60

We observed support for the *Region* model for DAIN and MYLELUSE (Table 2.3). Mean estimated probability of occupancy for DAIN was significantly higher in the Southern Coastal Plain than all other regions except the Blue Ridge (Figure 2.2). The mean occupancy probability estimate for MYLELUSE was highest in the Blue Ridge region, but it only significantly differed from the estimate for the Southeastern Plains (Figure 2.2).

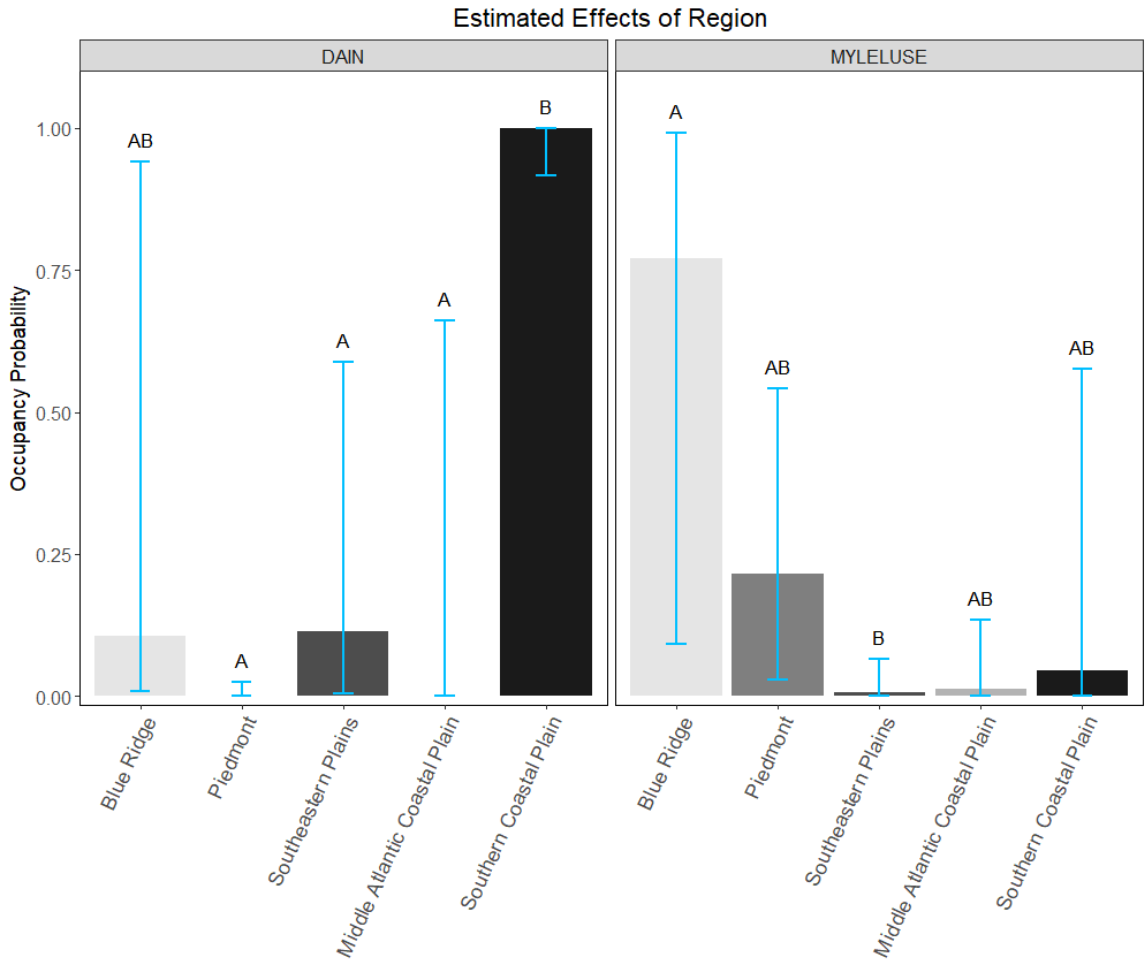


Figure 2.2: Mean estimated probability of occupancy within each ecoregion for species with *Region* retained in their top ranked models. Blue bars indicate 95% credible intervals. Within species, regions which share a letter above their intervals are not significantly different from one another. Refer to Table 1.1 for species code definitions.

We found support for the *Stream* + *F.ED* model for LACI and NYHU. Stream length did not significantly affect the probability of occupancy for either species (Table 2.1; Table A-6). Forest edge density significantly affected the probability of occupancy for LACI, but did not significantly affect the probability of occupancy for NYHU (Table 2.1; Table A-6). From the lowest (0.50 m/ha) to the highest (101.43 m/ha) forest edge

density, LACI probability of occupancy decreased from 98% to 5%, with a steep negative slope beginning at 50 m/ha (Figure 2.3). In addition to the *Stream + F.ED* model, we also found equivalent support for the single-term *F.ED* model for NYHU (Table 2.3), where the effect of forest edge density was negative but not statistically significant (Table 2.1; Table A-6).

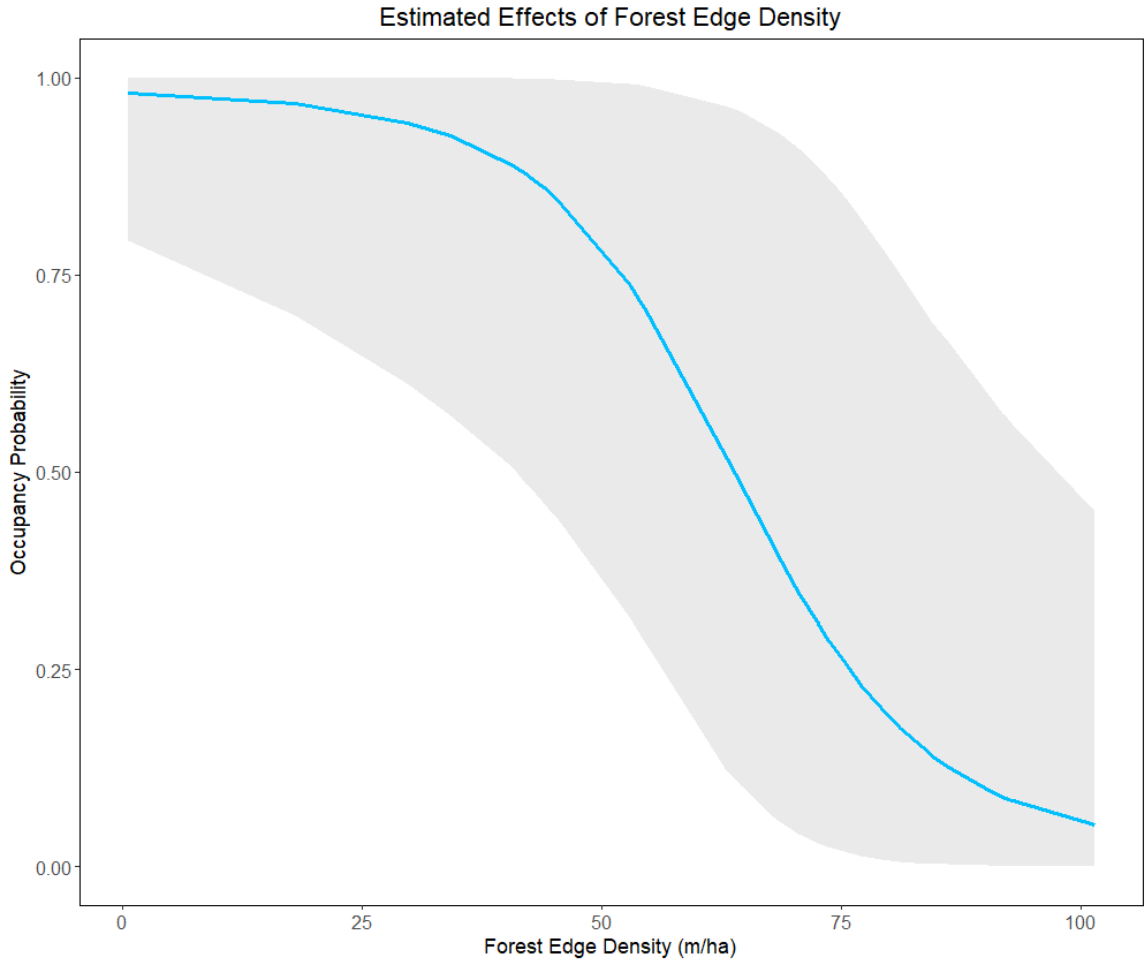


Figure 2.3: Estimated effect of forest edge density (m/ha) on LACI probability of occupancy based on the top ranked model. Gray shading indicates the 95% credible interval. Refer to Table 1.1 for species code definitions.

We found support for the *Ag* + *Dev* + *Forest* + *Qua* model for PESU (Table 2.3). Occupancy probability of PESU was negatively associated with increasing agricultural cover and positively associated with increasing developed land, forest cover, and quaternary road length (Table 2.1). However, none of these effects were statistically significant (Table 2.1; Table A-6).

We found support for the *Pri* + *Sec* + *Qua* model for TABR (Table 2.3). Occupancy probability was positively associated with increasing lengths of all road classes (Table 2.1). Increasing lengths of secondary and quaternary road classes had stronger positive effects on occupancy than the primary road class. However, none of these effects were significant (Table 2.1; Table A-6).

Based on the top ranked model(s), estimated mean probabilities of occupancy and turnover rates varied among species (Figure 2.4, Table 2.4). MYLELUSE had the lowest estimated mean probability of occupancy (0.12) and NYHU had the highest estimated mean probability of occupancy (0.96; Figure 2.4). Both top ranked models for NYHU produced the same estimates of occupancy. From 2015 to 2016, occupancy probabilities declined for EPFULANO, LACI, MYAUS, and NYHU and increased for DAIN, MYLELUSE, PESU, and TABR, but estimates did not significantly differ between years for any species (Figure 2.4). Turnover rates ranged from 0.02 for NYHU (both top ranked models) to 0.55 for MYLELUSE (Table 2.4) and 95% credible intervals were narrow for species with low turnover rates and wide for species with higher turnover rates (Table 2.4).

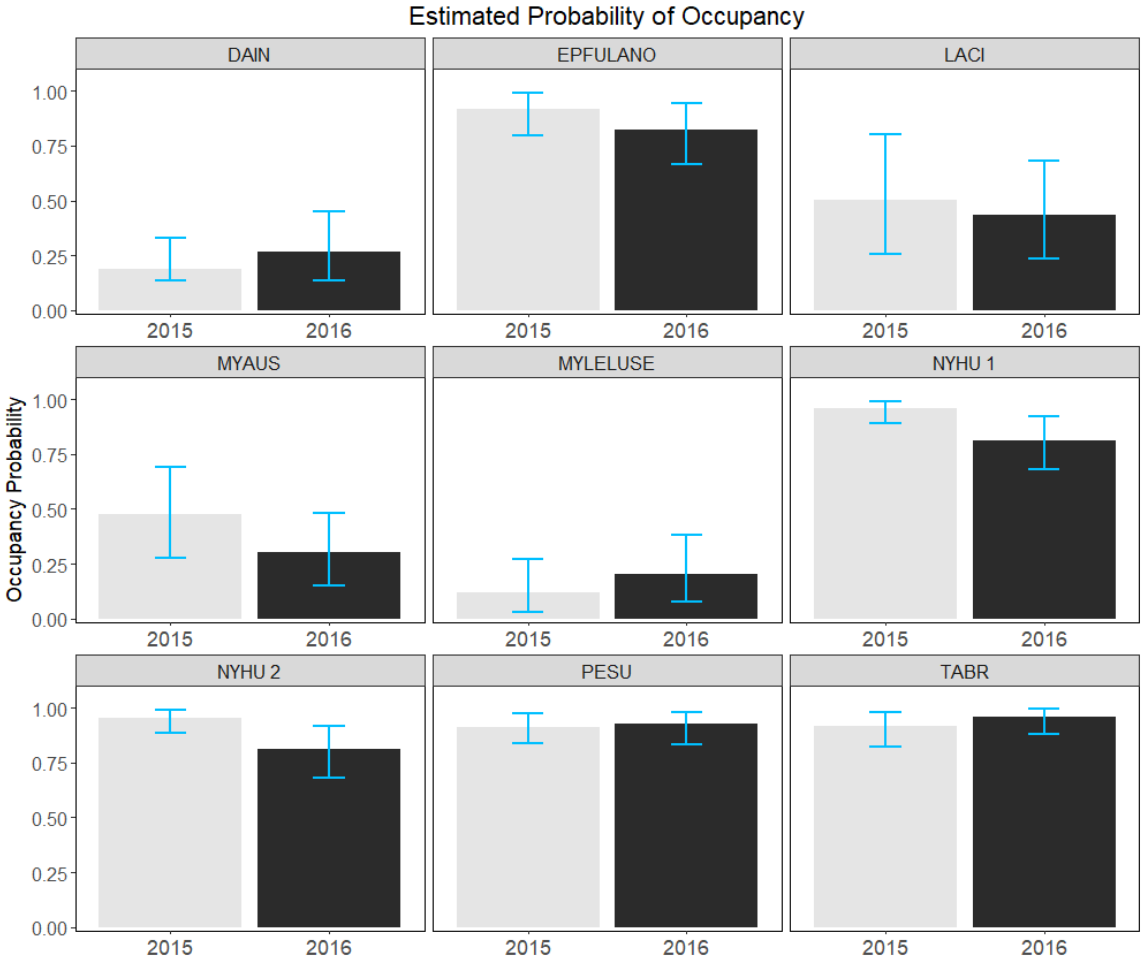


Figure 2.4: Estimated mean probability of occupancy each year based on the top ranked model for each species. NYHU had two top ranked models; NYHU 1 refers to the *F.ED* model and NYHU 2 refers to the *Stream* + *F.ED* model. Blue bars indicate 95% credible intervals. Refer to Table 1.1 for species code definitions.

Table 2.4: Estimated turnover rates and 95% credible intervals (Lower and Upper CI) based on the top ranked model for each species. Refer to Table 1.1 for species codes.

Species	Turnover	Lower CI	Upper CI
DAIN	0.44	0.10	0.73
EPFULANO	0.06	0.01	0.17
LACI	0.42	0.06	0.80
MYAUS	0.18	0.01	0.50
MYLELUSE	0.55	0.12	0.90
NYHU 1	0.02	5E-4	0.07
NYHU 2	0.02	6E-4	0.08
PESU	0.06	0.01	0.13
TABR	0.07	0.01	0.16

We found that predicted distribution maps based on top ranked occupancy models differed among species, but closely matched 2003 known ranges (Figure 2.5). DAIN and MYLELUSE, species for which *Region* was the top ranked model, each had high probabilities of occupancy in ecoregions that are completely within their 2003 known ranges, and neither species had a predicted occupancy greater than 30% outside these regions. NYHU 2003 known distribution was statewide, and both models predicted occupancy rates greater than 90% statewide. TABR 2003 known distribution was also statewide, and the model predicted occupancy rates greater than 90% in most areas, except in areas with fewer roads, but rates were consistently above 50%. PESU 2003 known distribution was statewide, but predicted occupancy rates were lowest, down to 15%, in areas with proportionally high agricultural land cover, and high throughout the rest of the state. LACI predicted occupancy rates were highest in the Blue Ridge region, which fully encompasses their 2003 known range. However, LACI occupancy was also

high in much of the Southern Coastal Plain region and areas of other regions where stream length was high and forest edge density was low.

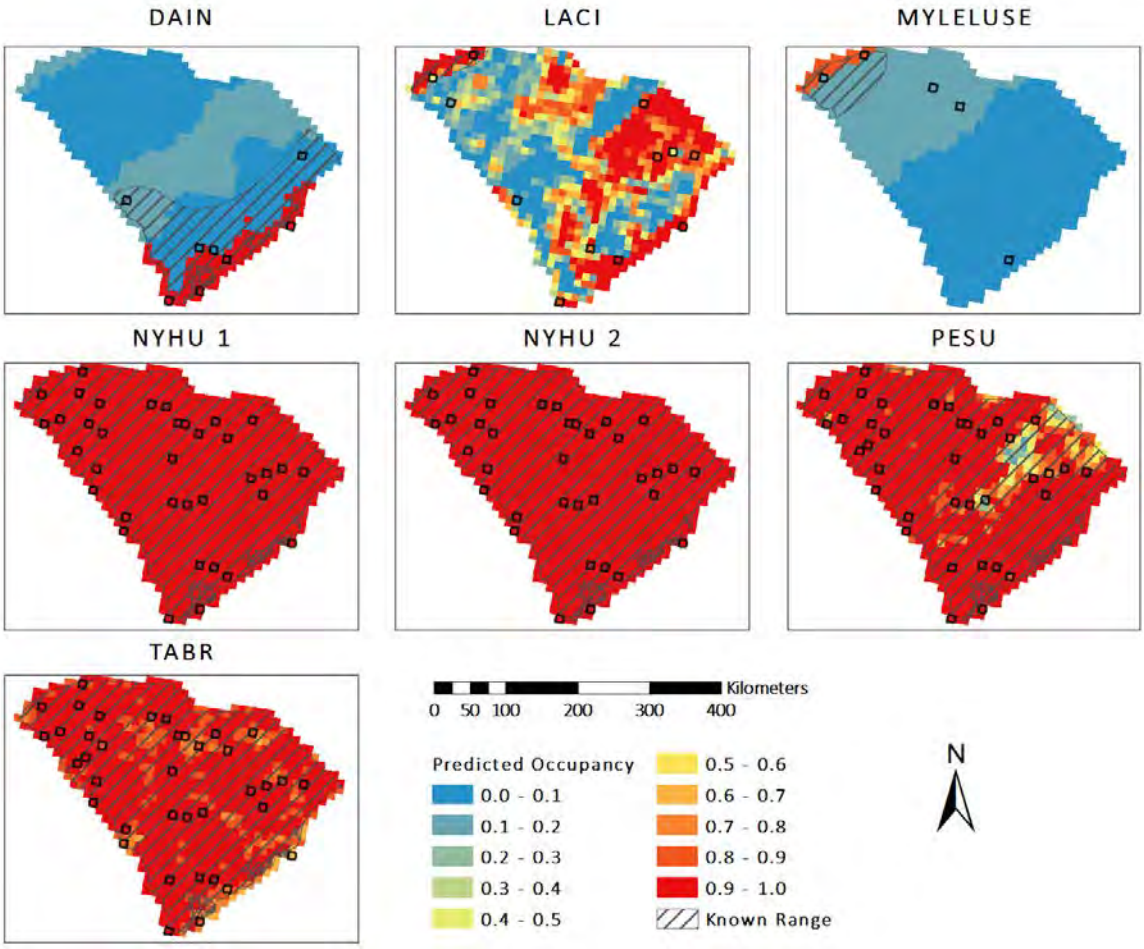


Figure 2.5: Predicted occurrence maps for each species based on effect estimates in their top ranked occupancy model, if non-null, and measures of environmental covariates. Black-outlined squares indicate cells where species were detected in 2015, 2016, or both years. Known summer ranges are based on a 2003 report (Menzel et al. 2003). Refer to Table 1.1 for species code definitions.

DISCUSSION

We found that the NABat framework was effective at detecting most bat species that occur in South Carolina, but that our ability to evaluate how environmental factors

influence landscape occupancy rates was generally limited and highly variable among species. This suggests, depending on the species being studied, that data collected from NABat acoustic surveys may be better suited for analyses of effects of finer-scale habitat conditions, which is similar to the findings of other bat occupancy studies (Loeb and O'Keefe 2006, Hein et al. 2009, Bender et al. 2015). However, for most species, our predicted distribution maps appear to closely match what is known about the summer habitat associations of these species. Our data can be incorporated into future analyses and may be used to study changes in bat habitat usage, population declines, and changes in their distributions over time (Rodhouse et al. 2015).

At a landscape scale, U.S. Level III Ecoregions describe many environmental characteristics, including habitat types and quality of resources (U.S. Environmental Protection Agency 2011), and may incorporate many of the features we predicted affect bat species occupancy. As we expected, we found the highest probability of occupancy for DAIN in the Southern Coastal Plain region. In our study area, the known range of DAIN includes the Southern Coastal Plain, Mid Atlantic Coastal Plain, and southern Southeastern Plains (Menzel et al. 2003). Therefore, we expected to find a significant difference in DAIN occupancy between Blue Ridge and Southern Coastal Plain regions, but did not. The Blue Ridge region was only a small part of our study area and had only one priority cell, which led to a very wide 95% credible interval for the estimate in this region and relatively low predictive performance of the *Region* model (AUC = 0.65). For MYLELUSE, we found a significantly higher mean estimated probability of occupancy in the Blue Ridge region than the Southeastern Plains region, as we expected based on

their known occurrence in the Blue Ridge region (Menzel et al. 2003). We did not expect to find a lack of significant difference between the Blue Ridge and coastal plain regions, but, like the case with DAIN, this may again be due to only one priority cell in the Blue Ridge region leading to a wide credible interval. Additionally, we detected MYLELUSE in three of the five regions, but they were not detected in many cells throughout each region, which likely led to overlapping credible intervals among regions and relatively low predictive performance (AUC = 0.62). However, regional mean occupancy estimates appear to reflect known ranges of the species within the MYLELUSE group, in addition to a slight probability of occupancy in the Southern Coastal Plain, due to one detection of MYSE. This detection was later verified by subsequent mist-net captures of MYSE in the Southern Coastal Plain (White et al. 2017, in review; http://www.dnr.sc.gov/news/2017/july/jul7_longearbats.html).

We also predicted significant effects of *Region* for LACI and MYAUS, but we did not find this result. LACI appear to have more widespread summer distributions within our study area than expected, reducing the significance of *Region*. MYAUS were not detected in many cells within the regions they are thought to occupy, possibly due to their preference for forested wetlands (Reid 2006, Carver and Ashley 2008) that may occur in small isolated patches and therefore were not heavily surveyed. When conducting NABat surveys, it may therefore be especially important to conduct stationary survey points in a variety of habitats within each cell to improve probability of detection for species with such specific habitat requirements, as suggested in the NABat plan (Loeb et al. 2015).

We found a negative association between forest edge density and LACI occupancy, but this effect may not be representative of LACI foraging preferences. Other studies of LACI foraging activity found preferences for forest edge and openings (Ford et al. 2005, Jantzen and Fenton 2013), forest interior (Veilleux et al. 2009), or no strong preference between forest edges and opening interiors (Brooks et al. 2017). LACI exhibit migratory behavior and the majority of individuals may move north, out of our study area during summer (Cryan 2003), which may explain why we found a very low mean probability of detection (4.0%; Chapter 1), relatively high turnover rate (0.42; Table 2.4), and a poor predictive performance for the top ranked occupancy model. Although LACI occupancy estimates were relatively high (averaged about 49%), the relatively high turnover rate may be a further indication of transient individuals opportunistically using habitat. Further investigation of LACI summer habitat use (e.g., radio tracking of individuals) may explain the potential effects of forest edge density.

For many species in our study, it may be the case that analysis at the cell level was too broad to detect significant factors that affected occupancy probabilities. The null model likely ranked highest for EPFULANO because it is a generalist species (Reid 2006), so landscape scale environmental covariates were likely unable to explain slight differences in occupancy. Compared to EPFULANO, MYAUS is more of a habitat specialist and typically roosts in tree cavities in wetland habitats and forages near streams (Reid 2006, Carver and Ashley 2008). Due to their specific habitat requirements, measures of forested wetlands and stream lengths at the cell level may not have been appropriate for modeling MYAUS occupancy, and a finer scale analysis may be more

suitable. Although environmental models ranked higher than the null model for NYHU, PESU, and TABR, none of the effects were significant. These species, along with EPFULANO, occupied most of our study area, among a variety of habitats, which could explain why we failed to find significant effects of any landscape scale environmental factors for these species. In a similar occupancy study in Missouri, Starbuck et al. (2015) found greater effects of habitat conditions at the landscape scale than habitat conditions at more localized scales. However, they analyzed occupancy and assessed effects of habitat conditions at each survey point, while we analyzed occupancy and assessed habitat conditions at the scale of 100 km^2 cells, and landscape factors that affect bat occupancy at points near cell edges may not be represented by cell-level metrics. Thus, conducting multi-scale analyses, with landscape occupancy at the cell-level and site use at the level of stationary points and mobile transects, accompanied by more localized measures of habitat conditions, may produce significant results that reflect bat species roosting and foraging preferences.

We were able to generate predicted occurrence maps for species with non-null top ranked occupancy models, and these maps appear to accurately represent what is known about the summer habitat use of most species. For instance, DAIN and MYLELUSE occurrence predictions are highest in regions where they were known to occur (but see Chapter 1 for a discussion of MYSE extra-range detection), PESU are positively associated with forest cover (Farrow and Broders 2011) and were predicted to have lower occupancy rates in areas with low forest coverage and high agricultural coverage, and NYHU and TABR, typically found throughout the state, generally had high predicted

occupancy rates statewide. We observed low predictive performance of occupancy models for most species, especially for species with low probabilities of detection (see Chapter 1; Table 2.3), which could be an indication of biased AUC estimates due to false negatives (i.e., non-detection of species where they were actually present; Zipkin et al. 2012) However, our data may not have been suitable for assessment with k-fold cross validation and AUC because we had a small sample size (i.e., 38 cells), which was divided into subsets of 25 cells for training data and 13 cells for testing data. Our top ranked models for most species may therefore be sufficient for predicting landscape occupancy and could be used to guide future mist netting efforts and updating species range maps, even in cases where we determined covariate effects were not significant and found models had low predictive performance.

Although we classified more calls to species in 2016 than 2015, we did not find significantly differing mean estimated probabilities of occupancy between years for any species at the landscape scale over the two years of our study, but turnover rates varied among species. Estimated turnover rates appear to be related to species detection probabilities, where species with high detection probabilities had low turnover rates, averaging about 5%, and species with lower detection probabilities had higher turnover rates, averaging about 40% (see Chapter 1; Table 2.4). The higher turnover rate for LACI may be related to our potential detection of transient individuals, due to their migratory behavior (Cryan 2003). DAIN and MYLELUSE high turnover rates and low probabilities of detection could indicate false negatives (i.e., non-detection where species were actually present) each year. However, species with higher turnover rates also had wide

credible intervals, so these results should be interpreted with caution, and further study is needed. We may have been able to classify more calls to species in 2016 than 2015 due to the addition of three cells, more stationary points, one additional mobile transect, and more recording nights for three stationary points in 2016, but occupancy estimates did not differ between years for any species, and we do not believe this affected analyses. Overall, populations of some species may be declining in our study area (Loeb et al. 2016) but it was not evident in our study at the landscape scale. In contrast, other studies at similar spatial scales detected some changes in bat populations over time, but these studies were at longer temporal scales (eight to 15 years; Roche et al. 2012, Barlow et al. 2015, Rodhouse et al. 2015). Thus, if the monitoring we initiated is continued, it is likely that managers will be able to better detect impacts of WNS and other threats to bat populations in our study area (Roche et al. 2009).

For the first time in South Carolina, we implemented standardized, statewide acoustic monitoring of bats that revealed landscape scale effects on the probability of occupancy for some species and generated predicted occurrence maps which could be used to guide future studies and to update species range maps. Additionally, our study has provided baseline data on occurrence of many bat species, which can be analyzed at various scales, and may potentially reveal further effects of land cover variables on bat species occupancy and changes in bat populations over time if monitoring is continued (Roche et al. 2009, Loeb et al. 2015). Results of our study and future analyses of our data (e.g., with finer scale habitat measures, and more years of data) can therefore increase ecological knowledge of bats and be used to inform conservation priorities (Roche et al.

2012, Loeb et al. 2015, Rodhouse et al. 2015), which is critical to the sustainability of bat populations due to the numerous threats they currently face.

MANAGEMENT RECOMMENDATIONS

We recommend continuing NABat surveys throughout South Carolina to monitor bat populations throughout the state and determine if species decline over time. For occupancy analyses of NABat acoustic survey data, we recommend taking a multi-scale approach. This approach should account for probability of detection (see Chapter 1), utilize site-level (i.e., stationary point or mobile transect) habitat measures to model site-use, and utilize landscape scale (i.e., 10 x 10 km NABat cell) environmental measures to model landscape occupancy. These multi-scale analyses should reveal effects of habitat at multiple spatial scales, and could provide habitat management guidelines for all bat species, whether they are found statewide or have more limited distributions. Findings from these analyses can also be used to update species range maps.

APPENDICES

Appendix A

Tables

	Type	Duration	Clutter	Date	Issue	Temp	RH	Wind	Rain
Type	**1.00**	**1.00**	**0.81**	-0.12	0.01	-0.22	0.20	0.08	0.13
Duration	**1.00**	**1.00**	**0.80**	-0.15	0.00	-0.24	0.21	0.08	0.13
Clutter	**0.81**	**0.80**	**1.00**	0.04	-0.10	-0.08	0.09	0.12	0.07
Date	-0.12	-0.15	0.04	**1.00**	0.02	0.55	-0.43	0.20	-0.10
Issue	0.01	0.00	-0.10	0.02	**1.00**	0.11	0.02	0.03	0.03
Temp	-0.22	-0.24	-0.08	0.55	0.11	**1.00**	-0.61	0.26	-0.09
RH	0.20	0.21	0.09	-0.43	0.02	-0.61	**1.00**	-0.10	0.32
Wind	0.08	0.08	0.12	0.20	0.03	0.26	-0.10	**1.00**	0.27
Rain	0.13	0.13	0.07	-0.10	0.03	-0.09	0.32	0.27	**1.00**

Table A-1: Results of Pearson's correlation test for each detection covariate we tested. We considered an absolute r-value > 0.7 (black background with white text) as an indication of significant correlation and did not include those covariates in the same models.

Species	2015	2016	% within range 2015	% within range 2016	% within prediction 2015	% within prediction 2016	# outside range 2015	# outside range 2016
CORA	0	0	0.0	0.0	NA	NA	0	0
DAIN	3	7	37.5	63.6	50.0	100.0	0	0
EPFULANO	30	28	85.7	73.7	NA	NA	0	0
LABOLASE	35	38	100.0	100.0	NA	NA	0	0
LACI	6	8	100.0	50.0	23.5	21.1	4	7
MYAUS	11	7	47.6	25.0	NA	NA	1	1
MYLE	1	0	20.0	0.0	NA	NA	0	0
MYLU	2	2	50.0	100.0	NA	NA	1	0
MYSE	2	1	25.0	0.0	NA	NA	1	1
NYHU	34	31	97.1	81.6	97.1	81.6	0	0
PESU	33	36	94.3	94.7	97.1	94.6	0	0
TABR	33	38	94.3	100.0	94.3	100.0	0	0
MYOTIS	3	4	NA	NA	NA	NA	NA	NA
MYLELUSE	3	4	40.0	40.0	100.0	100.0	1	2

Table A-2: Number of NABat survey cells where we detected each species or grouping in 2015 and 2016. “% within range” and “% within prediction” columns list the percentage of cells surveyed within each species’ 2003 known range, and predicted range (see Chapter 2), respectively, in which they were detected each year. “# outside range” columns indicate number of cells surveyed outside each species’ 2003 known range in which they were detected each year. Refer to Table 1.1 for species code definitions.

Species	Parameter	β estimate	Lower CI	Upper CI	R-hat
DAIN	Intercept	-3.38	-4.89	-1.38	1.00
	Clutter 0	0.00	0.00	0.00	1.00
	Clutter 1	3.10	1.03	4.73	1.00
	Clutter 2	1.43	-0.78	3.32	1.00
	Clutter 3	-2.15	-64.03	59.73	1.00
	Issue	2.54	0.76	4.59	1.00
EPFULANO	Intercept	0.18	-0.31	0.69	1.00
	Clutter 0	0.00	0.00	0.00	0.00
	Clutter 1	2.39	1.59	3.20	1.00
	Clutter 2	1.06	0.39	1.73	1.00
	Clutter 3	-0.66	-1.89	0.62	1.00
	Issue	-0.26	-1.08	0.61	1.00
	Date	1.18	0.85	1.54	1.00
	Temp	0.69	0.31	1.07	1.00
	RH	0.26	-0.05	0.58	1.00
	Wind	-0.16	-0.43	0.10	1.00
	Rain	-0.35	-1.03	0.32	1.00
LABOLASE	Intercept	4.55	3.71	4.98	1.00
	Clutter 0	0.00	0.00	0.00	0.00
	Clutter 1	-0.57	-1.67	0.66	1.00
	Clutter 2	-0.36	-1.51	1.02	1.00
	Clutter 3	-3.25	-4.41	-1.89	1.00
	Issue	-2.08	-3.14	-0.94	1.00
LACI	Intercept	-3.32	-4.63	-2.04	1.00
	Duration	2.96	1.05	4.86	1.00
	Temp	-0.42	-0.77	-0.08	1.00
MYAUS	Intercept	-0.88	-1.35	-0.45	1.00
	Duration	0.93	0.42	1.51	1.00
	Issue	-1.39	-2.54	-0.39	1.00
MYLELUSE	Intercept	-2.60	-4.35	-1.09	1.00
	Clutter 0	0.00	0.00	0.00	0.00
	Clutter 1	-0.57	-2.88	1.64	1.00
	Clutter 2	2.15	0.46	4.05	1.00
	Clutter 3	4.09	1.13	7.69	1.00
	Issue	-3.11	-4.91	-0.49	1.00
NYHU	Intercept	1.58	1.32	1.85	1.00
	Issue	-0.93	-1.58	-0.30	1.00
	Date	-0.21	-0.45	0.03	1.00
	Wind	-0.20	-0.43	0.03	1.00

PESU	Intercept	1.36	1.12	1.60	1.00
	Duration	0.51	0.29	0.73	1.00
	Date	0.35	0.11	0.60	1.00
TABR	Intercept	2.03	1.50	2.59	1.00
	Clutter 0	0.00	0.00	0.00	0.00
	Clutter 1	0.92	0.10	1.77	1.00
	Clutter 2	-0.75	-1.40	-0.12	1.00
	Clutter 3	-1.22	-2.38	-0.03	1.00
	RH	0.28	0.02	0.55	1.00
	Issue	-1.37	-2.15	-0.59	1.00

Table A-3: Estimated β for intercepts and covariates in top ranked detection models for each species, their 95% credible intervals (Lower and Upper CI), and their convergence values (R-hat). See Table 1.1 for species code definitions.

	Region	**Ag**	**Dev**	**Forest**	**F.Wet**	**Contagion**	**F.ED**	**F.Wet.ED**	**Stream**	**Pri**	**Sec**	**Ter**	**Qua**
Region	**1.00**	-0.26	0.16	-0.36	-0.12	0.02	-0.21	-0.08	-0.37	0.20	0.00	0.18	-0.38
Ag	-0.26	**1.00**	-0.11	-0.30	0.25	-0.53	0.04	0.36	0.15	-0.16	0.04	-0.05	0.19
Dev	0.16	-0.11	**1.00**	-0.13	0.24	0.07	0.01	0.19	0.05	**0.72**	**0.83**	**0.95**	-0.20
Forest	-0.36	-0.30	-0.13	**1.00**	-0.62	0.37	0.68	-0.66	0.62	-0.08	0.02	-0.14	-0.18
F.Wet	-0.12	0.25	-0.24	-0.62	**1.00**	-0.53	-0.43	**0.94**	-0.30	-0.20	-0.27	-0.19	0.48
Contagion	0.02	-0.53	-0.07	0.37	-0.53	**1.00**	-0.25	-0.63	-0.04	0.02	-0.17	-0.19	-0.28

	Qua	Ter	Sec	Pri	Stream	F.Wet.ED	F.ED
Qua	1.00	-0.22	-0.19	-0.15	0.00	0.52	-0.03
Ter	-0.22	1.00	0.81	0.68	-0.04	-0.11	0.01
Sec	-0.19	0.81	1.00	0.58	0.15	-0.16	0.21
Pri	0.68	0.58	1.00	-0.16	-0.16	-0.24	0.59
Stream	-0.04	0.15	-0.16	1.00	-0.24	-0.17	-0.14
F.Wet.ED	-0.11	-0.16	-0.17	-0.24	1.00	-0.41	-0.41
F.ED	0.01	0.21	-0.14	0.59	-0.41	1.00	-0.25
	0.52	-0.19	-0.16	-0.17	-0.24	-0.41	-0.25
	0.00	-0.04	0.15	-0.16	0.59	-0.41	-0.43
	-0.15	0.68	0.58	1.00	-0.16	-0.17	-0.14
	-0.19	0.81	1.00	0.58	0.15	-0.16	0.21
	-0.22	1.00	0.81	0.68	-0.04	-0.11	0.01
	-0.19	-0.22	-0.19	-0.15	0.00	0.52	-0.03

Table A-4: Results of Pearson's correlation test for each occupancy covariate we tested. We considered an absolute r-value > 0.7 (black background with white text) as an indication of significant correlation and did not include those covariates in the same models.

Species	Detection Model	AUC
DAIN	*clutter + issue*	0.99
EPFULANO	*clutter + issue + date + temp + RH + wind + rain*	0.88
LACI	*duration + temp*	0.93
MYAUS	*duration + issue*	0.90
MYLELUSE	*clutter + issue*	0.97
NYHU	*issue + date + wind*	0.73
PESU	*duration + date*	0.68
TABR	*clutter + issue + RH*	0.88

Table A-5: Predictive performance (AUC) of the top ranked detection model for each species. “clutter” categorized the level of vegetation clutter at stationary points, “issue” denoted incomplete mobile surveys or stationary point equipment malfunctions, “date” was Julian day, “temp”, “RH”, and “wind” were average temperature, relative humidity, and wind speed during the survey periods, “rain” denoted the occurrence of rain during the survey periods, and “duration” was the length of the survey occasion in minutes. See Table 1.1 for species code definitions.

Species	Parameter	β estimate	Lower CI	Upper CI	R-hat
DAIN	Intercept	-2.07	-4.87	2.80	1.01
	Blue Ridge	0.00	0.00	0.00	0.00
	Mid-Atlantic Coastal Plain	-22.57	-70.25	3.92	1.00
	Piedmont	-25.68	-70.22	-0.85	1.00
	Southeastern Plains	0.02	-5.52	4.10	1.00
	Southern Coastal Plain	27.82	3.96	71.43	1.00
LACI	Intercept	0.14	-2.00	3.26	1.00
	Stream	2.17	-0.61	4.69	1.00
	F.ED	-2.89	-4.84	-0.49	1.00
MYLELUSE	Intercept	1.81	-2.39	4.83	1.00
	Blue Ridge	0.00	0.00	0.00	0.00
	Mid-Atlantic Coastal Plain	-27.46	-72.06	-3.25	1.00
	Piedmont	-3.32	-7.06	1.19	1.00
	Southeastern Plains	-28.03	-72.89	-3.97	1.00
	Southern Coastal Plain	-24.56	-70.47	0.40	1.00
NYHU 1	Intercept	4.06	2.49	4.96	1.00
	F.ED	-0.86	-2.94	1.57	1.00
NYHU 2	Intercept	4.17	2.68	4.97	1.00
	Stream	0.15	-2.08	2.57	1.00
	F.ED	-0.88	-3.16	1.80	1.00
PESU	Intercept	4.41	3.14	4.98	1.00
	Ag	-1.20	-3.36	1.58	1.00
	Dev	1.38	-1.07	4.32	1.00
	For	0.38	-2.15	2.77	1.00
	Qua	1.48	-0.62	4.22	1.00
TABR	Pri	1.06	-1.85	4.46	1.00
	Sec	1.38	-0.75	3.91	1.00
	Qua	1.37	-0.53	4.00	1.00

Table A-6: Estimated β for intercepts and covariates in top ranked occupancy models for each species, their 95% credible intervals (Lower and Upper CI), and their convergence values (R-hat). See Table 1.1 for species code definitions.

REFERENCES

AnalookW. 2016. Titley Scientific. <http://www.titley-scientific.com/us/downloads-support/firmware-software>.
Appel, G., A. López-Baucells, W. E. Magnusson, and P. E. D. Bobrowiec. 2017. Aerial insectivorous bat activity in relation to moonlight intensity. Mammalian Biology - Zeitschrift für Säugetierkunde 85:37–46.
Barlow, K. E., P. A. Briggs, K. A. Haysom, A. M. Hutson, N. L. Lechiara, P. A. Racey, A. L. Walsh, and S. D. Langton. 2015. Citizen science reveals trends in bat populations: The National Bat Monitoring Programme in Great Britain. Biological Conservation 182:14–26.
Bass, H. E., L. C. Sutherland, and A. J. Zuckerwar. 1990. Atmospheric absorption of sound: Update. Journal of the Acoustical Society of America 88:2019–2021.
Bender, M. J., S. B. Castleberry, D. A. Miller, and T. Bently Wigley. 2015. Site occupancy of foraging bats on landscapes of managed pine forest. Forest Ecology and Management 336:1–10.
Bennett, V. J., and A. A. Zurcher. 2013. When corridors collide: Road-related disturbance in commuting bats. The Journal of Wildlife Management 77:93–101.
Boyles, J. G., P. M. Cryan, G. F. McCracken, and T. H. Kunz. 2011. Economic importance of bats in agriculture. Science 332:41–42.
Britzke, E. R., J. E. Duchamp, K. L. Murray, R. K. Swihart, and L. W. Robbins. 2011. Acoustic identification of bats in the eastern United States: A comparison of parametric and nonparametric methods. The Journal of Wildlife Management 75:660–667.
Britzke, E. R., E. H. Gillam, and K. L. Murray. 2013. Current state of understanding of ultrasonic detectors for the study of bat ecology. Acta Theriologica 58:109–117.
Broders, H. G., G. J. Forbes, S. Woodley, and I. D. Thompson. 2006. Range extent and stand selection for roosting and foraging in forest-dwelling northern long-eared bats and little brown bats in the Greater Fundy Ecosystem, New Brunswick. Journal of Wildlife Management 70:1174–1184.
Brooks, J. D., S. C. Loeb, and P. D. Gerard. 2017. Effect of forest opening characteristics, prey abundance, and environmental factors on bat activity in the Southern Appalachians. Forest Ecology and Management 400:19–27.
Burnham, K. P., and D. R. Anderson. 2003. Model selection and multimodel inference: a practical information-theoretic approach. Springer Science & Business Media.
Carver, B. D., and N. Ashley. 2008. Roost tree use by sympatric Rafinesque's big-eared bats (Corynorhinus rafinesquii) and southeastern myotis (Myotis austroriparius). The American Midland Naturalist 160:364–373.
Clement, M. J., and S. B. Castleberry. 2011. Comparison of survey methods for Rafinesque's big-eared bats. Pages 147–157 *in*. Conservation and management of eastern big-eared bats: a symposium. US Forest Service Southern Research Station, Clemson, SC.
Comer, C. E., L. A. Stuemke, M. L. Morrison, and R. W. Maxey. 2014. Comparison of systematic roost searches and acoustic detection to determine occupancy of rare forest bats: Detection of forest bats. Wildlife Society Bulletin 38:103–110.

Cryan, P. M. 2003. Seasonal distribution of migratory tree bats (Lasiurus and Lasionycteris) in North America. Journal of Mammalogy 84:579–593.
Cryan, P. M., and R. M. Barclay. 2009. Causes of bat fatalities at wind turbines: hypotheses and predictions. Journal of Mammalogy 90:1330–1340.
Cumming, G. S. 2000. Using between-model comparisons to fine-tune linear models of species ranges. Journal of Biogeography 27:441–455.
Duchamp, J. E., and R. K. Swihart. 2008. Shifts in bat community structure related to evolved traits and features of human-altered landscapes. Landscape Ecology 23:849–860.
Farrow, L. J., and H. G. Broders. 2011. Loss of forest cover impacts the distribution of the forest-dwelling tri-colored bat (Perimyotis subflavus). Mammalian Biology - Zeitschrift für Säugetierkunde 76:172–179.
Federal Register. 2015. Endangered and threatened wildlife and plants; threatened species status for the northern long-eared bat with 4 (d) rule. Federal Register 80:17974–18033.
Fisher-Phelps, M., D. Schwilk, and T. Kingston. 2017. Mobile acoustic transects detect more bat activity than stationary acoustic point counts in a semi-arid and agricultural landscape. Journal of Arid Environments 136:38–44.
Ford, W. M., M. A. Menzel, J. L. Rodrigue, J. M. Menzel, and J. B. Johnson. 2005. Relating bat species presence to simple habitat measures in a central Appalachian forest. Biological Conservation 126:528–539.
Garmin. 2017. GPS 18x OEM | Garmin. Garmin. <https://buy.garmin.com/en-US/US/p/27594/pn/010-00321-34>. Accessed 22 Jun 2017.
Gooding, G., and J. R. Langford. 2004. Characteristics of tree roosts of Rafinesque's big-eared bat and southeastern myotis in northeastern Louisiana. L. K. Ammerman, editor. The Southwestern Naturalist 49:61–67.
Google. n.d. [Google Maps imagery of South Carolina]. <https://www.google.com/maps>. Accessed 20 Jan 2015.
Gorresen, P. M., P. M. Cryan, K. Montoya-Aiona, and F. J. Bonaccorso. 2017. Do you hear what I see? Vocalization relative to visual detection rates of Hawaiian hoary bats (Lasiurus cinereus semotus). Ecology and Evolution 7:6669–6679.
Grindal, S. D., J. L. Morissette, and R. M. Brigham. 1999. Concentration of bat activity in riparian habitats over an elevational gradient. Canadian Journal of Zoology 77:972–977.
Hammerson, G. A., M. Kling, M. Harkness, M. Ormes, and B. E. Young. 2017. Strong geographic and temporal patterns in conservation status of North American bats. Biological Conservation 212:144–152.
Hawbaker, T. J., V. C. Radeloff, R. B. Hammer, and M. K. Clayton. 2005. Road density and landscape pattern in relation to housing density, and ownership, land cover, and soils. Landscape Ecology 20:609–625.
Hayes, J. P. 1997. Temporal variation in activity of bats and the design of echolocation-monitoring studies. Journal of Mammalogy 78:514–524.
Hein, C. D., S. B. Castleberry, and K. V. Miller. 2009. Site-occupancy of bats in relation to forested corridors. Forest Ecology and Management 257:1200–1207.

Homer, C. G., J. A. Dewitz, L. Yang, S. Jin, P. Danielson, G. Xian, J. Coulston, N. D. Herold, J. D. Wickham, and K. Megown. 2015. Completion of the 2011 National Land Cover Database for the conterminous United States - Representing a decade of land cover change information. Photogrammetric Engineering and Remote Sensing 81:345–354.

Horn, J. W., E. B. Arnett, and T. H. Kunz. 2008. Behavioral Responses of Bats to Operating Wind Turbines. Journal of Wildlife Management 72:123–132.

Jantzen, M. K., and M. B. Fenton. 2013. The depth of edge influence among insectivorous bats at forest–field interfaces. Canadian Journal of Zoology 91:287–292.

Jones, G., D. Jacobs, T. Kunz, M. Willig, and P. Racey. 2009. Carpe noctem: the importance of bats as bioindicators. Endangered Species Research 8:93–115.

Kitzes, J., and A. Merenlender. 2014. Large Roads Reduce Bat Activity across Multiple Species. R. M. Brigham, editor. PLoS ONE 9:e96341.

Kunz, T. H. 1982. Roosting ecology of bats. Pages 1–55 *in*. Ecology of Bats. Springer, US.

Kunz, T. H., E. B. Arnett, W. P. Erickson, A. R. Hoar, G. D. Johnson, R. P. Larkin, M. D. Strickland, R. W. Thresher, and M. D. Tuttle. 2007. Ecological impacts of wind energy development on bats: questions, research needs, and hypotheses. Frontiers in Ecology and the Environment 5:315–324.

Kunz, T. H., E. Braun de Torrez, D. Bauer, T. Lobova, and T. H. Fleming. 2011. Ecosystem services provided by bats. Annals of the New York Academy of Sciences 1223:1–38.

Langwig, K. E., W. F. Frick, J. T. Bried, A. C. Hicks, T. H. Kunz, and A. Marm Kilpatrick. 2012. Sociality, density-dependence and microclimates determine the persistence of populations suffering from a novel fungal disease, white-nose syndrome. K. Lafferty, editor. Ecology Letters 15:1050–1057.

Law, B., L. Gonsalves, P. Tap, T. Penman, and M. Chidel. 2015. Optimizing ultrasonic sampling effort for monitoring forest bats. Austral Ecology 40:886–897.

Loeb, S. C. 2017. Adaptive response to land-use history and roost selection by Rafinesque's big-eared bats. Journal of Mammalogy 98:560–571.

Loeb, S. C., and J. M. O'Keefe. 2006. Habitat use by forest bats in South Carolina in relation to local, stand, and landscape characteristics. Journal of Wildlife Management 70:1210–1218.

Loeb, S. C., T. J. Rodhouse, L. E. Ellison, C. L. Lausen, J. D. Reichard, K. M. Irvine, T. E. Ingersoll, J. T. Coleman, W. E. Thogmartin, J. R. Sauer, and others. 2015. A plan for the North American bat monitoring program (NABat). General Technical Report, U.S. Department of Agriculture Forest Service, Southern Research Station, Asheville, North Carolina.

Loeb, S. C., E. A. Winters, L. K. L. Burns, and P. Sirajuddin. 2016. Will the South's bats rise again? The devastating effects of White-nose syndrome in South Carolina. Bat Research News 57:76.

Lorch, J. M., J. M. Palmer, D. L. Lindner, A. E. Ballmann, K. G. George, K. Griffin, S. Knowles, J. R. Huckabee, K. H. Haman, C. D. Anderson, P. A. Becker, J. B. Buchanan, J. T. Foster, and D. S. Blehert. 2016. First Detection of Bat White-Nose Syndrome in Western North America. K. McMahon, editor. mSphere 1:e00148-16.

Lucas, J. S., S. C. Loeb, and P. G. R. Jodice. 2015. Roost selection by Rafinesque's big-eared bats (Corynorhinus rafinesquii) in a pristine habitat at three spatial scales. Acta Chiropterologica 17:131–141.

MacKenzie, D. I. 2005. What are the issues with presence-absence data for wildlife managers? Vojta, editor. Journal of Wildlife Management 69:849–860.

Martinuzzi, S., J. C. Withey, A. M. Pidgeon, A. J. Plantinga, A. J. McKerrow, S. G. Williams, D. P. Helmers, and V. C. Radeloff. 2015. Future land-use scenarios and the loss of wildlife habitats in the southeastern United States. Ecological Applications 25:160–171.

Menzel, J. M., M. A. Menzel, J. C. Kilgo, W. M. Ford, J. W. Edwards, and G. F. McCRACKEN. 2005. Effect of habitat and foraging height on bat activity in the Coastal Plain of South Carolina. Russell, editor. Journal of Wildlife Management 69:235–245.

Menzel, M. A., J. M. Menzel, and J. C. Kilgo. 2003. Bats of the Savannah River Site and vicinity. Gen. Tech. Rep. SRS-68. Asheville, NC: U.S. Department of Agriculture, Forest Service, Southern Research Station.

Murray, K. L., E. R. Britzke, B. M. Hadley, and L. W. Robbins. 1999. Surveying bat communities: a comparison between mist nets and the Anabat II bat detector system. Acta Chiropterologica 1:105–112.

O'Donnell, C. 2000. Influence of season, habitat, temperature, and invertebrate availability on nocturnal activity of the New Zealand long-tailed bat (Chalinolobus tuberculatus). New Zealand Journal of Zoology 27:207–221.

O'Farrell, M. J., W. G. Bradley, and G. W. Jones. 1967. Fall and winter bat activity at a desert spring in southern Nevada. The Southwestern Naturalist 12:163.

Patriquin, K. J., and R. M. Barclay. 2003. Foraging by bats in cleared, thinned and unharvested boreal forest. Journal of Applied Ecology 40:646–657.

Patriquin, K. J., L. K. Hogberg, B. J. Chruszcz, and R. M. R. Barclay. 2003. The influence of habitat structure on the ability to detect ultrasound using bat detectors. Wildlife Society Bulletin 31:475–481.

Pauli, B. P., H. A. Badin, G. S. Haulton, P. A. Zollner, and T. C. Carter. 2015. Landscape features associated with the roosting habitat of Indiana bats and northern long-eared bats. Landscape Ecology 30:2015–2029.

Plummer, M. 2016. rjags: Bayesian Graphical Models using MCMC. <https://cran.r-project.org/web/packages/rjags/index.html>.

Racey, P. A. 1982. Ecology of Bat Reproduction. Pages 57–104 *in* T. H. Kunz, editor. Ecology of Bats. Springer US, Boston, MA. <https://doi.org/10.1007/978-1-4613-3421-7_2>.

Rebelo, H., P. Tarroso, and G. Jones. 2010. Predicted impact of climate change on European bats in relation to their biogeographic patterns. Global Change Biology 16:561–576.
Reid, F. A. 2006. A Field Guide to Mammals of North America North of Mexico. 4th edition. The Peterson field guide series, Houghton Mifflin Company, Boston, MA.
Roche, N., S. Langton, and T. Aughney. 2009. The car-based bat monitoring scheme for Ireland: Synthesis Report 2003-2008. Irish Wildlife Manuals 39.
Roche, N., S. Langton, and T. Aughney. 2012. Car-based bat monitoring in Ireland 2003-2011. Irish Wildlife Manuals 60.
Rodhouse, T. J., P. C. Ormsbee, K. M. Irvine, L. A. Vierling, J. M. Szewczak, and K. T. Vierling. 2012. Assessing the status and trend of bat populations across broad geographic regions with dynamic distribution models. Ecological Applications 22:1098–1113.
Rodhouse, T. J., P. C. Ormsbee, K. M. Irvine, L. A. Vierling, J. M. Szewczak, and K. T. Vierling. 2015. Establishing conservation baselines with dynamic distribution models for bat populations facing imminent decline. J. Franklin, editor. Diversity and Distributions 21:1401–1413.
Rydell, J. 1989. Feeding activity of the northern bat Eptesicus nilssoni during pregnancy and lactation. Oecologia 80:562–565.
Sing, T., O. Sander, N. Beerenwinkel, and T. Lengauer. 2005. ROCR: visualizing classifier performance in R. Bioinformatics 21:3940–3941.
South Carolina Department of Natural Resources. 2015. South Carolina bat conservation plan. Columbia, South Carolina.
Starbuck, C. 2013. Bat occupancy of forests and managed savanna and woodland in the Missouri Ozark region. University of Missouri - Columbia.
Starbuck, C. A., S. K. Amelon, and F. R. Thompson. 2015. Relationships between bat occupancy and habitat and landscape structure along a savanna, woodland, forest gradient in the Missouri Ozarks: Bat Occupancy in Savannas. Wildlife Society Bulletin 39:20–30.
Tonos, J. M., B. P. Pauli, P. A. Zollner, and G. S. Haulton. 2014. A comparison of the efficiency of mobile and stationary acoustic bat surveys. Proceedings of the Indiana Academy of Science. Volume 123. <http://search.ebscohost.com/login.aspx?direct=true&profile=ehost&scope=site&authtype=crawler&jrnl=00736767&AN=110454901&h=lVcSohmDfiPOfGQr%2BP2YiNuXYMKXHPfZXZ1WPiLuGcRFZkL5HWgBL2Nf%2FIdlNJrR6YmyJFeQipZuqVWNmgtyIg%3D%3D&crl=c>. Accessed 8 May 2016.
Turner, G. G., and D. M. Reeder. 2009. Update of white-nose syndrome in bats, September 2009. Bat Research News 50:47–53.
United States Fish and Wildlife Service. 2014. White-Nose Syndrome The devastating disease of hibernating bats in North America. <http://digitalcommons.unl.edu/usfwspubs/453/>. Accessed 27 Nov 2015.
U.S. Environmental Protection Agency. 2011. U.S. Level III and IV Ecoregions. U.S. EPA Office of Research and Development, Corvallis, OR.

U.S. Forest Service. 2015*a*. National Forest System Roads. U.S. Forest Service. <https://data.fs.usda.gov/geodata/edw/datasets.php>. Accessed 20 Jan 2015.
U.S. Forest Service. 2015*b*. S_USA.BasicOwnership. U.S. Forest Service. <https://data.fs.usda.gov/geodata/edw/datasets.php>. Accessed 20 Jan 2015.
U.S. Geological Survey. 2014. NLCD 2011 Land Cover (2011 Edition, amended 2014) - National Geospatial Data Asset (NGDA) Land Use Land Cover. U.S. Geological Survey. <http://gisdata.usgs.gov/tdds/downloadfile.php?TYPE=nlcd2011_lc_state&ORIG=META&FNAME=NLCD2011_LC_South_Carolina.zip>. Accessed 20 Jan 2015.
U.S. Geological Survey, National Geospatial Program. 2017. USGS National Hydrography Dataset (NHD). U.S. Geological Survey. <https://viewer.nationalmap.gov>. Accessed 2 Feb 2017.
U.S. Geological Survey, National Geospatial Technical Operations Center. 2016. US Forest Service Roads. U.S. Geological Survey. <https://www.sciencebase.gov/catalog/item/56b9fc25e4b08d617f648ecc>.
USGS, National Geospatial Technical Operations Center. 2014. USGS National Transportation Dataset (NTD) FileGDB 10.1. U.S. Geological Survey. <ftp://rockyftp.cr.usgs.gov/vdelivery/Datasets/Staged/Tran/FileGDB101/TRAN_45_South_Carolina_GU_STATEORTERRITORY.zip>. Accessed 20 Jan 2015.
Vehtari, A., A. Gelman, and J. Gabry. 2016. loo: Efficient leave-one-out cross-validation and WAIC for Bayesian models. <https://CRAN.R-project.org/package=loo>.
Veilleux, J. P., P. R. Moosman, D. Scott Reynolds, K. E. LaGory, and L. J. Walston. 2009. Observations of summer roosting and foraging behavior of a hoary bat (Lasiurus cinereus) in southern New Hampshire. Northeastern Naturalist 16:148–152.
Whitby, M. D., T. C. Carter, E. R. Britzke, and S. M. Bergeson. 2014. Evaluation of Mobile Acoustic Techniques for Bat Population Monitoring. Acta Chiropterologica 16:223–230.
White, T., J. E. Walea, and J. Robinson. 2017. New record of northern long-eared bats in coastal South Carolina. Southeastern Naturalist.
Wolbert, S. J., A. S. Zellner, and H. P. Whidden. 2014. Bat Activity, Insect Biomass, and Temperature Along an Elevational Gradient. Northeastern Naturalist 21:72–85.
Yates, M. D., and R. M. Muzika. 2006. Effect of Forest Structure and Fragmentation on Site Occupancy of Bat Species in Missouri Ozark Forests. Journal of Wildlife Management 70:1238–1248.
Zipkin, E. F., E. H. C. Grant, and W. F. Fagan. 2012. Evaluating the predictive abilities of community occupancy models using AUC while accounting for imperfect detection. Ecological Applications 22:1962–1972.
Zurcher, A. A., D. W. Sparks, and V. J. Bennett. 2010. Why the Bat Did Not Cross the Road? Acta Chiropterologica 12:337–340.

Final Report
for
WM-0288:
Carolinas Regional Acoustic Bat Survey

November 2014 through April 2017

North American Bat Monitoring Program North Carolina (NABat NC)

Dr. Han Li
Dr. Matina Kalcounis-Rueppell
University of North Carolina at Greensboro
Department of Biology
321 McIver St.
312 Eberhart Building
Greensboro, NC 27412
May 2017

TABLE OF CONTENTS

HIGHLIGHTS

HIGHLIGHTS OF NABAT IMPLEMENTATION

- We sampled 57 NABat grid cells (41 with mobile transect survey, 40 with stationary point survey) in two years and collected over 300,000 bat acoustic files.
- We developed four protocol guides and two datasheets for future NABat implementation.
- Development of a multi-stakeholder collaboration network was initiated for future NABat implementation.

HIGHLIGHTS OF FINDINGS OF BATS

- We detected 14 species of bats.
- Mobile transect surveys and stationary point surveys revealed similar general patterns of bat distributions in NC.
- Federally list species:
    - MYSO - mainly concentrated in the Cherokee National Forest near the NC/TN border
    - MYGR - mainly distributed along the Appalachian Mountains between Asheville and Boone
    - MYSE - more found in the coastal plain region than the rest of NC

SUMMARY

The North American Bat Monitoring Program (NABat) in North Carolina was initiated in November 2014 as part of the Carolinas Regional Acoustic Bat Survey project. The goal of the project was to establish standardized summer acoustic surveys across North Carolina to monitor bat distributions and relative abundance, and contribute data to continental trend analyses through a two-year pilot project. In 2015 and 2016, we visited 57 highly ranked NABat grid cells and developed mobile transect surveys for 41 grid cells and stationary point survey for 40 grid cells. Through over 300,000 acoustic files, we generated NABat grid cell specific presence/no detection information. We also generated species specific activity density maps via ArcGIS mapping. We suggest that the future implementation of NABat should involve multiple partners with a centralized coordinator to facilitate participation and integrate data. We also discuss issues of concern that need to be addressed in the future.

LIST OF TABLES

LIST OF FIGURES

TASK

There is a profound need to evaluate trends in bat populations and distributions for assessment of wind energy projects, hydroelectric projects, timber harvest, and state and federal listing of rare and endangered species. In North Carolina, 10 species of bats are listed in the state wildlife action plan as species of conservation concern that require systematic monitoring (North Carolina Wildlife Resources Commission. 2015). Therefore, the North American Bat Monitoring Program (NABat) in North Carolina was initiated in November 2014 as part of the Carolinas Regional Acoustic Bat Survey project.

The project is administered through the University of North Carolina at Greensboro (UNCG) in partnership with the North Carolina Wildlife Resource Commission (NCWRC). The goal of the project was to establish standardized summer acoustic surveys across North Carolina to monitor bat distributions and relative abundance and contribute data to continental trend analyses. Specifically, we adopted protocols in Loeb et al. (2015) to pilot a two-year statewide summer bat acoustic survey project. Through the pilot survey, we expected to select sites and develop site-specific survey protocols suitable to further implement NABat in the future. We also expected to initiate a long-term broad-scale bat monitoring partnership network for NC and start accumulating baseline data for NC bat species. Through the first two-years, we expected to generate baseline information of bats in NC and provide suggestions on how to implement NABat in the future.

APPROACH

NABAT SURVEY PROTOCOLS

The NABat framework provides general guidance for components throughout the bat survey process (Loeb et al. 2015). As much as possible given our logistic constraints, we followed the NABat protocols for sampling site design, mobile transect survey, stationary point survey, acoustic species identification, data management and analysis. As NABat suggests two field survey protocols, mobile transect survey and stationary point survey, as complementary methods, we treated each method as an independent protocol and implemented them separately.

SAMPLING DESIGN

NABat uses a generalized random-tessellation stratified (GRTS) master survey design algorithm and divides the continental United States into 133,307 10- by 10-km (100-km2) grid cell sample units. The GRTS master design assigns a spatially balanced and randomized ordering to all cells, which allows subsampling to be spatially balanced yet randomized by following the GRTS order. This survey design also allows subsampling within a political or natural boundary. We used 100 top ranked GRTS grid cells in North Carolina as the candidate sites to choose from. We selected NABat grid cells for mobile transect survey and stationary point separately, considering travel logistics.

GEOGRAPHICAL SUBSAMPLING

Geographically, North Carolina falls into three regions: the Appalachian Mountains (21% land area) formed mostly by the Blue Ridge and Great Smoky Mountains, the Piedmont Plateau (34%), and the Coastal Plain region (45%). Each region has a unique sub-climate and vegetation which influences a unique bat species composition. An equal probability subsampling strategy was chosen to select grid cells. Thus, the survey effort within a survey season was divided by the proportion of land area. Grid cells in each region were selected by their GRTS order within a region (un-sampleable grid cells replaced by the next ranked in line within a

geographical region). It is important to note that grid cells that crossed regional boundaries were either eliminated or designed in a way that all surveys could be conducted in one side of the cell for just one geographical region.

The field season for NABat is approximately 9 weeks from June to July. Thus, our sampling effort was divided as 1.5 weeks for the mountain region, 3 weeks for the piedmont region, and 4.5 weeks for the coastal region. Scheduling-wise, we further divided the piedmont and coast region efforts into half. We started the field work in the coastal region, then the piedmont, then the mountains. After the mountain region survey, we would return to the piedmont and finished the season in the coastal region. In this way we were able to balance the seasonal variation within June and July. This travel plan also allows the timing of survey at each specific grid cell to be relatively consistent among years.

GRID CELL SELECTION

First, we eliminated grid cells that were mainly overlying neighboring states or large water bodies or mountain tops. Next, within each geographical region, we selected grid cells for mobile transect survey and stationary point survey separately. Due to accessibility and safety reasons, stationary point survey grid cells were harder to locate than mobile transect survey grid cells. Additionally, we incorporated volunteer participants into the mobile transect survey; whereas the survey team conducted all stationary point surveys. Therefore, we had different criteria for selecting grid cells based on survey methods.

Stationary point survey grid cells

Accessible lands within a grid cell or lies very closely (less than 500 m) to a grid cell was our selection criteria for grid cells that were sampled by the stationary point survey method. Any grid cells that did not have accessible lands were eliminated following the GRTS order.

Mobile transect survey grid cells

We had two types og grid cells for mobile transect surveys: volunteer grid cells and survey team grid cells. The selection for these two types of grid cells was different and outlined below.

Volunteer grid cells

In both years, we involved volunteers in the mobile transect survey. In 2015 we focused on involving citizen scientists; whereas we focused on trained biologists in 2016. A comparison of these two approaches is stated in the future implementation section. When selecting grid cells for volunteers, we subsampled the top 100 NABat grid cells in NC to a geographical range within which volunteers were willing to travel. We followed the GRTS order within the travel range for each volunteer to select highly ranked grid cells. A volunteer might take more than one grid cells.

Survey team grid cells

When selecting mobile transect survey grid cells for the survey team, we followed the general GRTS order except if: 1) a cell was taken by a volunteer; 2) a cell did not have enough roads or enough roads that were safe to survey; 3) there was difficult to find lodging or a site for camping. As the survey team was also responsible for setting up the stationary point survey grid cells, cells that had been chosen for stationary point survey were prioritized unless they were not suitable for mobile transect survey. Specifically, we had to eliminate a few cells that were more than 100 miles away from any lodging sites used during the field season. Instead, a 100-mile subsampling zone around a lodging site was used to choose highest ranked grid cells.

Replace grid cells between years

Due to land accessibility changes, grid cells sampled in one year might no longer be available for sampling the following year. The GRTS design is suitable for this scenario where the order would be followed. In addition to replacing grid cells, how each survey method is implemented within a grid cell might have to change between years. This aspect is discussed in each method section below.

MOBILE TRANSECT SURVEY

According to the NABat protocol, the mobile transect survey is conducted with a AnaBat SD 2 bat detector (Titley Scientific, Australia) fixed to the roof of a vehicle, which is driven at a set speed (20 mph) 45 minutes after sunset. The transect needs to be surveyed twice within 7 days in June or July on nights when there is no rain or fog and low wind. The transect route should be mainly on secondary or tertiary roads (at least 2 lanes) with as few stops as possible. The route should cover common habitat types important for bats in as much of the grid cell as possible. The transect should not double back or include gates that require opening and closing. Sections of the route should be > 100 m from each other if the route contains many curves or switchbacks. Areas with dense forested corridors or low canopies (< 3m) should be avoided. When driving the transect, drivers should use their hazard lights. If the vehicle needs to be stopped for more than just a few seconds or go over 20 mph, the detector needs to be paused and noted. The transect route was also recorded via a Mouse GPS unit (Titley Scientific, Australia).

We strictly followed the protocol to plan the transect route using ArcGIS and printed maps. Even though some of our transects ran out of a grid cell, they all had the required length within the grid cell. Some extended length of transects were planned for a convenient stop. All transects were test driven before they were sampled during the field season. We summarized each transect as a map and a turn-by-turn guide (appendix 1). We also developed a data sheet to record covariates for the mobile transect survey (appendix 2). As volunteers were involved in mobile transect surveys, we developed a training manual for all survey participants (appendix 3).

It was possible that a pre-determined transect might become unavailable during the field season or between seasons. If this was the case, a slightly modified route could be recorded automatically with the GPS system without the transect surveyor. For safety reasons, we recommended a team of two for the mobile transect survey, unless the surveyor was extremely familiar with the road and the area without the need for direction.

STATIONARY POINT SURVEY

According to the NABat protocol, the stationary point survey involves 2-4 bat detectors placed for 4 consecutive nights at pre-selected sites within a grid cell in June or July. If both transect and stationary surveys will be conducted in a grid cell, the transect needs to be surveyed on nights when the stationary survey is happening. Stationary detectors run the entire night, from 15 minutes prior to sunset to 15 minutes after sunrise. The stationary sites should be selected to maximize the number and quality of recordings as well as the diversity of species. Clutter (dense vegetation) and large flat reflective surfaces (like pavement) should be avoided. When 4 sites are used, each site should be within a quadrat of the grid cell. If the grid cell is very homogenous, the number of detectors can be reduced to 2. The detector microphone can be mounted to a pole or a tripod or a tree and needs to be at least 1.5 m above the ground. Weatherproofing gear should be used.

When implementing this protocol in the field, we had to modify it for many logistic reasons. First, as volunteers were involved in transect surveys, it was not possible to schedule both surveys to be done within the same time window. Even for cells that were surveyed by the survey team, a slight mismatch happened on

rainy nights. Second, due to accessibility, stationary point survey sites might be outside of a grid cell (less than 500 m) and could not be placed in each quadrat of a grid cell. The number of stationary point survey sites within a grid cell was determined by both habitat variety and land accessibility. In general, we attempted to sample the following structures within a cell: an open field, a forest-field edge, a forest corridor, and a forest interior. When water features were available and accessible, we would incorporate the water body in one or some of the structures mentioned above. Additionally, if a grid cell contains a large proportion of human settlement, we would attempt to include a stationary point survey site in urban habitats. The stationary survey setup and timing followed the NABat protocol strictly.

We developed a data sheet to record covariates for the stationary point survey (appendix 2). Based on two field seasons, we realized that stationary point survey sites varied significantly between seasons. Specific sites within a grid cell might have to be selected every year due to accessibility changes, land management activities, and vegetation growth. Thus, we did not provide a guide for stationary point survey sites. We did summarize a guide on equipment used in the stationary point survey and how to choose and set up a stationary point survey sites (appendix 4). This guide was also used for guiding partners for equipment purchase and survey planning. More details are discussed in the later sections.

LANDS INVOLVED IN THE STATIONARY POINT SURVEYS

A total of 63 properties was involved in the stationary point surveys. Among them, 18 were privately owned properties. Due to privacy reason, the owner information and addresses of these properties are not included in this report. How to involve these private land owners in the future is discussed in the future implementation section. Owner's names are listed as those who assisted in the fieldwork in the acknowledgements.

Federal lands

- Alligoator River National Wildlife Refuge
- Blue Ridge Parkway
- Croatan National Forest
- Fort Bragg Military Base
- Nantahala National Forest
- Pisgah National Forest
- Uwharrie National Forest

Tribal lands

- Eastern Band of Cherokee Indian tribal lands

State lands

Game lands and other NCWRC properties

- Brinkleyville Game Land
- Brunswick Game Land
- Buffalo Cove Game Land
- Bullard and Branch Game Land
- Catawba Game Land (no longer leased)
- Chatham Game Land
- Chowan Swamp Game Land

- Cold Mountain Game Land
- Croatan National Forest Game Land
- Elk Knob Game Land
- Hyco Game Land
- Lower Roanoke River Wetlands Game Land
- Nantahala National Forest Game Land
- NCWRC Hertford Depot (not a game land)
- NCWRC Lennons Bridge Boat Ramp (not a game land)
- Neuse River Game Land
- Pee Dee River Game Land
- Pisgah National Forest Game Land
- Sandhills Game Land
- Shocco Creek Game Land
- Sutton Lake Game Land
- Upper Roanoke River Wetlands Game Land
- Uwharrie National Forest Game Land

State parks

- Carolina Beach State Park
- Chimney Rock State Park
- Crowders Mountain State Park
- Elk Knob State Park
- Eno River State Park
- Hanging Rock State Park
- Lake Norman State Park
- Lake Waccamaw State Park
- Lumber River State Park
- Mayo River State Park
- Medoc Mountain State Park

Other state properties

- NC Aquarium at Fort Fisher

County lands

- Mecklenburg County Property
- Wake County property

ACOUSTIC SPECIES IDENTIFICATION

The NABat protocol expects the individual surveyor or agency to process acoustic recordings and identify bats from recordings. It suggests using multiple automated identification programs and emphasizes the importance of manual verification of calls. However, no specific workflow for acoustic species identification was provided. Therefore, we developed the following workflow and summarized it into a manual (appendix 5).

1. Use AnaLook 4.1z (Chris Corben, www.hoarybat.com) to visually display and screen analyzable calls. Call quality criteria are:
    a. At least 3 pulses within 5 seconds
    b. Calls are not fragmentary
    c. Background noise does not overlap with calls in the frequency domain
2. Use two automated identification programs to assign species identification to each analyzable file. The programs are:
    a. Bat Call Identification (BCID) version 2.7c (C. Ryan Allen, www.batcallid.com)
    b. EchoClass v3.1 (Eric Britzke, US Army Engineer Research and Development Center, Vicksburg, MS)
3. Manually verify all transect recordings to assign identification to all files. Manually verify selected stationary recordings to obtain species presence/no detection information for each grid cell (combining all sites all nights for each season, see note below). The criteria used for manual species verification include:
    a. Assume no distribution range expansion of any species. Only species that are known present in a region (coastal, piedmont, mountains) based on common knowledge and capture records from NCWRC and other bat biologists will be considered for identification. For example, southeastern Myotis, *Myotis austroriparius*, would be present in the coastal region but not in the mountain region. Whereas Indiana bats, *Myotis sodalis*, should only be present in the mountain region. The list of species considered (the species abbreviations used in the following sections):
        - CORA - *Corynorhinus rafinesquii*, Rafinesque's big-eared bat
        - EPFU – *Eptesicus fuscus*, big brown bat
        - LABO – *Lasiurus borealis*, red bat
        - LACI – *Lasiurus cinereus*, hoary bat
        - LANO – *Lasionycteris noctivagans*, silver haired bat
        - MY spp. – bats from genus *Myotis* (for transect data only)
        - MYAU – *Myotis austroriparius*, southeastern myotis (coastal/piedmont only)
        - MYGR – *Myotis grisescens*, gray bat (piedmont/mountain only)
        - MYLE – *Myotis leibii*, eastern small-footed myotis (mountain only)
        - MYLU – *Myotis lucifugus*, little brown bat
        - MYSE – *Myotis septentrionalis*, northern long eared bat
        - MYSO – *Myotis sodalis*, Indiana bat (mountain only)
        - NYHU – *Nycticeius humeralis*, evening bat
        - PESU – *Perimyotis subflavus*, tricolored bat
        - TABR – *Tadarida brasiliensis*, Mexican free-tailed bat
    b. For all Myotis species, files that were identified as the same species by either programs will be examined. These recordings will also be compared across the entire state. Due to the quality of recordings, *Myotis* species from transect data will only be identified to genus level as *Myotis* spp. Multiple verified identifications (usually at least 3 – 5 call files) are needed to confirm the presence of a certain Myotis species in a grid cell on a given night.
    c. For *Corynorhinus rafinesquii*, all program identified files will be compared with reference calls published by (Loeb and Britzke 2010).

d. For *Tadarida brasiliensis*, all program identified files will be compared with calls collected for work conducted by (Li and Wilkins 2014, 2015).
e. For other species, files that were identified as the same species by both two programs will be examined. One or two verified identifications are needed to confirm the presence of a certain species in a grid cell.

Due to the large number of acoustic files collected in each field season, manually vetting of files needs to be limited to selected files. For the longterm, broad scale analysis of bat distribution and population trends, within grid cell variations and within season variations are not high priority for NABat data analysis protocols. Thus, we combined all stationary point survey sites together for each grid cell.

AUTOMATED IDENTIFICATION PROGRAM SETTINGS

For the general presence/no detection identification analysis, we used the following settings. When we used acoustic data to inference bat activity level, we increased the BCID setting min discriminant probability for species ID to 0.5:

BCID settings

Filter setting:

- Minimum percentage of pulses for species ID 0
- Minimum percentage of pulses for group ID 0
- Min discriminant probability for species ID 0
- S1(OPS) Minimum -9999, Maximum 9999
- Dur (ms) Minimum 1, Maximum 20
- Minimum Number of Calls Present 5 within (sec) 15

Species setting:

For grid cells in different regions, we use the potential species within the region. Thus, we did not include MYAU – *Myotis austroriparius*, southeastern myotis for the mountain region; MYLE – *Myotis leibii*, eastern small-footed myotis and MYSO – *Myotis sodalis*, Indiana bat for the piedmont region; and MYGR – *Myotis grisescens*, gray bat, MYLE – *Myotis leibii*, eastern small-footed myotis and MYSO – *Myotis sodalis*, Indiana bat for the coastal region.

EchoClass settings

We used Species Set 1, which is recommended for North Carolina as the candidate species. No other setting options is available for this program. Regional species-specific option is not available for this program.

DATA MANAGEMENT AND ANALYSIS

Originally NABat suggested the Bat Population Database from U.S. Geological Survey as the platform for data management. However, due to technical difficulties, the online national database will not be available for upload data until the end of 2017. Therefore, we developed a guide for both acoustic species identification and data management to store data locally (appendix 5). The guide is suitable for managing data locally and provides a data frame structure to local storage. Currently our data has been stored in three different external hard drives and will be upload to the national database when it becomes available.

NABat suggests that both presence/no detection and relative activity level can be the response variables from the acoustic data collected in this project. It also notes that pilot data will be needed to test model assumptions before a specific statistical test is used. Currently we are collaborating with NABat national statisticians to identify the suitable statistics approaches for NABat data. Therefore, in this report we present our results as the presence/no detection information and the relative activity level information.

RESULTS AND INTERPRETATION

GIRD CELLS SAMPLED

We visited a total of 37 grid cells in 2015 and 56 grid cells in 2016. All grid cells sampled in 2015 were revisited except for one (grid cell 9). Grid cell 9 was sampled by transect only in 2015. Due to road changes, the cell was not sampled in 2016. Additionally, the following cells sampled in 2015 had to be sampled using a different method for the following reasons.

- Grid cell 2 was sampled by both stationary and transect in 2015. Due to accessibility change (Fort Bragg military base), the cell was only sampled by stationary survey in 2016.
- Grid cell 54 was sampled by stationary only in 2015. Due to accessibility change (Game Land no longer leased), the cell was sampled by transect survey in2016.
- Grid cell 29 was sampled by transect only in 2015. Stationary sites were added in 2016.
- Grid cell 84 was sampled by transect only in 2015. Stationary sites were added in 2016.
- Grid cell 65 was sampled by stationary sites only in 2015. A transect was added in 2016.

In 2015, 9 transects were surveyed by project volunteers. In 2016, 17 transects were surveyed by project volunteers. Additionally, one grid cell (cell 105) was sampled by USFWS wildlife biologists using USFWS equipment. Some long-term partnerships were developed for project participants. More details are discussed in the future implantation section. Project participants who assisted in the fieldwork are listed in the acknowledgements.

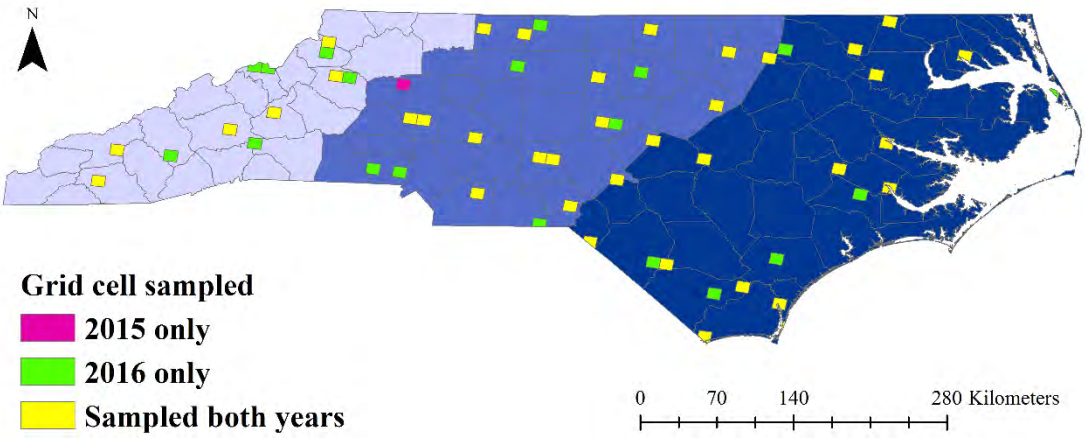


Figure 1 NABat grid cells sampled in 2015 and 2016 by either mobile transect survey or stationary point survey or both

GRID CELLS BY GEOGRAPHICAL REGIONS

Geographically North Carolina falls into three regions: the Appalachian Mountains, the Piedmont Plateau, and the Coastal Plain region (Figure. 1, indicated by different shades). Grid cells visited by region and by year are listed below:

Table 1 Grid cells visited by region in 2015 and 2016

Geographical region	2015	2016
Appalachian Mountains	6	12
Piedmont Plateau	15.5*	21
Coastal Plain	15.5*	23

*Grid Cell 101 in Franklin County and Nash County covers both the Piedmont Plateau and the Eastern or Coastal Plain regions. Two stationary sites in each region (a total of 4) were set during the survey, resulting in a mixed grid. In 2016 only stationary sites in the coastal region were sampled

GRID CELLS BY SAMPLING METHODS

Due to logistic (mainly labor and effort required for stationary site set up) reasons and volunteer participation, different survey methods were used in different grid cells. The number of grid cells sampled by survey method is listed below:

Table 2 Grid cells sampled by survey method in 2015 and 2016

Survey methods	2015	2016
Driving transect and stationary	17	23
Driving transect only	15	16
Stationary only	5	17

In 2016, a different survey travel strategy was implemented than in 2015. This caused the significant increase of number of cells surveyed. More details are discussed in the future implementation section.

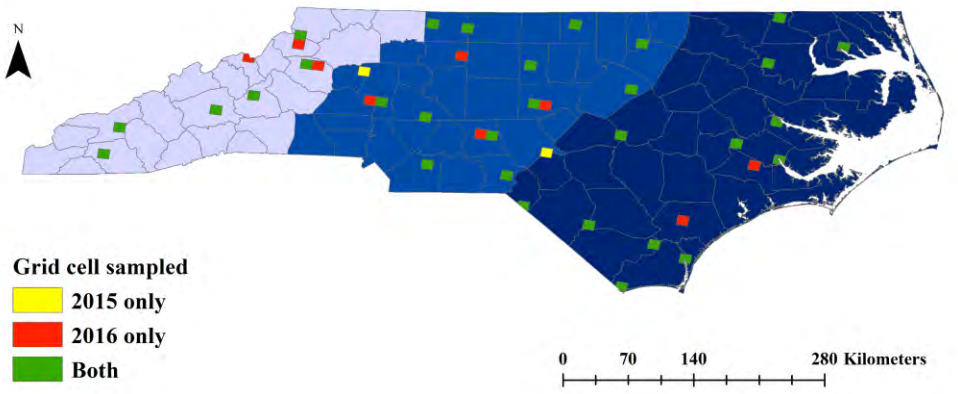


Figure 2 NABat grid cells sampled in 2015 and 2016 by mobile transect survey

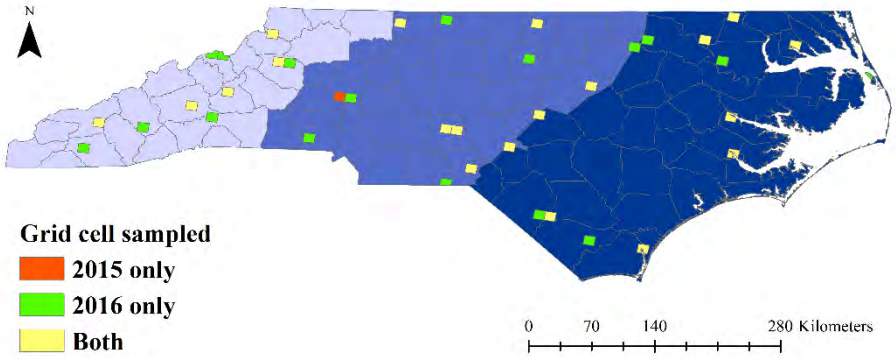


Figure 3 NABat grid cells sampled in 2015 and 2016 by stationary point survey

TOTAL NUMBERS OF ACOUSTIC FILES COLLECTED

The numbers of acoustic files collected in both years by sampling method and recording quality are listed below. The high numbers of files collected affected acoustic species identification strategies.

Table 3 Number of acoustic files collected in 2015 and 2016 (listed as 2015 number/2016 number)

Survey method	Total acoustic files	Files with bat calls	Files with analyzable bat calls
Driving transect	5104/12036	3322/6394	2145/4739
Stationary	98313/215451	62075/160485	33592/105349

MOBILE TRANSECT SURVEY RESULTS

MOBILE TRANSECT SURVEY PRESENCE/NO DETECTION TABLES BY YEAR

For mobile transect survey data, we manually vetted all bat calls with at least 5 pulses to species. The presence/no detection tables by year are presented here. P stands for presence; N/D stands for no detection.

Table 4 Species detected in mobile transect surveys in 2015

	Cell	EPFU	LABO	LACI	LANO	*Myspp*	NYHU	PESU	TABR
	cell-0	N/D	P	N/D	N/D	P	P	P	P
	cell-1	P	P	N/D	P	N/D	P	N/D	P
	cell-2	P	P	P	P	P	P	P	N/D
	cell-3	N/D	P	N/D	P	P	P	P	P
	cell-5	P	P	P	P	N/D	P	P	P
	cell-6	P	P	P	P	N/D	P	P	P
	cell-7	N/D	P	N/D	N/D	P	P	P	N/D
	cell-9	P	P	N/D	P	P	P	P	P
	cell-12	P	P	N/D	P	N/D	P	P	P
	cell-13	N/D	P	N/D	N/D	N/D	P	P	N/D
	cell-14	N/D	P	P	P	N/D	P	P	P
	cell-15	N/D	P	P	P	N/D	P	P	P
	cell-18	P	P	N/D	P	P	P	P	P
	cell-19	P	P	P	N/D	P	P	P	N/D
	cell-20	N/D	P	P	P	N/D	P	P	P
	cell-21	P	P	P	P	P	P	P	P
	cell-22	N/D	P	P	N/D	N/D	P	P	N/D
	cell-23	N/D	P	P	P	P	P	P	P
	cell-24	N/D	P	P	N/D	P	P	P	N/D
	cell-26	P	P	N/D	P	P	P	P	N/D
	cell-27	P	P	N/D	P	P	P	P	P
	cell-29	N/D	P	N/D	N/D	P	P	P	P
	cell-30	P	P	N/D	P	P	P	P	P
	cell-31	P	P	N/D	P	P	P	P	P
	cell-32	P	P	N/D	P	P	P	P	P
	cell-33	P	P	P	P	P	P	P	P
	cell-37	N/D	P	N/D	P	P	P	P	P
	cell-39	P	P	P	P	P	P	P	P
	cell-42	P	P	P	P	P	P	P	P
	cell-44	P	P	P	P	P	P	N/D	P
	cell-47	P	P	N/D	P	P	P	P	P
	cell-84	P	P	P	P	P	P	P	N/D

Table 5 Species detected in mobile transect surveys in 2016.

Cell	EPFU	LABO	LACI	LANO	*Myspp*	NYHU	PESU	TABR
cell-0	N/D	P	P	P	N/D	P	N/D	N/D
cell-1	N/D	P	N/D	P	P	P	P	N/D
cell-3	N/D	P	N/D	P	N/D	P	P	N/D
cell-5	P	P	P	P	P	P	P	P
cell-6	P	P	P	P	P	P	P	P
cell-7	N/D	P	N/D	N/D	P	P	N/D	P
cell-12	P	P	P	P	P	P	P	P
cell-13	N/D	P	N/D	N/D	P	P	P	N/D
cell-14	N/D	P	N/D	N/D	N/D	P	P	N/D
cell-15	P	P	P	P	N/D	P	P	P
cell-18	P	P	P	N/D	N/D	P	P	P
cell-19	P	P	N/D	P	N/D	P	P	P
cell-20	P	P	N/D	P	N/D	P	P	P
cell-21	P	P	N/D	P	P	P	P	P
cell-22	P	P	N/D	P	N/D	P	P	P
cell-23	P	P	N/D	P	N/D	P	P	P
cell-24	N/D	P	N/D	P	N/D	P	N/D	P
cell-26	P	P	N/D	P	P	P	P	N/D
cell-27	N/D	P	N/D	N/D	P	P	P	N/D
cell-29	N/D	P	N/D	N/D	P	P	P	P
cell-30	P	P	P	P	N/D	P	N/D	P
cell-31	P	P	N/D	N/D	N/D	P	P	N/D
cell-32	P	P	P	N/D	P	P	N/D	P
cell-33	P	P	N/D	P	P	P	P	P
cell-37	N/D	P	P	P	N/D	P	P	P
cell-39	P	P	P	P	N/D	P	P	N/D
cell-42	P	P	P	P	P	P	P	P
cell-44	P	P	P	P	P	P	P	P
cell-47	P	P	N/D	P	P	P	P	N/D
cell-52	P	P	P	P	P	P	P	N/D
cell-53	P	P	N/D	P	P	P	P	N/D
cell-54	P	P	N/D	P	P	P	P	P
cell-59	P	P	N/D	P	P	P	P	P
cell-65	P	P	N/D	P	P	P	P	P
cell-66	P	P	N/D	P	N/D	P	P	P
cell-84	P	P	P	P	P	P	P	P
cell-90	P	P	P	P	N/D	N/D	P	P
cell-94	P	P	N/D	P	P	P	P	N/D
cell-100	P	P	P	P	P	P	P	P

MOBILE TRANSECT SURVEY BAT ACTIVITY DENSITY MAPS BY SPECIES

Because all transect recordings have been identified to species, it is possible to calculate species-specific activity level (number of calls/transect or recording hour). The design of NABat driving transect protocol (survey speed and transect spatial structure) allows bat activity level obtained from this survey as an indirect index of abundance (Loeb et al. 2015). Below we generated bat relative abundance maps (Figure 4 - 10) using ArcGIS interpolation function. Specifically, we used inverse distance weighted (IDW) interpolation method to determine bat relative abundance for areas that were not sample. IDW uses a linearly weighted combination of a set of sample points. The weight is a function of inverse distance. The surface being interpolated should be that of a location-dependent variable. We combined two years or data for grid cells that were sampled in both years and calculated the average. It must be noteed that the map quality was limited by the number of grid cells sampled. It can be interpreted as a broad scale distribution of each species. It should not be used to predict species presence in any local site or estimate the population size at any scale. We did not generate a map for the *Myotis* genus group. Instead, we discuss the unique recordings for species collected through the mobile transect survey. In the maps below, the darker shade of color indicates higher bat activity. Major cities in NC are listed as references.

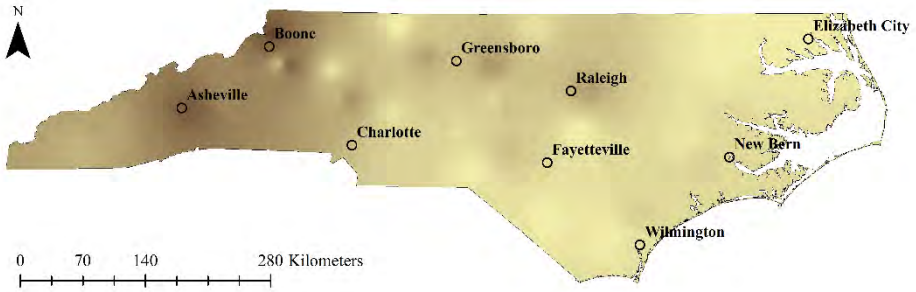


Figure 4 EPFU relative abundance density plot using IDW interpolation method based on mobile transect survey data

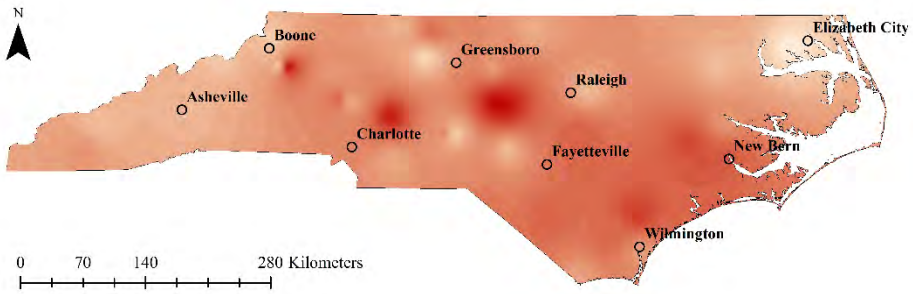


Figure 5 LABO relative abundance density plot using IDW interpolation method based on mobile transect survey data

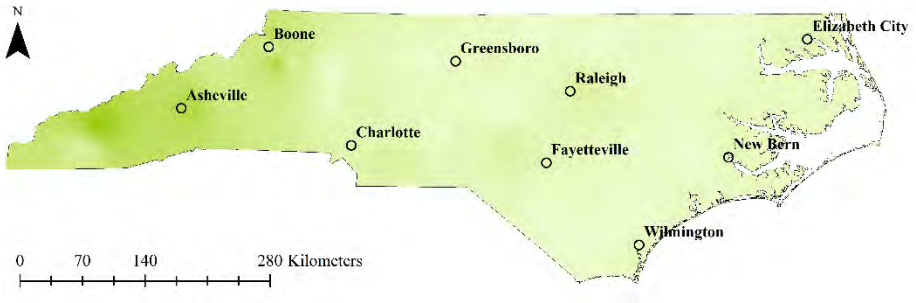


Figure 6 LACI relative abundance density plot using IDW interpolation method based on mobile transect survey data

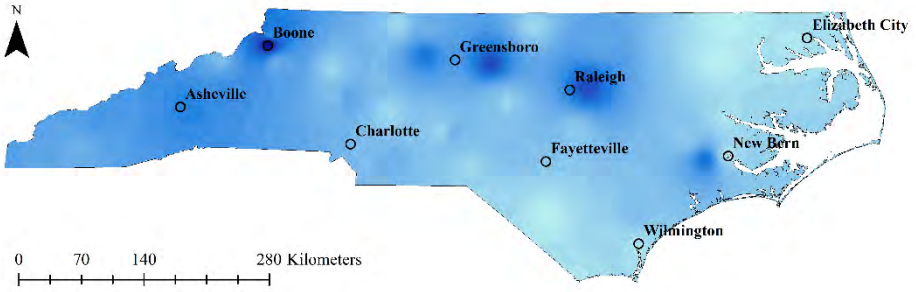


Figure 7 LANO relative abundance density plot using IDW interpolation method based on mobile transect survey data

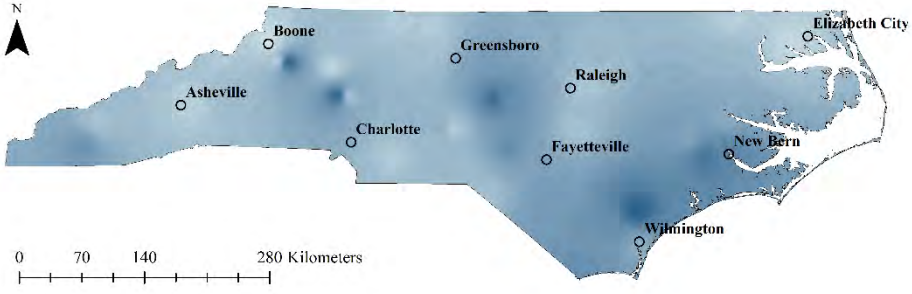


Figure 8 NYHU relative abundance density plot using IDW interpolation method based on mobile transect survey data

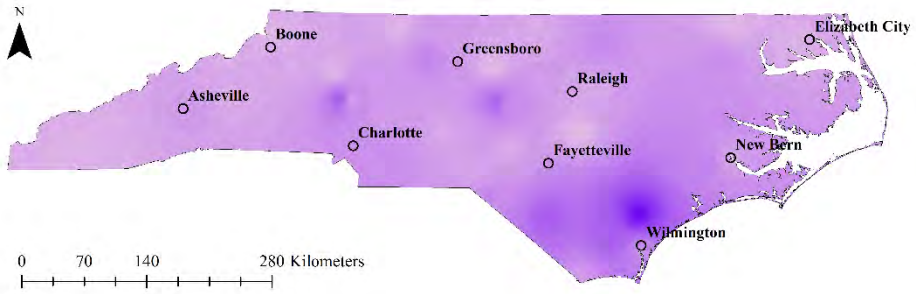


Figure 9 PESU relative abundance density plot using IDW interpolation method based on mobile transect survey data

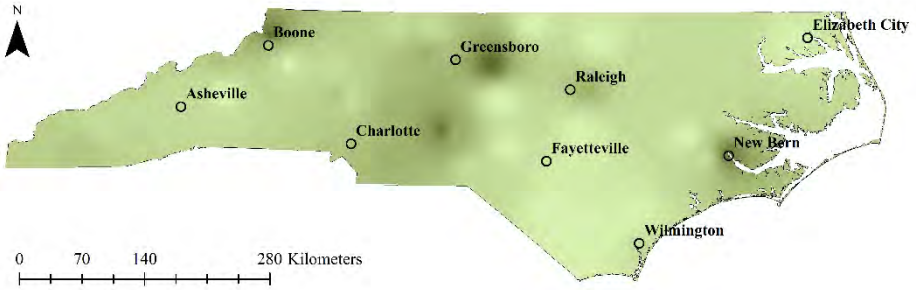


Figure 10 TABR relative abundance density plot using IDW interpolation method based on mobile transect survey data

UNIQUE SPECIES RECORDINGS

In general, mobile transect survey recordings do not include high numbers of good quality *Myotis* species calls. This is because 1) *Myotis* species are relatively rare, therefore less likely to be encountered and 2) many *Myotis* species prefer forest interior over other habitats and roads used for the mobile transect survey usually do not go through forest interiors. However, there were still some high quality *Myotis* recording obtained through the mobile transect survey in both years. For example, in grid cell 42 near Asheville in both years the mobile transect surveys recorded high quality MYGR calls (Figure 11). Some permanent summer MYGR roosts have been located. For MYLU, in both years we recorded high numbers of MYLU calls in grid cells between Charlotte and Uwharrie National Forest. These areas should be investigated through mist-netting if MYLU needs to be further studied.

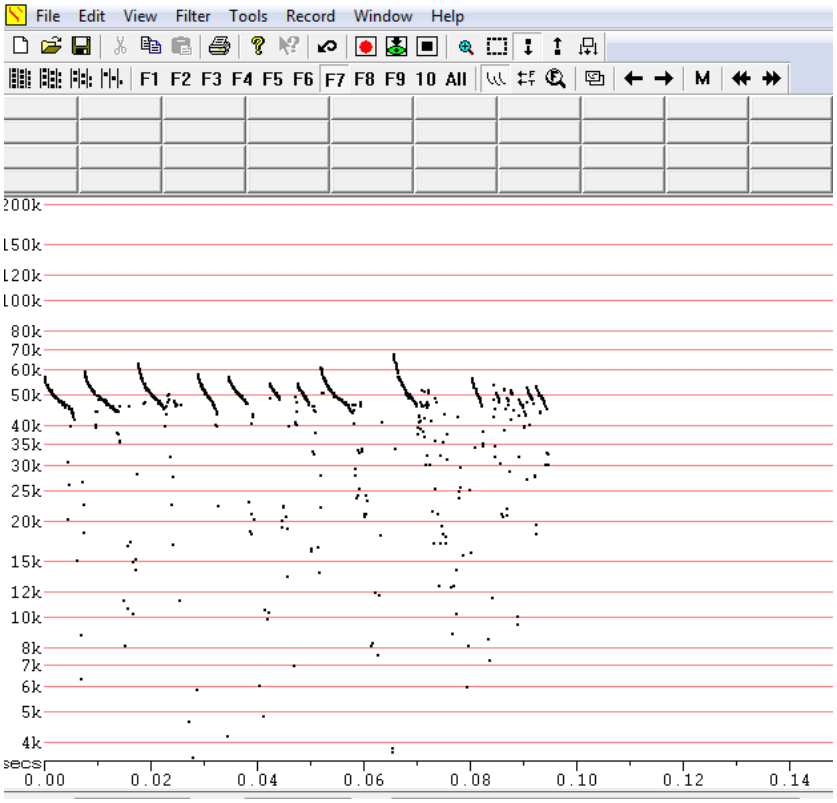


Figure 11 Example of MYGR calls collected via mobile transect survey in gird cell 42 near Asheville in 2015

STATIONARY POINT SURVEY RESULTS

STATIONARY POINT SURVEY PRESENCE/NO DETECTION TABLES BY YEAR

For stationary point survey data, we manually vetted selected bat calls, with at least 5 pulses, to species. For each grid cell, we selected call files for each species with all sites and all nights combined. The presence/no detection tables by year are presented here. P stands for presence; N/D stands for no detection.

Table 6 Species detected in stationary point surveys in 2015

cell	Cell	CORA	EPFU	LANO	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYHU	PESU	TABR
1	cell-1	N/D	N/D	N/D	P	N/D	N/D	N/D	N/D	P	N/D	N/D	P	N/D	N/D
2	cell-2	N/D	P	P	P	N/D	P	N/D	N/D	P	N/D	N/D	P	P	P
5	cell-5	P	P	P	P	P	N/D	P	N/D	P	N/D	N/D	P	P	N/D
6	cell-6	P	P	P	P	P	N/D	P	P	P	P	P	P	P	P
7	cell-7	P	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
10	cell-10	N/D	P	P	P	N/D	P	N/D	N/D	P	N/D	N/D	P	P	P
12	cell-12	N/D	P	P	P	P	N/D	N/D	N/D	N/D	N/D	N/D	N/D	P	P
13	cell-13	N/D	P	P	P	N/D	N/D	N/D	N/D	P	P	N/D	P	P	N/D
14	cell-14	N/D	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
18	cell-18	P	P	P	P	P	N/D	N/D	N/D	P	N/D	N/D	P	P	P
19	cell-19	N/D	P	P	P	N/D	P	N/D	N/D	N/D	N/D	N/D	P	P	P
20	cell-20	N/D	P	P	P	N/D	P	N/D	N/D	N/D	N/D	N/D	P	P	N/D
22	cell-22	N/D	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	N/D
24	cell-24	N/D	P	P	P	P	P	N/D	N/D	P	N/D	N/D	P	P	P
37	cell-37	N/D	P	P	P	P	N/D	N/D	N/D	P	N/D	N/D	P	P	P
39	cell-39	P	P	P	P	N/D	N/D	P	N/D	P	N/D	N/D	P	N/D	N/D
40	cell-40	P	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	N/D
42	cell-42	N/D	P	P	P	P	N/D	P	N/D	P	N/D	P	P	P	P
44	cell-44	N/D	P	P	P	N/D	N/D	P	N/D	P	N/D	P	P	N/D	P
54	cell-54	N/D	P	P	P	N/D	P	P	N/D	P	N/D	N/D	P	P	P
65	cell-65	N/D	P	P	P	P	N/D	P	N/D	P	P	N/D	P	P	P
108	cell-108	N/D	P	P	P	P	P	N/D	N/D	N/D	N/D	N/D	P	P	N/D

Table 7 Species detected in stationary point surveys in 2016

	Cell	CORA	EPFU	LANO	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYHU	PESU	TABR
	cell-1	N/D	P	P	P	N/D	P	N/D	N/D	P	N/D	N/D	P	P	P
	cell-5	N/D	P	P	P	P	N/D	P	N/D	P	N/D	P	P	P	N/D
	cell-6	P	P	P	P	P	N/D	P	N/D	P	N/D	P	P	P	P
	cell-7	N/D	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
	cell-10	N/D	P	P	P	N/D	P	N/D	N/D	P	N/D	N/D	P	P	P
	cell-12	N/D	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
	cell-13	N/D	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	P
	cell-14	N/D	P	P	P	P	N/D	N/D	N/D	P	P	N/D	P	P	P
	cell-18	P	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
	cell-19	N/D	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	P
	cell-20	N/D	P	P	P	P	N/D	N/D	N/D	P	N/D	N/D	P	P	P
	cell-21	N/D	P	P	P	N/D	N/D	P	N/D	N/D	N/D	N/D	P	P	P
	cell-22	P	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	N/D
	cell-24	N/D	P	P	P	P	N/D	P	N/D	N/D	N/D	N/D	P	P	P
	cell-29	N/D	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	N/D
	cell-34	P	P	P	P	N/D	P	N/D	N/D	P	N/D	N/D	P	P	P
	cell-37	N/D	P	P	P	P	N/D	N/D	N/D	P	N/D	N/D	P	P	P
	cell-38	N/D	P	P	P	N/D	N/D	N/D	N/D	N/D	N/D	N/D	P	P	N/D
	cell-39	P	P	P	P	N/D	N/D	P	N/D	P	P	P	P	P	P
	cell-40	N/D	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
	cell-42	P	P	P	P	P	N/D	P	N/D	P	N/D	P	P	P	P
	cell-44	N/D	P	P	P	P	N/D	P	N/D	P	N/D	P	P	N/D	P
	cell-46	N/D	P	P	P	N/D	P	N/D	N/D	N/D	N/D	N/D	P	P	P
	cell-48	P	P	P	P	N/D	P	N/D	N/D	P	P	N/D	P	P	P
	cell-52	P	P	P	P	P	N/D	P	N/D	P	P	P	P	P	N/D
	cell-58	N/D	P	P	P	P	P	N/D	N/D	P	N/D	N/D	P	P	P
	cell-62	N/D	P	P	P	N/D	N/D	N/D	N/D	P	N/D	N/D	P	P	P
	cell-65	N/D	P	P	P	P	N/D	N/D	N/D	P	P	N/D	P	P	N/D
	cell-66	N/D	P	P	P	N/D	N/D	N/D	N/D	N/D	N/D	N/D	P	P	P
	cell-74	P	P	P	P	N/D	N/D	P	N/D	P	P	P	P	P	N/D
	cell-75	N/D	P	P	P	N/D	N/D	N/D	N/D	N/D	P	N/D	P	P	P
	cell-78	N/D	P	P	P	P	N/D	N/D	N/D	P	N/D	N/D	P	P	P
	cell-79	N/D	P	P	P	N/D	N/D	N/D	N/D	P	N/D	N/D	P	P	N/D
	cell-81	N/D	P	P	P	P	P	N/D	N/D	P	P	N/D	P	P	P
	cell-83	N/D	P	P	P	N/D	N/D	N/D	P	P	P	N/D	P	P	N/D
	cell-84	P	P	P	P	N/D	N/D	N/D	N/D	P	N/D	P	P	P	N/D
	cell-95	N/D	P	P	P	P	N/D	P	N/D	P	N/D	P	P	P	P
	cell-100	N/D	P	P	P	P	N/D	P	N/D	P	P	P	P	P	N/D
	cell-105	N/D	P	P	P	N/D	N/D	N/D	N/D	N/D	N/D	N/D	P	P	N/D
	cell-108	N/D	P	P	P	N/D	P	N/D	N/D	P	N/D	N/D	P	P	P

STATIONARY POINT SURVEY BAT ACTIVITY DENSITY MAPS BY SPECIES

We conducted the same IDW interpolation for stationary point survey data (Figure 12 -23). However, it must be noted that due to the design of stationary point surveys bat activity level cannot be interpreted as relative abundance. It can only indicate how active bats were in each grid cell during the survey season. It must also be noted that due to the large amount of call files, we were not able to manually vet all files to generate bat activity level used in interpolation. Instead, we selected a very conservative setting in the automated identification program BCID. We only included call files with at least 0.50 discriminant probability for each individual call file. For each grid cell, we combined all sites and all nights to calculate the average for each grid cell. We also combined two years or data for grid cells that were sampled in both years and calculated the average. These maps can only be used to indicate how active bats distributed at a broad scale. It should not be used to predict species presence in any local site or estimate the population size at any scale. For certain species with limited distribution in NC, we only generated regional maps. In the maps below, the darker shade of color indicates higher level of bat activity. Major cities in NC are listed as references. Due to the program limitation, we did not include TABR in to the mapping section. CORA maps are listed in a separated section for further discussion.

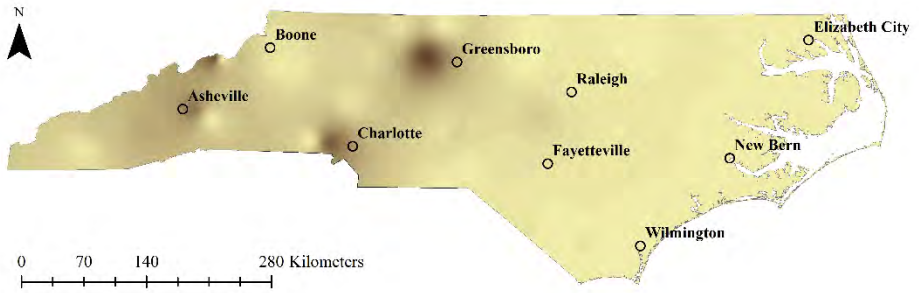


Figure 12 EPFU activity level density plot using IDW interpolation method based on stationary point survey data

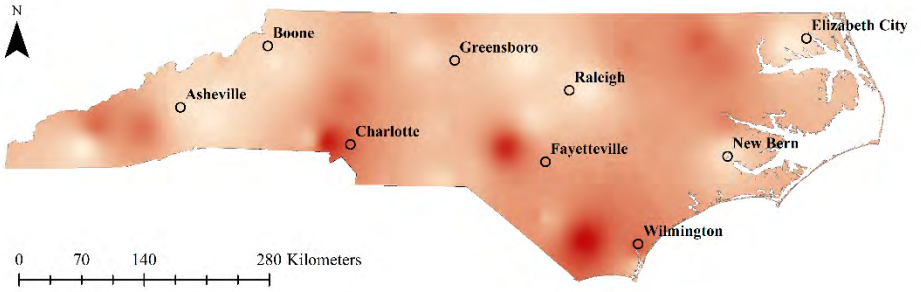


Figure 13 LABO activity level density plot using IDW interpolation method based on stationary point survey data

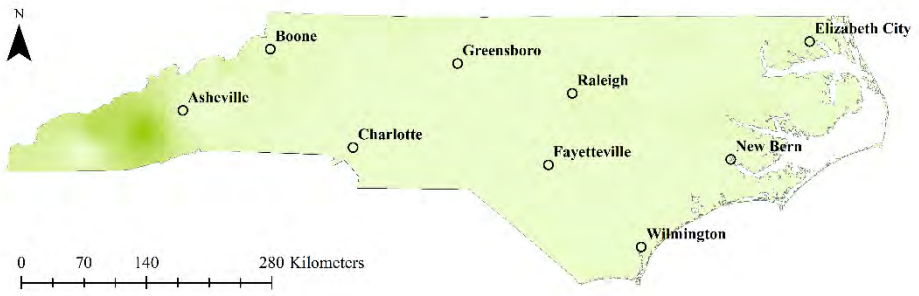


Figure 14 LACI activity level density plot using IDW interpolation method based on stationary point survey data

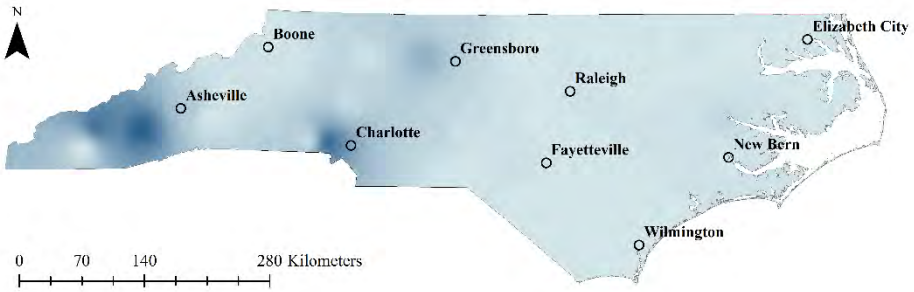


Figure 15 LANO activity level density plot using IDW interpolation method based on stationary point survey data

Figure 16 MYAU activity level density plot using IDW interpolation method based on stationary point survey data

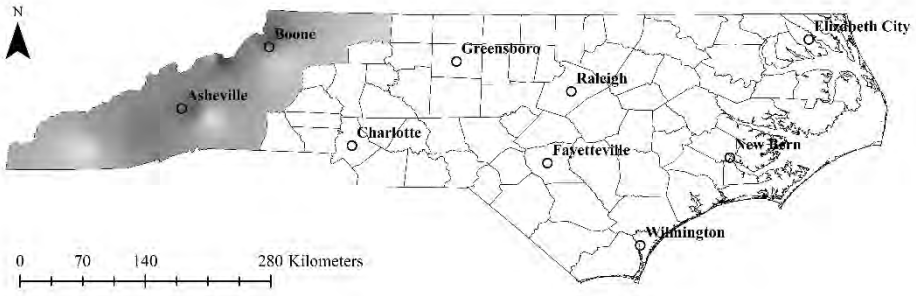


Figure 17 MYGR activity level density plot using IDW interpolation method based on stationary point survey data

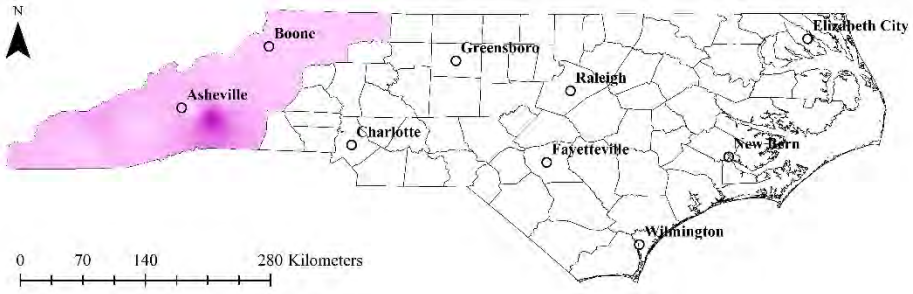


Figure 18 MYLE activity level density plot using IDW interpolation method based on stationary point survey data

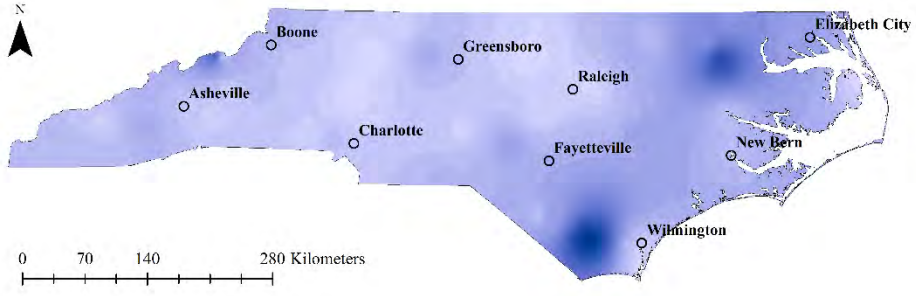


Figure 19 MYLU activity level density plot using IDW interpolation method based on stationary point survey data

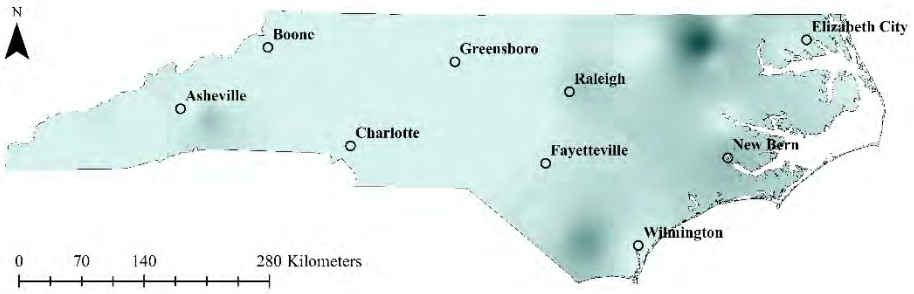


Figure 20 MYSE activity level density plot using IDW interpolation method based on stationary point survey data

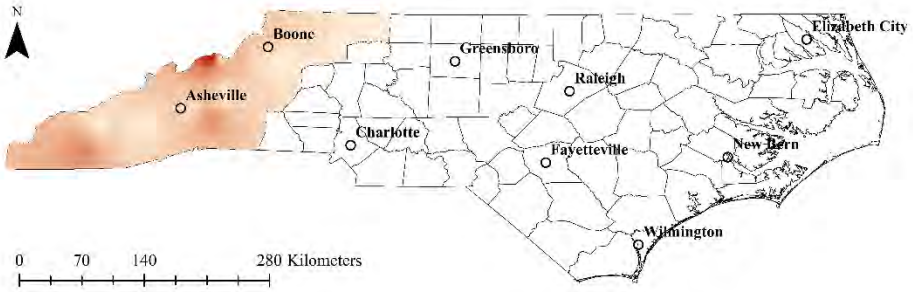


Figure 21 MYSO activity level density plot using IDW interpolation method based on stationary point survey data

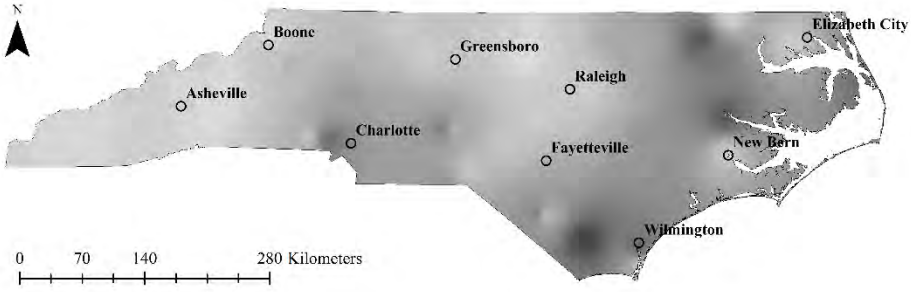


Figure 22 NYHU activity level density plot using IDW interpolation method based on stationary point survey data

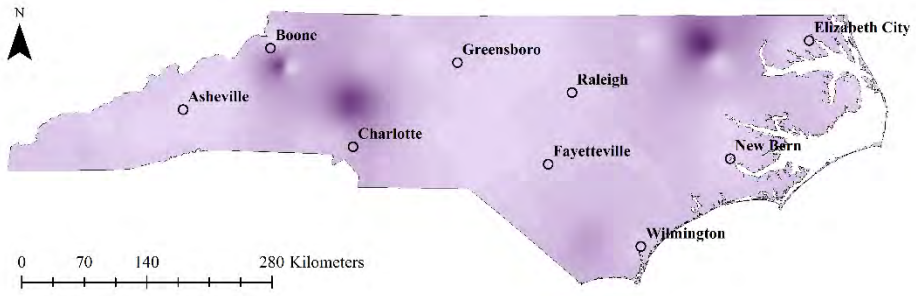


Figure 23 PESU activity level density plot using IDW interpolation method based on stationary point survey data

UNIQUE SPECIES RECORDINGS

It was suggested that CORA might not be adequately surveyed acoustically. However, our field experience showed that CORA could be successfully recorded by placing the stationary point survey sites strategically. We recorded CORA at multiple sites. Due to its unique echolocation calls, its identification contains high confidence (Figure 24). In grid cell 6 on the Eastern Band of Cherokee Indian property, we recorded CORA in both years, which was unknown to the area. In fall 2016, tribal biologists located a CORA roost near one of the NABat stationary point survey sites. We used the IDW interpolation methods to generate two separate CORA maps to reflect its mountain and coast populations (Figure 25, 26).

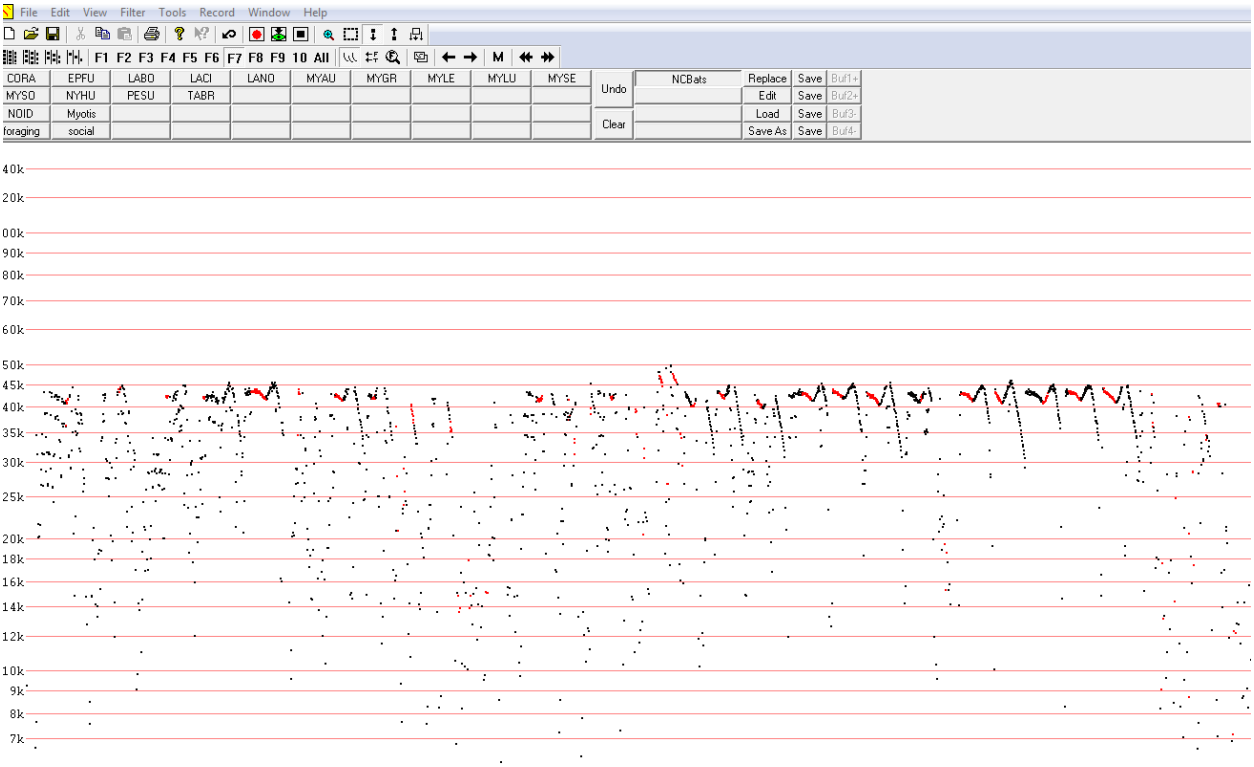


Figure 24 Example of CORA calls collected via stationary point survey in gird cell 48 near Lake Waccamaw in 2016

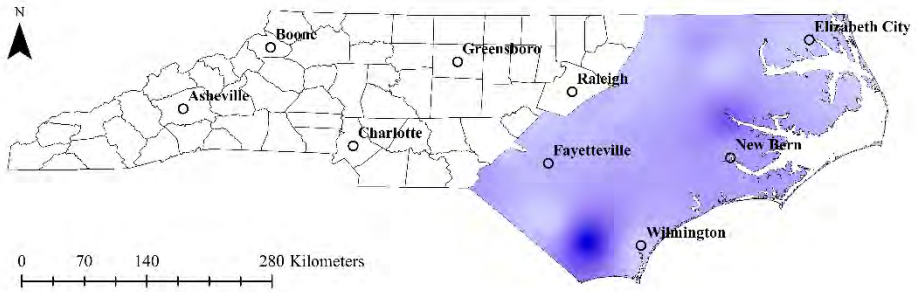


Figure 25 CORA activity level density plot using IDW interpolation method based on stationary point survey data in the coast region

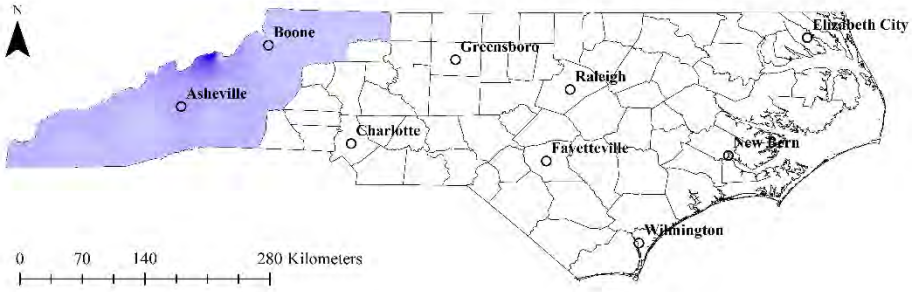


Figure 26 CORA activity level density plot using IDW interpolation method based on stationary point survey data in the mountain region

INTERPRETING THE ACTIVITY DENSITY MAPS

It is important to acknowledge that activity density maps generated by the mobile transect survey and by the stationary point survey cannot be interpreted in the same manner. The mobile transect survey activity density maps can loosely translate into the relative abundance of each species based on the survey design. The stationary point survey activity density maps only show the activity level. However, both types of maps for each species still show similar patterns. For example, both types of maps suggest a higher density of activity of EPFU in the mountain and piedmont regions than the coast region. LACI in both maps concentrates in the mountain region. These are generally consistent with NCWRC mist netting results in the past two years. In both types of maps NYHU shows higher activity density in the coast region than the mountain region. This is also consistent with mist netting records and the common knowledge of local bat biologists. PESU shows higher activity densities near water sources such as near Lake Norman, Lake Jordan, Cape Fear River, Chowan River. This is consistent with this species being a aquaticzone specialist (Kalcounis-Rueppell et al. 2007).

We used a very conservative setting to automatically identify bats for the stationary point survey data. The activity density maps generated by the stationary point survey data shows patterns consistent with current knowledge. MYSE is the only federally listed species with a statewide range in NC. The pattern of highest activity density in the coast region is consistent with local efforts of mist netting and current literature (Grider et al. 2016). This may suggest the coast potentially could function as a refuge for this species. Both MYGR and MYSO maps show the patterns that these species distribute in NC along the Appalachian Mountain with high activity density near the NC and TN border. The recent MYGR summer roosts in NC is also indicated by the higher activity density in the region.

The limitation of IDW interpolation mainly includes 1) no consideration of habitats, 2) no consideration of annual variation, and 3) map quality limited by the number of samples. Therefore, neither types of maps should be used for prediction purposes. In the future, habitat information can be added to produce models and maps for prediction. The current sample size and duration of data make it not suitable to generate annually different maps for estimating trends. As the survey is being continued over years, in the future this method can be used to investigate population trends for bats. The sample size is also the limiting factor to estimating population size. As the dataset grows, the mobile transect survey data can potentially be used to estimate population size.

IMPLEMENTATION OF NABAT

INFORMING CONSERVATION AND MANAGEMENT ACTIONS

One goal of implementing NABat is to establish long-term baseline bat monitoring to provide information for conservation and management actions. Through the pilot two years of surveys, our data suggests NABat is suitable to monitor most species of conservation concerns in the state wildlife action plan. For example, the activity density maps generated from different datasets show similar patterns for species distribution and are consistent with current knowledge. One direct outcome of the monitoring is to guide local surveys, such as mist-netting or roosts/hibernacula surveys. There have been incidences described above that roosts have been found near acoustic survey sites. We have made suggestions of the 2017 mist netting locations to the NCWRC biologists. The similar information also has led other agencies to mist netting plans. Another way to inform conservation and management through NABat is providing large scale guides on land management and acquisition. As data accumulate over time, the activity density maps and possibly habitat maps will become more mature and accurate. These maps can suggest if and where additional conservation lands should be.

FORMING MONITORING AND CONSERVATION PARTNERSHIPS

NABat protocol based surveys emphasize the broad spatial scale. The GRTS sample design can be applied to any size or level of jurisdiction. Therefore, forming multiple agencies, multiple stakeholders monitoring and conservation partnerships is a major component of long-term implementation of NABat. During our pilot work, many agencies and organizations expressed interest and needs for monitoring bats for different purposes. For example, both the Eastern Band of Cherokee Indian and Mecklenburg County have a wildlife inventory need. Many universities and science education organizations have teaching or outreach needs. Each partner can potentially contribute bat data at the local scale by adopting the NABat protocols. In this partnership system, more sites can be included for sampling and improve the quality of baseline bat data. We suggest a centralized coordinator to facilitate the partnership and incorporate the data for further analysis.

ADOPTING DIFFERENT SURVEY PROTOCOLS

Two different survey protocols increase the flexibility of participation. The monetary investment differs between these two methods. By choosing different bat detectors, equipment for the mobile transect survey can cost less than $500 whereas over $5000 is the least amount of investment needed for the stationary point survey. Even though both methods can provide informative data, more studies are needed to compare them systematically.

ADOPTING MOBILE TRANSECT SURVEY

Iit is easier for less experienced participants to adopt the mobile transect survey. There is no access issue related to the mobile transects. In 2015 we invited mostly citizen scientists for the mobile transect survey. In 2016 we invited mostly professional wildlife biologists. Overall we received higher quality data in 2016 than in 2015. The major issues for citizen scientists was the lack of scientific knowledge or experience for field data collection. For all non-biologists involved in 2015, we gave each one over 4 hours of training. Most participants had no issue using the detector. However, issues arose in collecting metadata, transferring data, or diagnosing common equipment errors. One example was that one citizen scientist had a loose GPS cable connection and did not collect GPS information through the season. However, it was not discovered until the data was sent back for archiving. Additionally, NABat suggests that for long-term monitoring consistence, each transect should be driven during the same time of each year over many years. This is an inherent issue for any volunteer based project. Very few volunteers can commit to a specific time window years in advance. It generally took more time for citizen scientists to complete a grid cell than professional biologists, which might be a commitment issue as well. In this way, over a field season, a bat detector was used less efficiently with citizen scientists than with professional biologists.

As the technology advances, a variety of detectors become available. Some of them are designed for amateurs with simpler designs that are more user-friendly. We recommend investigating the possibility to use these less expensive detectors in the future if a citizen science component is needed. Compared to other citizen scientist projects in NC, equipment used in NABat is very expensive and complicated. Training of participants and coordination among individuals is time consuming as the equipment is limited and complicated. Another issue of including citizen scientists at the broad scale project is the transfer/transportation of equipment between participants. With current detectors, the shipping cost is prohibitive. Another issue is the safety of participants during driving. The survey protocol requires a vehicle speed that is lower than other vehicles on the road. Thus, there needs to be appropriate liability protections for both the organizers and the participants.

ADOPTING STATIONARY POINT SURVEY

In the pilot project, we only invited a few professional wildlife biologists to participate in the stationary point survey through partnerships. The outcome was positive. Most partners have professional experience that help them conduct the survey. One inherent issue of stationary point survey is that the placement of a bat detector affects data quality and quantity. As the environment changes over time by either vegetation growth or management, almost all stationary point survey sites need to be re-evaluated every year. This site evaluation process requires special knowledge from very experienced bat biologists and cannot be done by others. This is one reason we suggested a centralized coordinator as this person should be responsible for stationary point site selection and detector placement decision. Based on our experience in the past two years, this can be done within a month prior the season.

In general, we do not recommend involving citizen scientists for stationary point surveys. First it is hard to provide access legally to all volunteers. In the current survey network, all public lands require a specific permit to conduct research. It is not possible to include citizen scientists on these permits. Second, stationary point survey requires more specific timing for data integrity. The level of work commitment cannot be fulfilled by volunteers. Third, the effort for equipment maintenance and physical labor is not appropriate for most volunteers. Instead, we suggest involving volunteers through professional partners. For example, local science education organizations may adopt the stationary point survey sites and be responsible for surveying these sites. Those organizations can recruit volunteers independently and incorporate NABat surveys as part of their education programs. In this way, the level of guidance for citizen scientists will be much higher. The education

can be conducted by professionals. The quality of data is controlled by both the education organization and the person responsible for integrating data.

Another way to involve volunteers is through land owners who provide land access for the stationary point survey. In the past two years, we had over 15 private land owners involved in this project. Most land owners provided not only land access, but also information of the land (such as topography and vegetation). Many land owners also routinely checked the equipment. We suggest that those landowners' works should be counted as volunteer hours. Even though the data collection was essentially conducted by professionals, the effort from land owners should be documented.

ISSUE OF DATA MANAGEMENT AND ANALYSIS

The major unsolved issue in implementing NABat is data management and analysis. As shown earlier, the number of files collected in the project is very large. We have not identified the ideal platform for managing the data. Currently external hard drives are used to store data. However, an efficient way to index, archive, and store files should be developed. This issue is faced by all scientists who use acoustic methods and might need professionals from other fields (such as computer science or engraining) to solve.

How to accurately identify bats to species through acoustic files is also an unsolved issue. We attempted different ways including completely manual identification, completely automatic identification with different programs and different program settings, and combinations of automatic identification and manual vetting. As presented above, it is manageable to manually identify transect all mobile transect survey data. But the amount of stationary point survey data is too large to be process in the same manner. We believe the ideal method for acoustic species identification is through the combination of automatic identification and manual vetting. However, more investigations are needed to identify how to balance these two components to be efficient and manageable.

RESEARCH BEYOND SURVEYS

The NABat survey provided opportunities to address many scientific questions beyond the inventory baseline survey. In the past two years, many research projects were developed in the NABat framework. These projects include:

- Separating the effects of urbanization and water quality degradation on bat distribution. This project has been summarized into a manuscript and is under review.
- Determining species-specific nightly bat activity in areas with different urbanization intensity. This is part of a master student thesis and being prepared as a manuscript for review.
- Investigating the effectiveness of a shortened NABat mobile transect for sampling bats in urban areas. This is part of a master student thesis and being prepared as a manuscript for review.
- Evaluating inherent observation probability variations related to NABat protocols. This project needs more in depth statistical analysis and is expected to be summarized into a manuscript in the fall of 2017.
- Identifying threshold patterns in bats responses to urbanization. This project needs more in depth statistical analysis and is expected to be summarized into a manuscript in the fall of 2017.
- Investigating the spatial configuration effects of landscape element on bat distribution. This project needs either another year of data or data from other states for spatial replicates.

Overall, NABat provides research opportunities to involve more scientists. Through partnership and data sharing more questions can be answered. We have heard that participants are interested in implementing NABat protocols to make their research more relevant and comparable with data available at larger scales.

CONCLUSION

In the past two years, we piloted the NABat in NC. We set up a summer acoustic survey framework and started collecting baseline information at a broad scale. We initiated a partnership network to involve professional wildlife biologists and citizen scientists to participate in NABat. There are still some issues such as data management and analysis, and population trend estimation that need to be solved. We anticipate that these problems will be solved with more data in the future and through innovative collaborations. The highlights of this projects are listed below:

- We sampled 57 NABat grid cells (41 with mobile transect survey, 40 with stationary point survey) in two years and collected over 300,000 bat acoustic files.
- We developed four protocol guides and two datasheets for future NABat implementation.
- Development of a multi-stakeholder collaboration network was initiated for future NABat implementation.
- We detected 14 species of bats.
- Mobile transect surveys and stationary point surveys revealed similar general patterns of bat distributions in NC.
- Federally list species:
    - MYSO - mainly concentrated in Cherokee National Forest near the NC/TN border
    - MYGR - mainly distributed along Appalachian Mountains between Asheville and Boone
    - MYSE - more found in the coastal plain than the rest of NC

SUMMARY OF PRESENTATIONS BASED ON THIS PROJECT

Urbanization effects on bats across multiple North Carolina cities within the NABat sampling framework. **Han Li**, Ashley Matteson, Katherine Caldwell, and Matina Kalcounis-Rueppell. NASBR annual conference, Monterey, California, October, 2015

Urbanization effects on bats across multiple North Carolina cities within the NABat sampling framework. **Han Li**, Ashley Matteson, Katherine Caldwell, and Matina Kalcounis-Rueppell. South Carolina/North Carolina joint bat working group meeting, Crowders Mountain state park, North Carolina, December, 2015

Urbanization effects on bats across multiple North Carolina cities within the NABat sampling framework. **Han Li**, Katherine Caldwell, and Matina Kalcounis-Rueppell. 21ST annual meeting of the southeastern bat diversity network and 26th annual colloquium on the conservation of mammals in the southeastern US, Guntersville state park, Alabama, February, 2016

Understanding urban and landscape ecology of North Carolina's bats using the North American Bat Monitoring Program (NABat) sampling framework, **Matina Kalcounis-Rueppell**, Han Li, and Katherine Caldwell. TWS North Carolina Chapter annual meeting, Haw River State Park, North Carolina, March, 2016

Landscape scale analysis of urbanization effects on bat distribution in North Carolina, **Han Li**, Katherine Caldwell, and Matina Kalcounis-Rueppell. US-IALE annual meeting, Asheville, North Carolina, April, 2016

Landscape scale analyses on the effects of urban land covers on bat distributions in North Carolina, **Han Li**, Katherine Caldwell, and Matina Kalcounis-Rueppell. The 101th Ecological Society of America annual meeting, Fort Lauderdale, Florida, August, 2016

Landscape level interactions of water quality and urbanization on bats, Han Li and **Matina Kalcounis-Rueppell**. The 17th International Bat Research Conference, Durban, South Africa, August, 2016

Urbanization effects on bats across multiple North Carolina cities within the NABat Sampling Framework, **Han Li**, Katherine Caldwell, and Matina Kalcounis-Rueppell. The 23rd annual conference of The Wildlife Society, Raleigh, North Carolina, October, 2016

Detection probability differences within the North American Bat Monitoring Program (NABat), **Kevin Parker Jr.**, Han Li, and Matina Kalcounis-Rueppell. The 23rd annual conference of The Wildlife Society, Raleigh, North Carolina, October, 2016

Determining species-specific nightly bat activity in sites with varying urban intensity, **Sarah Schimpp**, Han Li, and Matina Kalcounis-Rueppell. The 23rd annual conference of The Wildlife Society, Raleigh, North Carolina, October, 2016

Urbanization effects on bats across multiple North Carolina cities within the NABat Sampling Framework, **Han Li**, Katherine Caldwell, and Matina Kalcounis-Rueppell. Native American Fish and Wildlife Society Southeast and Northeast Regional Meeting, Cherokee, North Carolina, October, 2016

Two years of NABat survey: Separating the effects of water quality and urbanization on bats at landscape scale, **Han Li**, Katherine Caldwell, and Matina Kalcounis-Rueppell. North Carolina bat working group meeting, Haw River state park, North Carolina, December, 2016

Separating the effects of urbanization and water quality degradation on bat distributions. **Han Li** and Matina Kalcounis-Rueppell. 22nd annual meeting of the southeastern bat diversity network and 27th annual colloquium on the conservation of mammals in the southeastern US, Asheville, North Carolina, February, 2017

Effectiveness of a shortened NABat protocol for sampling bats in urban areas. **Sarah Schimpp**, Han Li, and Matina Kalcounis-Rueppell. 114th annual meeting of the North Carolina Academy of Science (NCAS), High Point, North Carolina, March, 2017

Comparing the effectiveness of the North American Bar Monitoring Program (NABat) acoustic survey protocols. **Han Li**, Kevin Parker Jr., and Matina Kalcounis-Rueppell. 2nd international symposium on bat echolocation research: learn to listen, Tucson, Arizona, March, 2017

Effectiveness of a shortened NABat protocol for sampling bats in urban areas. **Sarah Schimpp**, Han Li, and Matina Kalcounis-Rueppell. 2nd international symposium on bat echolocation research: learn to listen, Tucson, Arizona, March, 2017

MAJOR PROJECT EQUIPMENT

A list of major equipment purchased is below:

Table 8 Equipment purchased for project

Item	Amount
AnaBat SD2 Bat Detector with Stainless Microphone	11
AnaBat Car mount with suction cup and 3m detachable cable	4
Additional stainless Microphone	2
Mouse GPS powered through USB port with an included AA battery pack	4
Water proof setup including: 1400 Pelican case configured for detector and 12-volt battery, internal cabling, 1 battery connector lead, 3m microphone extension cable, weather proof weather head with stainless microphone	8
AnaBat Equalizer - Chirper II, Jig, and Equalizer Software which electronically controls the detector and sets the sensitivity of the detectors	1
4 AA Low Self Discharge 2000mAH Rechargeable NiMH Batteries	6
Battery Maintenance Kit with connectors that match those used in AnaBat Weather Proof Setups including: 12 volt 1.25 amp battery charger, volt meter (to measure and health), and 3 Y-connectors (to enable up to battery voltage 4 batteries at once)	3
12 Volt 7.5 Amp Sealed Lead Acid Battery	16
Battery Connector to Power Lead	8
Tripod Mount	4
T- Post Mounting Bracket	2
Belt and Ratchet Mounting Bracket	2
Dual Angle Weather Head Mount to attach to poles: 1 inch	4
Python Lock by Master Lock	8
Solid brass padlock	8
Memory CF Card 4GB	5
Memory Card Reader - USB 2.0 CF and SD	2

LITERATURE CITED

- Grider, John F., Angela L. Larsen, Jessica A. Homyack, and Matina C. Kalcounis-Rueppell. 2016. "Winter Activity of Coastal Plain Populations of Bat Species Affected by White-Nose Syndrome and Wind Energy Facilities." PLOS ONE 11 (11): e0166512. doi:10.1371/journal.pone.0166512.
- Kalcounis-Rueppell, M. C., V. H. Payne, S. R. Huff, and A. L. Boyko. 2007. "Effects of Wastewater Treatment Plant Effluent on Bat Foraging Ecology in an Urban Stream System." Biological Conservation 138 (1–2): 120–30. doi:10.1016/j.biocon.2007.04.009.
- Li, Han, and Kenneth T. Wilkins. 2014. "Patch or Mosaic: Bat Activity Responds to Fine-Scale Urban Heterogeneity in a Medium-Sized City in the United States." Urban Ecosystems, no. 17: 1013–31. doi:10.1007/s11252-014-0369-9.
- Li, Han, and Kenneth T. Wilkins. 2015. "Selection of Building Roosts by Mexican Free-Tailed Bats (Tadarida Brasiliensis) in an Urban Area." Acta Chiropterologica 17 (2): 321–30. doi:10.3161/15081109ACC2015.17.2.007.

- Loeb, Susan C., and Eric R. Britzke. 2010. "Intra- and Interspecific Responses to Rafinesque's Big-Eared Bat (Corynorhinus Rafinesquii) Social Calls." Acta Chiropterologica 12 (2): 329–36. doi:10.3161/150811010X537909.
- Loeb, Susan C., Thomas J. Rodhouse, Laura E. Ellison, Cori L. Lausen, Jonathan D. Reichard, Kathryn M. Irvine, Thomas E. Ingersoll, et al. 2015. "A Plan for the North American Bat Monitoring Program (NABat)." http://www.treesearch.fs.fed.us/pubs/48442
- North Carolina Wildlife Resources Commission. 2015. North Carolina wildlife action plan. Raleigh, NC: North Carolina Wildlife Resources Commission. http://www.ncwildlife.org/Plan

ACKNOWLEDGEMENTS

The following people contributed to this project in various manners. We sincerely thank for their helps:

- NCWRC: William Ridgeway, Maria Dunn, David Allen, Kacy Cook, John Carpenter, Kendrick Weeks, Allen Boynton, Brandon Sherrill, Gabrielle Graeter, Isaac Harrold, Allison Melford;
- NCDEQ: Michael Walters;
- North Carolina Coastal Land Trust: Janice Allen, Jesica Blake;
- US Army: Janice Patten, Eric Britzke;
- Eastern Band of Cherokee Indians: Caleb Hickman, Dallas Bradley, Josh Paris, Michael LaVoie;
- USFS: Sheryl Bryan, Deb Walker, Susan Loeb;
- USFWS: Laura Fogo, Susan Cameron, Michael Morse;
- Private volunteers: Garcy and Allison Ward, Shane Staples, David and Cathy Fields, Karen Curry, Stacey Weddle, Michelle Amato, Emilie Travis, Bob Bryden, Barbara and Keith Bollman, Kristy King, Tim Tapio, Laura Ellison, Kevin and Gretchen Parker, Tom and Mary Vann, Scott Perry, Jim Willis, Michael Whitby, Bradley Saul, Ashely Matteson;
- Wake County: Deborah Fowler;
- Mecklenburg County: Catherine Luckenbaugh and fellow biologists;
- NC Aquarium: Peggy Sloan, Joanna Zazzali, Andrew Moore;
- Salem College: Traci Porter;
- NC Zoological Park: Melissa Vindigni, Nell Allen, Corinne Kendall, Bailey Morgan, Megan Cline, Heidi Faris;
- NC Division of Parks and Recreation: Ed Corey;
- UNCG: Stan Faeth, Paul Hidalgo, Omar Raya, Kevin Parker;
- NC Museum of Natural Sciences: Stephanie Schuttler;
- UNCW: David Webster;
- Titley Scientific: Kim Livengood, Chris Corben,

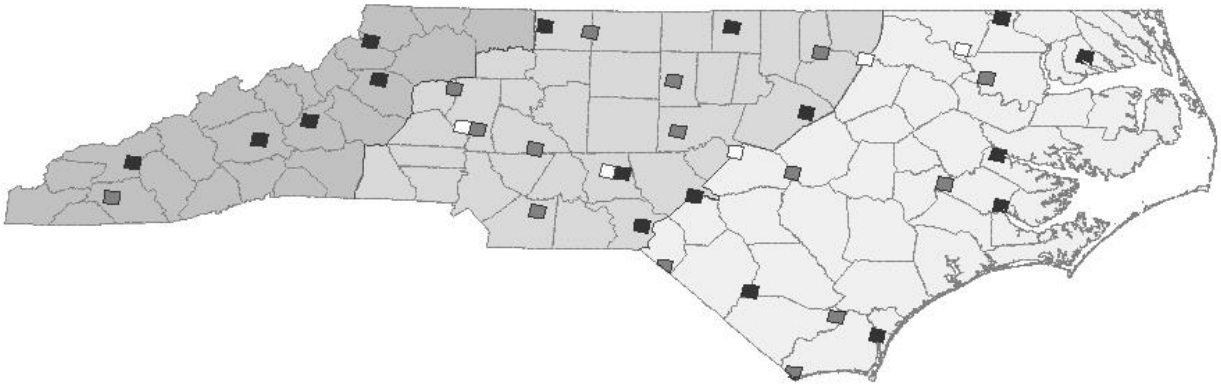
APPENDICES

- Appendix 1 – Protocol-NABat mobile transect survey guide
- Appendix 2 – Data Sheet-Various data sheets for NABat NC
- Appendix 3 – Protocol-NABat AnaBat training manual

- Appendix 4 – Protocol-NABat stationary point survey guide
- Appendix 5 – Protocol-NABat local data management and acoustic analysis guide

Driving Transect Survey
Route Directions and Maps

North American Bat Monitoring Program
North Carolina Division



Dr. Han Li
Ashley Matesson
Dr. Matina Kalcounis-Rueppell

University of North Carolina at Greensboro
Department of Biology
321 McIver St.
312 Eberhart Building
Greensboro, NC 27412

September 2015

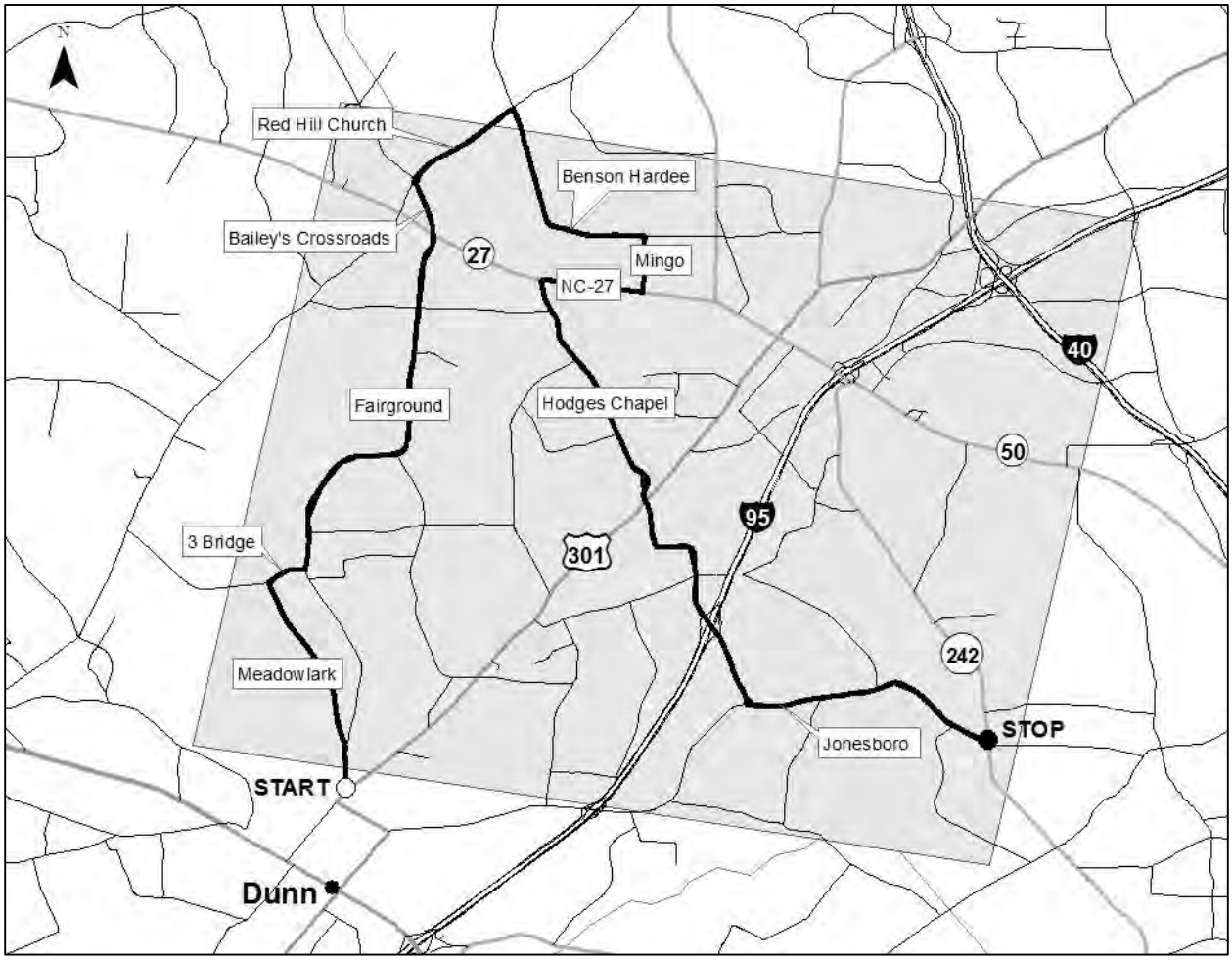
Table of Contents

Grid 0 – Dunn - Harnett County

	Latitude	Longitude	Intersection
Start	35.326218	-78.608077	Old Fairground Rd. and Meadowlark Rd.
Stop	35.326550	-78.520276	Dragstrip Rd. and NC-242

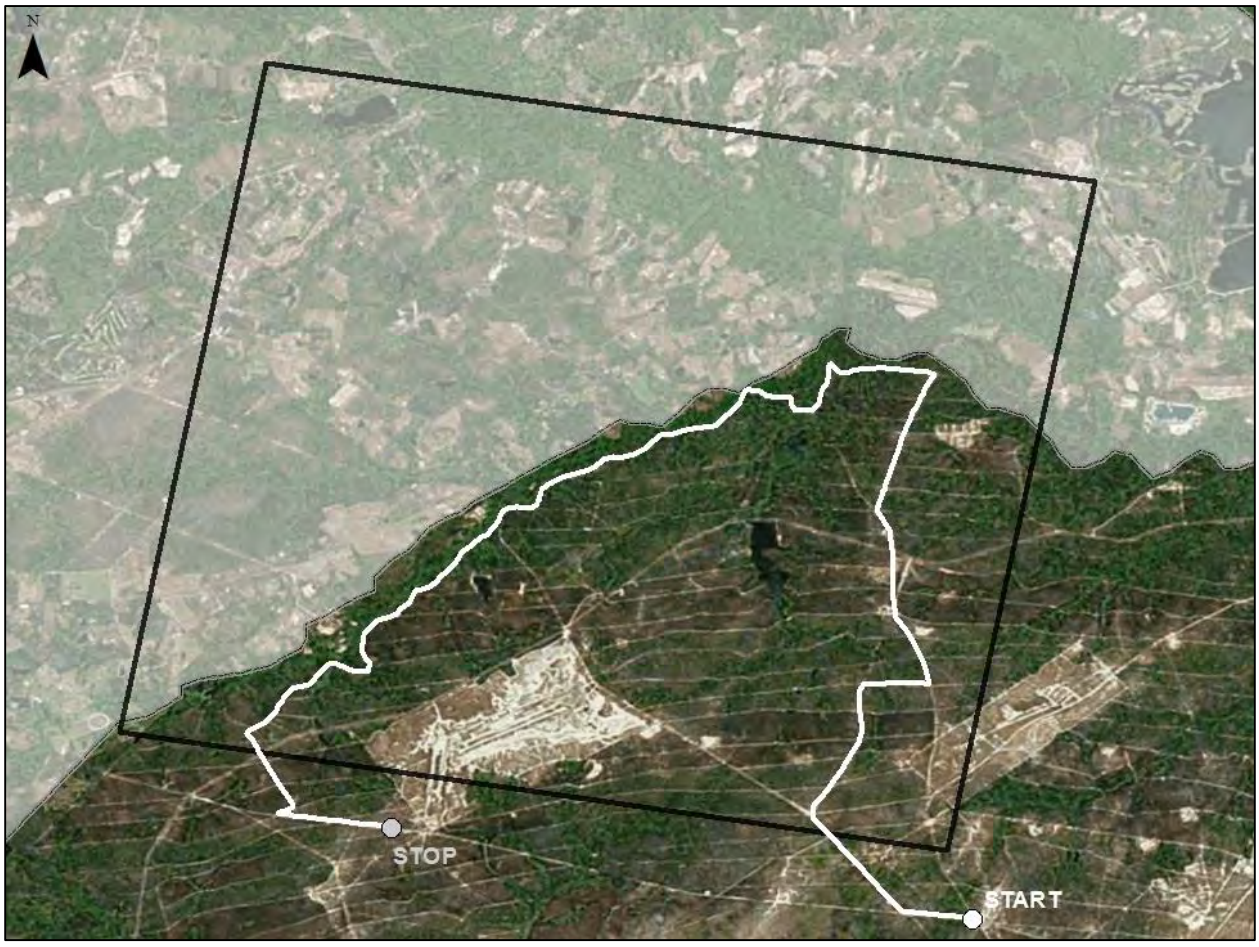
Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	Y No stop sign	Meadowlark Rd.	SR-1715	2
Right	T Stop sign	Three Bridge Rd.	SR-1722	0.3
Left	T Stop sign	Fairground Rd.	SR-1705	3.7
Straight	+ Stop sign	Baileys Crossroads Rd.	SR-1551	0.5
Right	+ No stop sign	Red Hill Church Rd. ** Denning Rd. (after crossing county line)*	SR-1703 *SR-1168*	0.2 *0.8*
Right	+ Stop sign	Benson Hardee Rd.	SR-1303	1.9
Right	T No stop sign	Mingo Rd.	SR-1302	0.5
Right	+ Stop sign	NC-27 West ** if necessary drive 55mph*	NC-27 W	0.8
Left	T No stop sign	Hodges Chapel Rd.	SR-1709	4.6
Right	T Stop sign	Jonesboro Rd. ** Dragstrip Rd. (after crossing county line)*	SR-1808 *SR-1107*	1.3 *0.8*

Grid 0 – Dunn


N
Red Hill Church
Benson Hardee
Bailey's Crossroads
27
Mingo
NC-27
40
Fairground
Hodges Chapel
50
95
3 Bridge
301
242
Meadowlark
STOP
Jonesboro
START
Dunn

Grid 2 – Fort Bragg – Hoke County

	Latitude	Longitude
Start	35.145208	-79.30726
Stop	35.133287	-79.229825

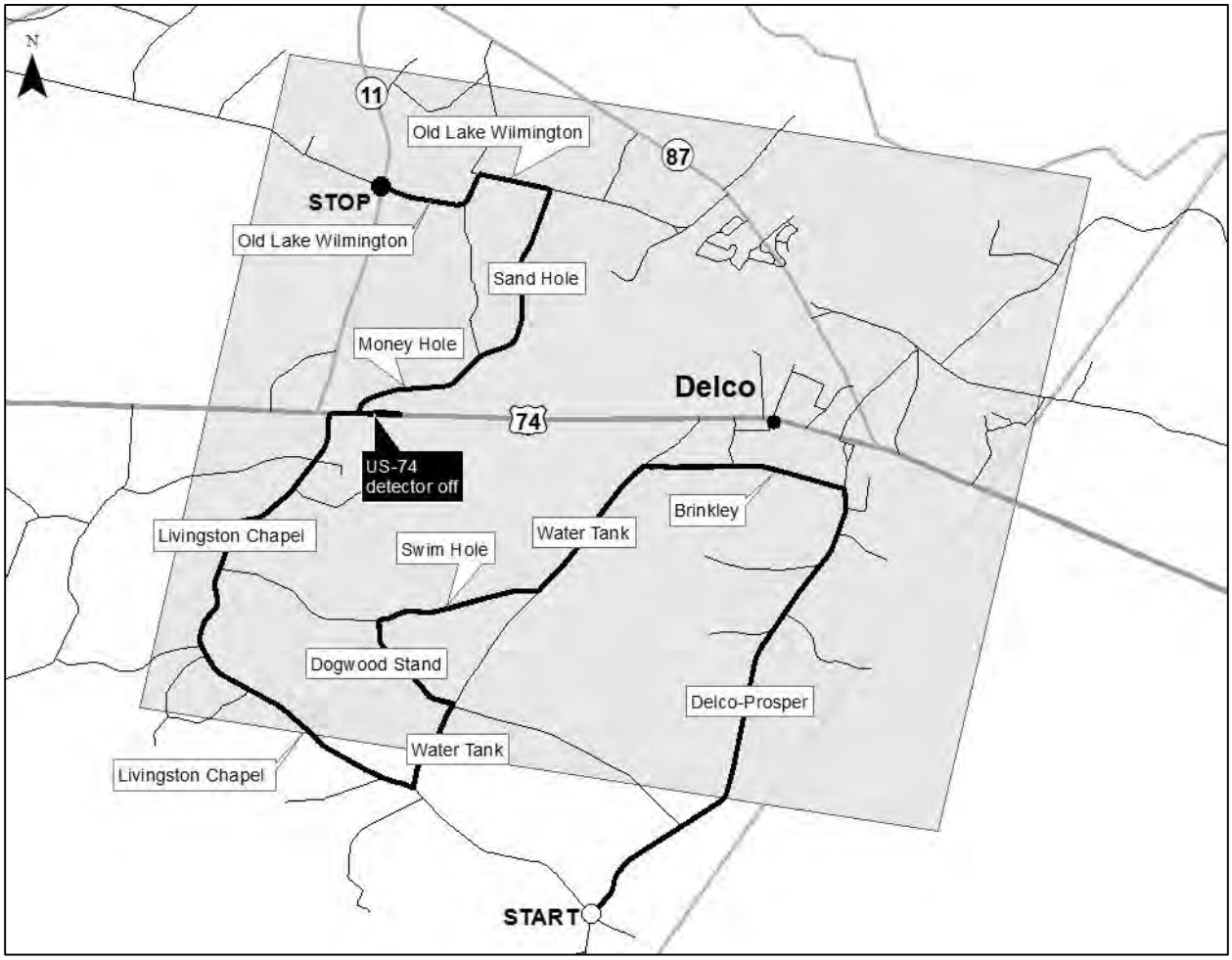


Grid 3 – Delco – Columbus County

	Latitude	Longitude	Intersection
Start	34.249722	-78.248821	Delco-Prosper Rd. and Horseshoe Rd.
Off	34.317067	-78.284045	
On	34.317177	-78.280193	
Stop	34.347624	-78.276919	Old Lake Rd. and NC-11

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	+ Stop sign	Delco-Prosper Rd.	SR-1849	4.6
Left	T No stop sign	Brinkley Rd.	SR-1851	1.6
Left	T Stop sign	Water Tank Rd.	SR-1824	1.4
Right	T No stop sign	Swim Hole Rd.	SR-1831	1.3
Left	T No stop sign	Dogwood Stand Rd.	SR-1830	1
Right	T Stop sign	Water Tank Rd.	SR-1824	0.8
Right	T Stop sign	Livingston Chapel Rd.	SR-1843	4.7
Right	T Stop sign	Andrew Jackson Hwy **** turn off detector and drive normal highway speed***	US-74 E	1.3
Left	U-turn No stop sign	Andrew Jackson Hwy	US-74 W	1
Right	T No stop sign	Money Hole Rd. **** turn on detector and drive transect speed***	SR-1845	1.1
Right	Y No stop sign	Sand Hole Rd.	SR-1846	1.8
Left	T Stop sign	Old Lake Wilmington Rd.	SR-1740	0.6
Left	T No stop sign	Old Lake Wilmington Rd. ** road name change to Old Lake Rd.*	SR-1740	1

Grid 3 – Delco

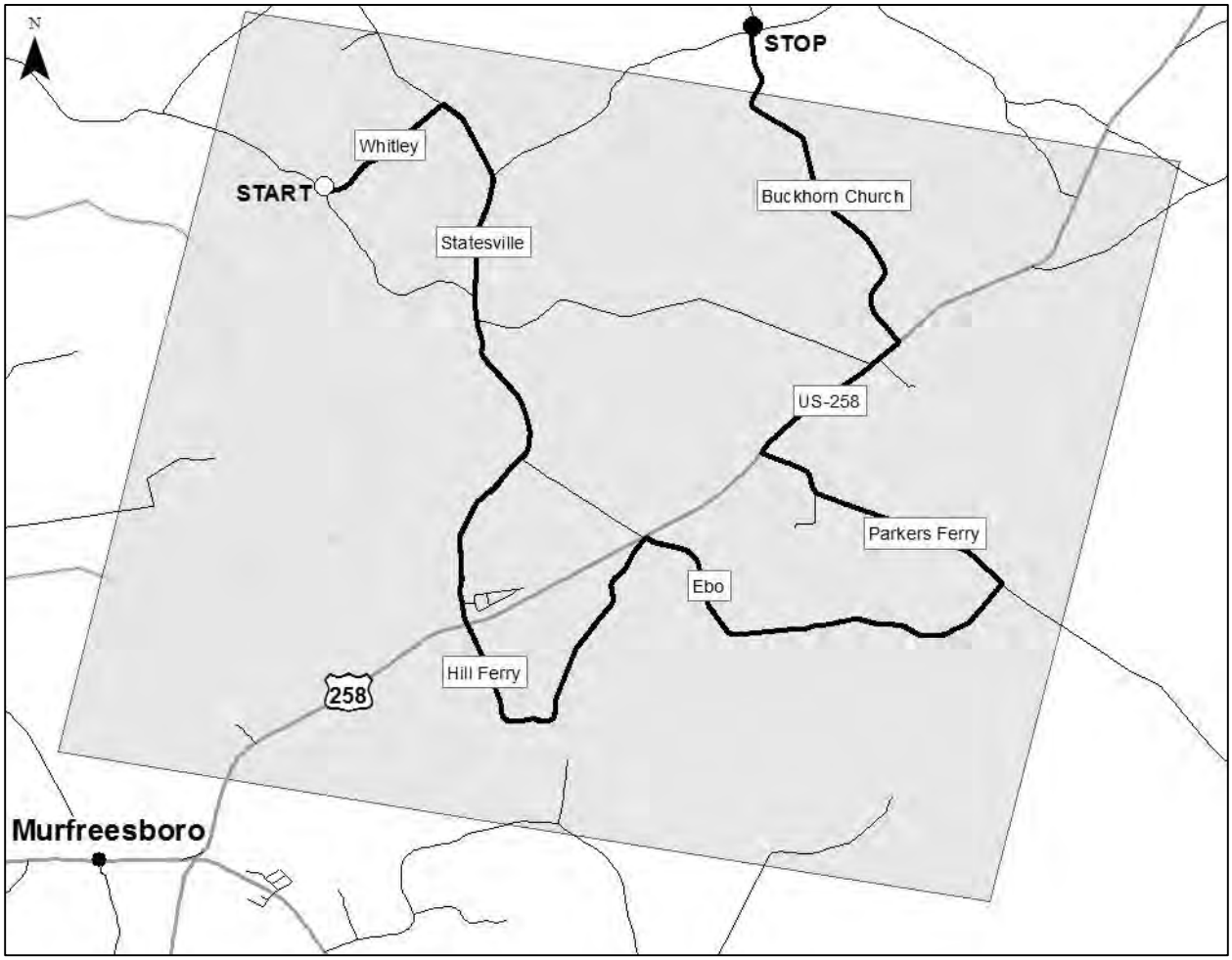

N
11
Old Lake Wilmington
87
STOP
Old Lake Wilmington
Sand Hole
Money Hole
Delco
74
US-74
detector off
Brinkley
Livingston Chapel
Swim Hole
Water Tank
Dogwood Stand
Delco-Prosper
Water Tank
Livingston Chapel
START

Grid 7 – Murfreesboro – Hertford County

	Latitude	Longitude	Intersection
Start	36.521445	-77.071682	Boone's Bridge Rd. and Whitley Rd.
Stop	36.540213	-77.021747	Buckhorn Church Rd. and New Hope Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T No stop sign	Whitley Rd.	SR-1314	1
Right	T Stop sign	Statesville Rd.	SR-1310	4.6
Straight	+ Stop sign	Hill Ferry Rd.	SR-1309	2.8
Right	5-way Stop sign	Ebo Rd.	SR-1308	3.1
Left	T Stop sign	Parkers Ferry Rd.	SR-1306	2
Right	T Stop sign	US-258	US-258	1.3
Left	T No stop sign	Buckhorn Church Rd.	SR-1316	2.9

Grid 7 – Murfreesboro

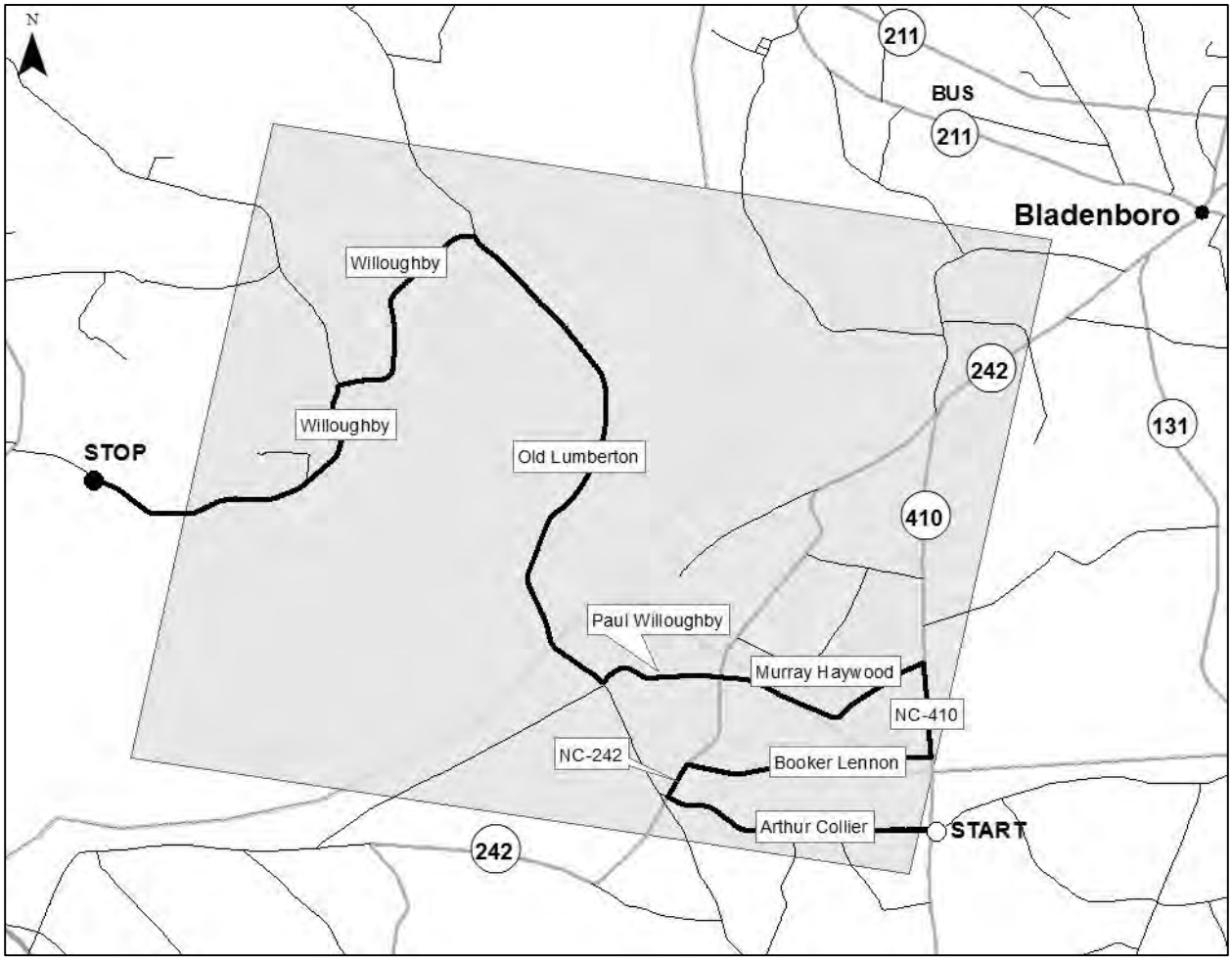


Grid 13 – Bladenboro – Bladen County

	Latitude	Longitude	Intersection
Start	34.453081	-78.825576	Arthur Collier Rd. and NC-410 (Joe Brown Hwy)
Stop	34.501886	-78.941401	Willoughby Rd. and NC-72

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	+ No stop sign	Arthur Collier Rd.	SR-1519	2.2
Right	+ Stop sign	Haynes Lennon Hwy	NC-242	0.3
Right	T No stop sign	Booker Lennon Rd.	SR-1518	2
Left	T Stop sign	Joe Brown Hwy	NC-410	0.9
Left	T No stop sign	Murray Haywood Rd. ** at 0.8 mile veer right staying on Murray Haywood Rd.*	SR-1185	1.9
Straight	+ Stop sign	Paul Willoughby Rd.	SR-1185	1
Right	T Stop sign	Old Lumberton Rd.	SR-1002	5
Left	T No stop sign	Willoughby Rd. ** at 0.1 there is a Y-intersection stay left on Willoughby Rd.*	SR-2121	2.1
Left	T Stop sign	Willoughby Rd.	SR-2121	3.6

Grid 13 – Bladenboro

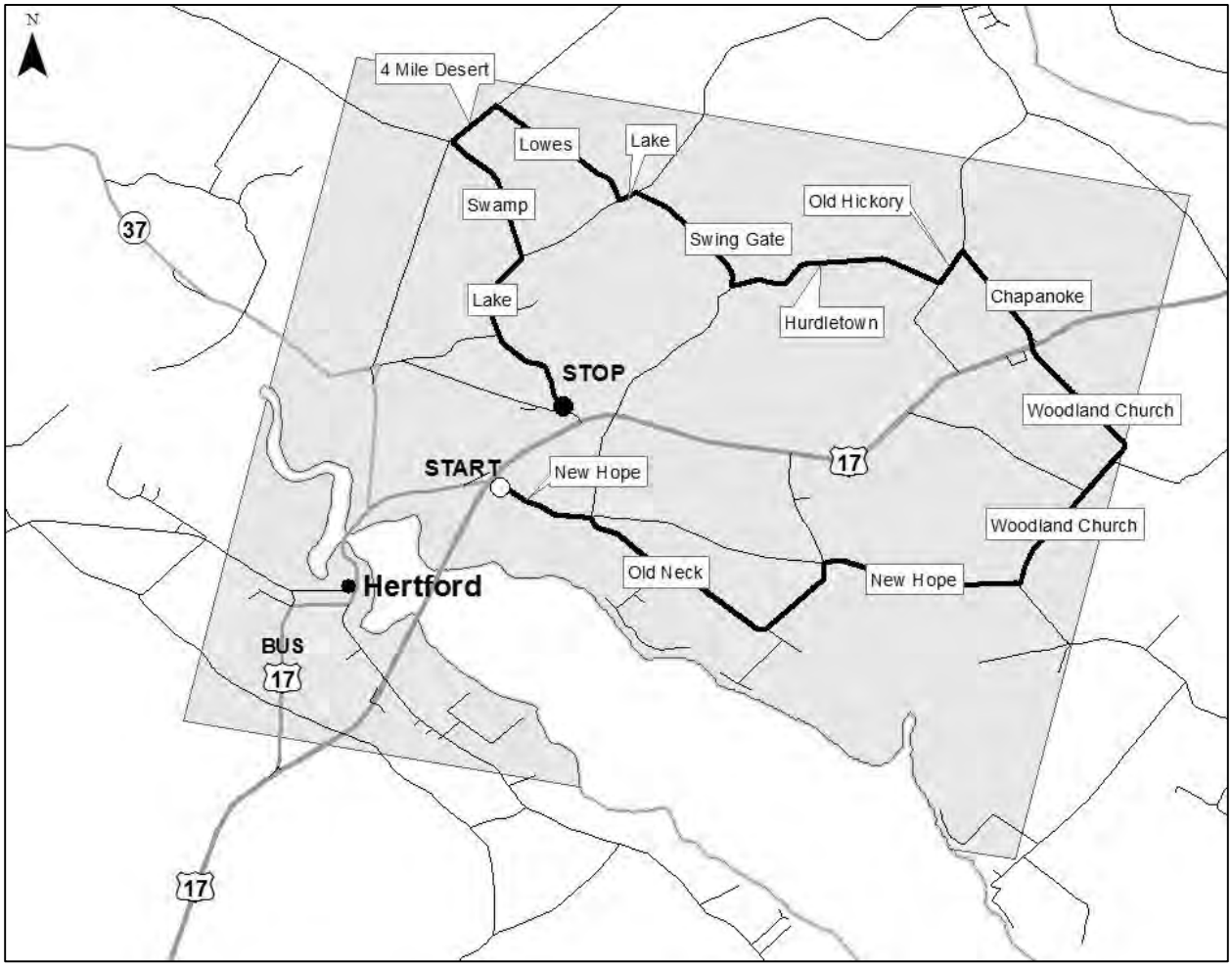


Grid 14 – Hertford – Perquimans County

	Latitude	Longitude	
Start	36.203874	-76.446475	US-17/ NC-37 and New Hope Rd.
Stop	36.213645	-76.437852	Lake Rd. and Wiggins Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	+ Traffic light	New Hope Rd.	SR-1300	0.8
Right	+ No stop sign	Old Neck Rd. ** at 1.6 miles 90° left turn, staying on Old Neck Rd*.	SR-1301	2.4
Right	+ Stop sign	New Hope Rd.	SR-1300	1.5
Left	Y No stop sign	Woodland Church Rd.	SR-1303	1.5
Left	Y No stop sign	Woodland Church Rd.	SR-1303	1.1
Straight	+ Stop sign	Chapanoke Rd. ** cross US-17*	SR-1225	1
Left	Y No stop sign	Old Hickory Rd.	SR-1226	0.3
Right	T No stop sign	Hurdletown Rd.	SR-1227	1.7
Right	T Stop sign	Swing Gate Rd.	SR-1228	1.1
Left	T Stop sign	Lake Rd.	SR-1221	0.1
Right	T No stop sign	Lowes Ln.	SR-1222	1.3
Left	T Stop sign	4 Mile Desert Rd.	SR-1223	0.5
Left	T Stop sign	Swamp Rd.	SR-1214	1.2
Right	T Stop sign	Lake Rd.	SR-1221	1.7

Grid 14 – Hertford

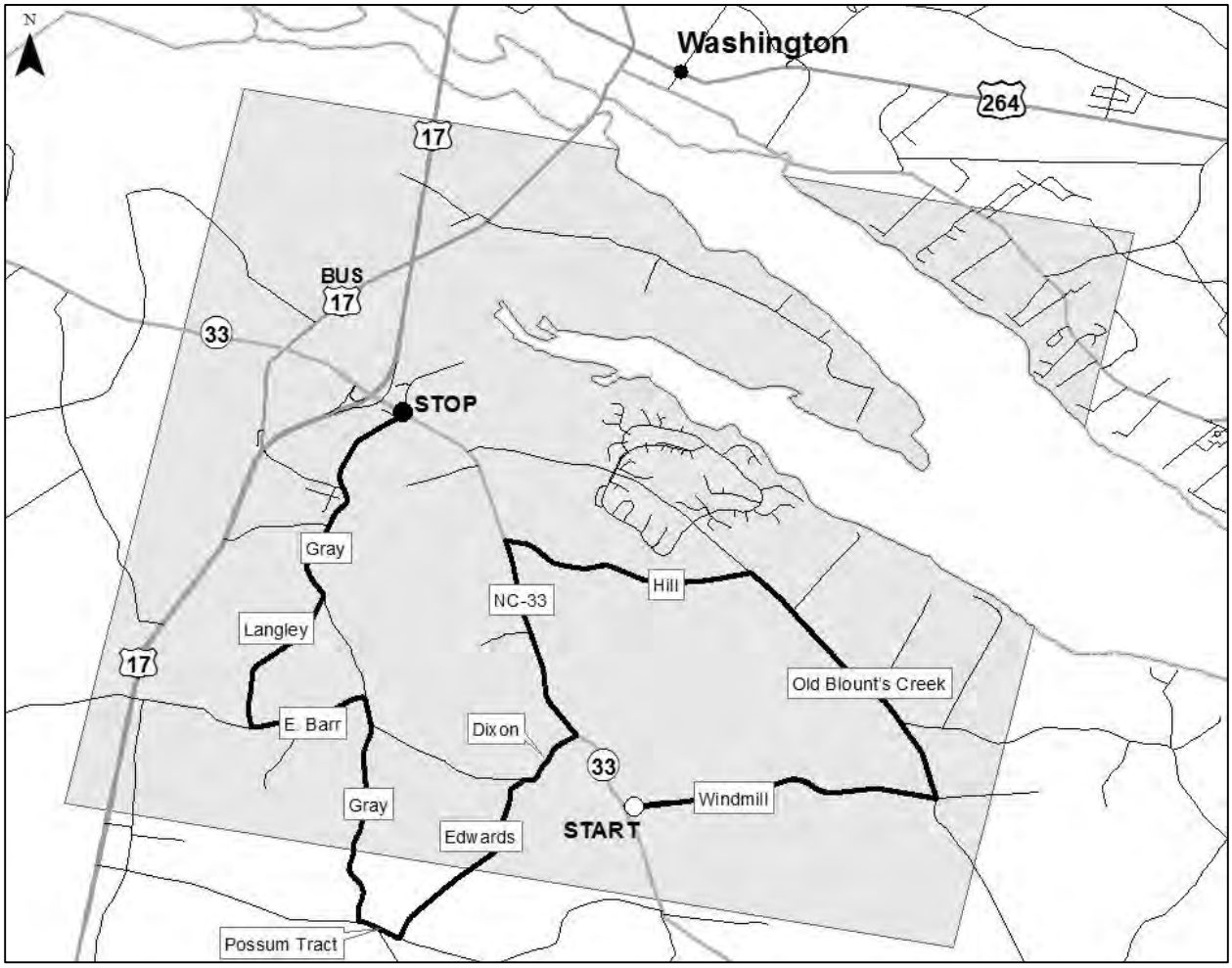


Grid 18 – Washington – Beaufort County

	Latitude	Longitude	Intersection
Start	35.456490	-77.057454	NC-33 and Windmill Rd.
Stop	35.504213	-77.086653	Gray Rd. and N -33

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T No stop sign	Windmill Rd.	SR-1124	2.2
Left	+ Stop sign	Old Blount's Creek Rd.	SR-1114	2.3
Left	Y No stop sign	Hill Rd.	SR-1125	1.8
Left	T Stop sign	NC-33	NC-33	1.7
Right	T No stop sign	Dixon Rd.	SR-1138	0.6
Left	Y No stop sign	Edwards Rd.	SR-1137	1.6
Right	T Stop sign	Possum Tract Rd.	SR-1127	0.3
Right	T No stop sign	Gray Rd.	SR-1136	2
Left	Y No stop sign	E. Barr Rd.	SR-1152	0.9
Right	T No stop sign	Langley Rd.	SR-1151	1.3
Left	Y Stop sign	Gray Rd.	SR-1136	1.8

Grid 18 – Washington

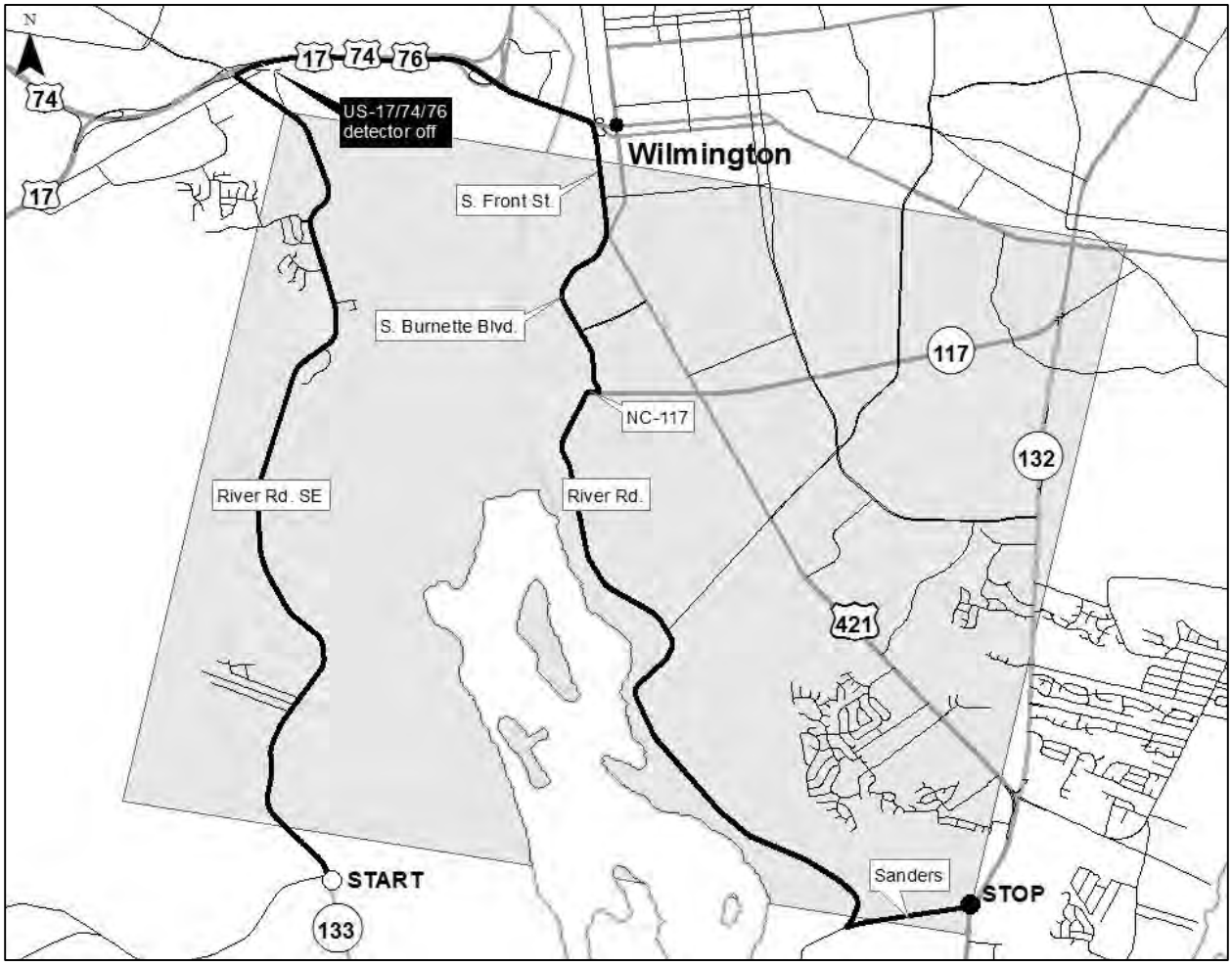


Grid 19 – Wilmington – New Hanover County

	Latitude	Longitude	Intersection
Start	34.130465	-77.981710	River Rd. SE and Daws Creek Rd.
Off	34.231224	-77.992489	
On	34.212008	-77.946032	
Stop	34.125922	-77.900181	Sanders Rd. and US-421

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T Stop sign	River Rd. SE	NC-133	8
Right	+ Traffic light	US-17/ 74/ 76 **** turn off detector and drive normal highway speed***	US-17/ 74/ 76	3
Right	Ramp	S. Front St. ** South Port is on the exit sign*	--	1
Right	+ Traffic light	S. Burnette Blvd. **** turn on detector and drive transect speed***	--	1.5
Right	T Stop sign	US-117	US-117	<0.1
Left	T No stop sign	River Rd.	SR-1576	5.8
Left	Y No stop sign	Sanders Rd.	SR-1187	1

Grid 19 – Wilmington



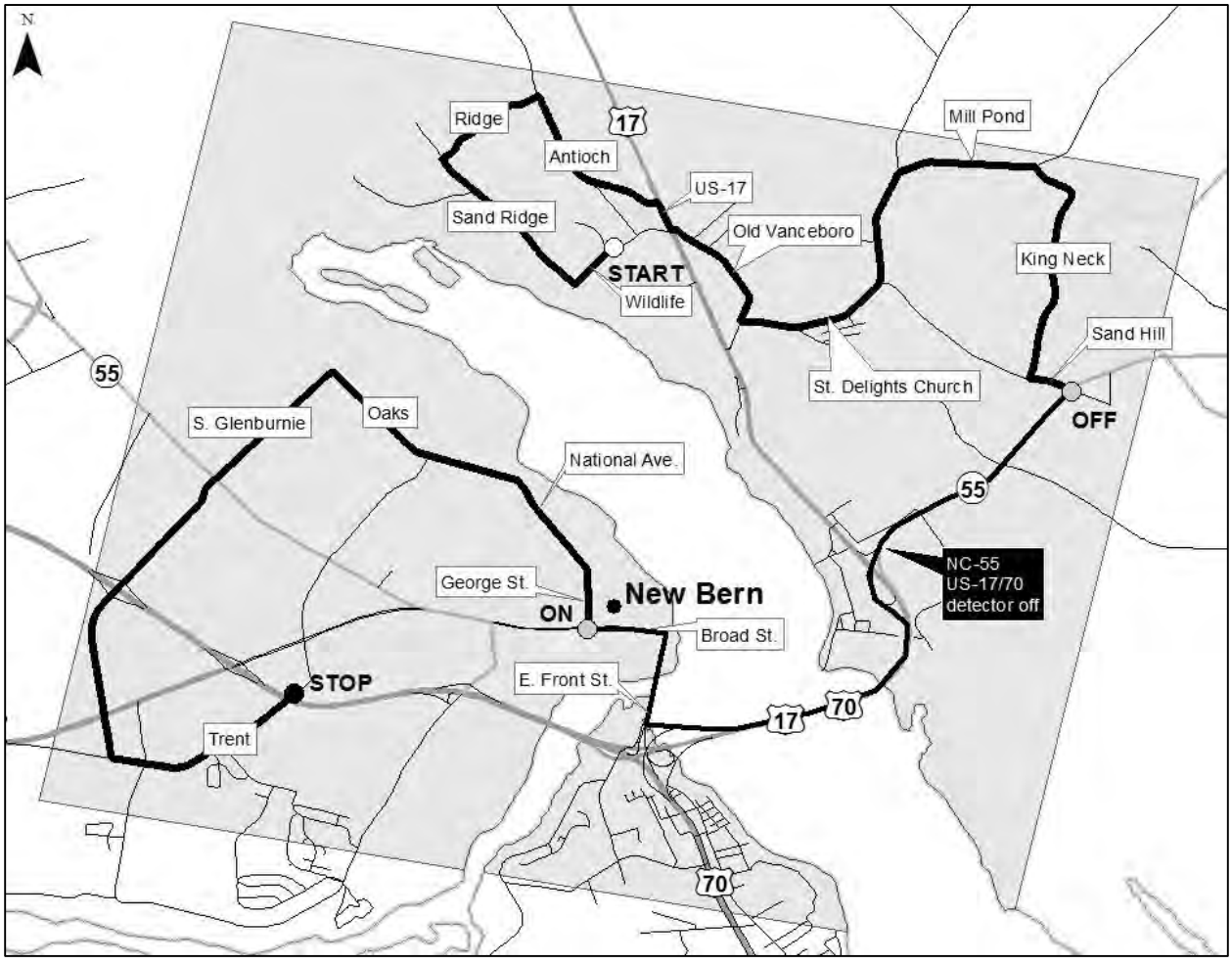
Grid 22 – New Bern – Craven County

	Latitude	Longitude	Intersection
Start	35.150888	-77.041871	Avery Rd. and Wildlife Rd.
Off	35.134999	-76.990538	
On	35.108372	-77.044182	
Stop	35.100078	-77.078245	Trent Rd. passes under US-17/ 70

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Straight	T No stop sign	Wildlife Rd.	SR-1503	0.35
Right	T No stop sign	Sand Ridge Rd.	SR-1492	1.3
Right	T No stop sign	Ridge Rd.	SR-1490	0.8
Right	T Stop sign	Antioch Rd.	SR-1433	0.7
Left	Y No stop sign	Antioch Rd.	SR-1433	0.5
Right	T Stop sign	US-17	US-17	0.2
Left	T No stop sign	Old Vanceboro Rd.	SR-1616	0.9
Left	T No stop sign	St. Delights Church Rd.	SR-1615	0.4
Left	Y No stop sign	St. Delights Church Rd.	SR-1615	1.5
Right	T No stop sign	Mill Pond Rd.	SR-1613	0.9
Right	Y No stop sign	King Neck Rd.	SR-1613	1.8
Left	T Stop sign	Sand Hill Rd.	SR-1614	0.2
Right	T Stop sign	NC-55 **** turn detector off and drive normal highway speed***	NC-55	1.7
Straight	Ramp	US-17 South NC-55 West	US-17 S NC-55 W	2.2
Right	Ramp Exit 417 A-B	US-70 East New Bern	US-70 E	0.3
Right	Ramp Exit 417 A	E. Front St. New Bern	US-70 BUS	0.3
Right	+ Traffic light	E. Front St.	--	0.7

Right	Traffic circle 2nd exit	Broad St.	--	0.5
Right	+ Traffic light	George St. **** turn detector on and drive transect speed*** ** road name changes to National Ave. and then to Oaks Rd.*	--	2.7
Left	T Traffic light	S. Glenburnie Rd. ** SR-1402 changes to SR-1309*	SR-1402	3.5
Left	+ Traffic light	Trent Rd.	SR-1278	1.5

Grid 22 – New Bern

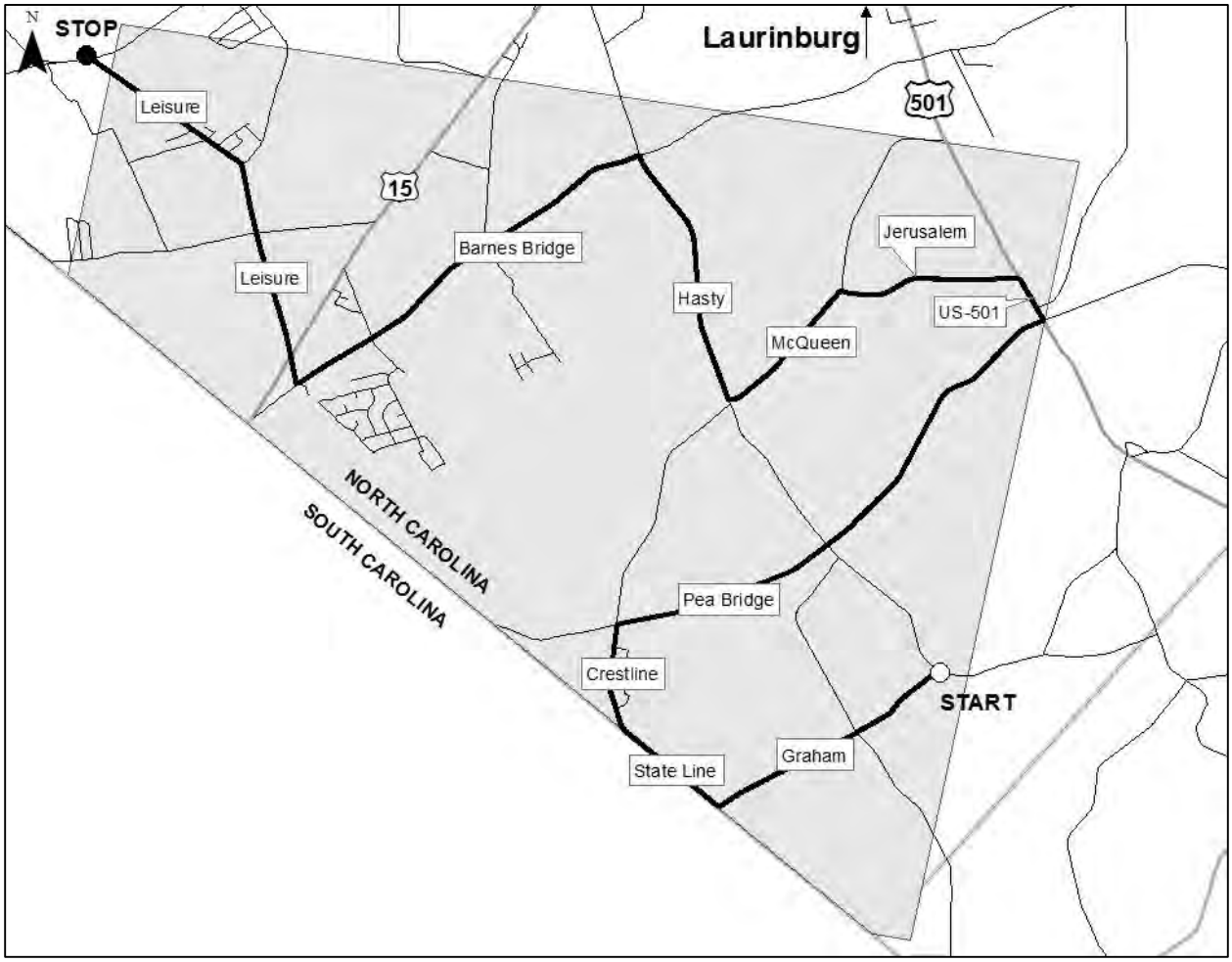


Grid 23 – Laurinburg – Scotland County

	Latitude	Longitude	Intersection
Start	34.659771	-79.454934	Hasty Rd. and Graham Rd.
Stop	34.729229	-79.549995	Leisure Rd. and X-way Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T No stop sign	Graham Rd.	SR-1624	1.7
Right	T No stop sign	State Line Rd.	SR-1625	0.9
Right	Y No stop sign	Crestline Rd.	SR-1622	0.8
Right	+ Stop sign	Pea Bridge Rd.	SR-1619	3.8
Left	+ Stop sign	Johns Rd.	US-501	0.3
Left	T No stop sign	Jerusalem Rd.	SR-1620	1.2
Left	T Stop sign	McQueen Rd.	SR-1621	1.1
Right	T Stop sign	Hasty Rd.	SR-1615	2
Left	+ No stop sign	Barnes Bridge Rd.	SR-1614	2.8
Right	T No stop sign	Leisure Rd.	SR-1100	1.5
Left	Y No stop sign	Leisure Rd.	SR-1100	1.3

Grid 23 – Laurinburg

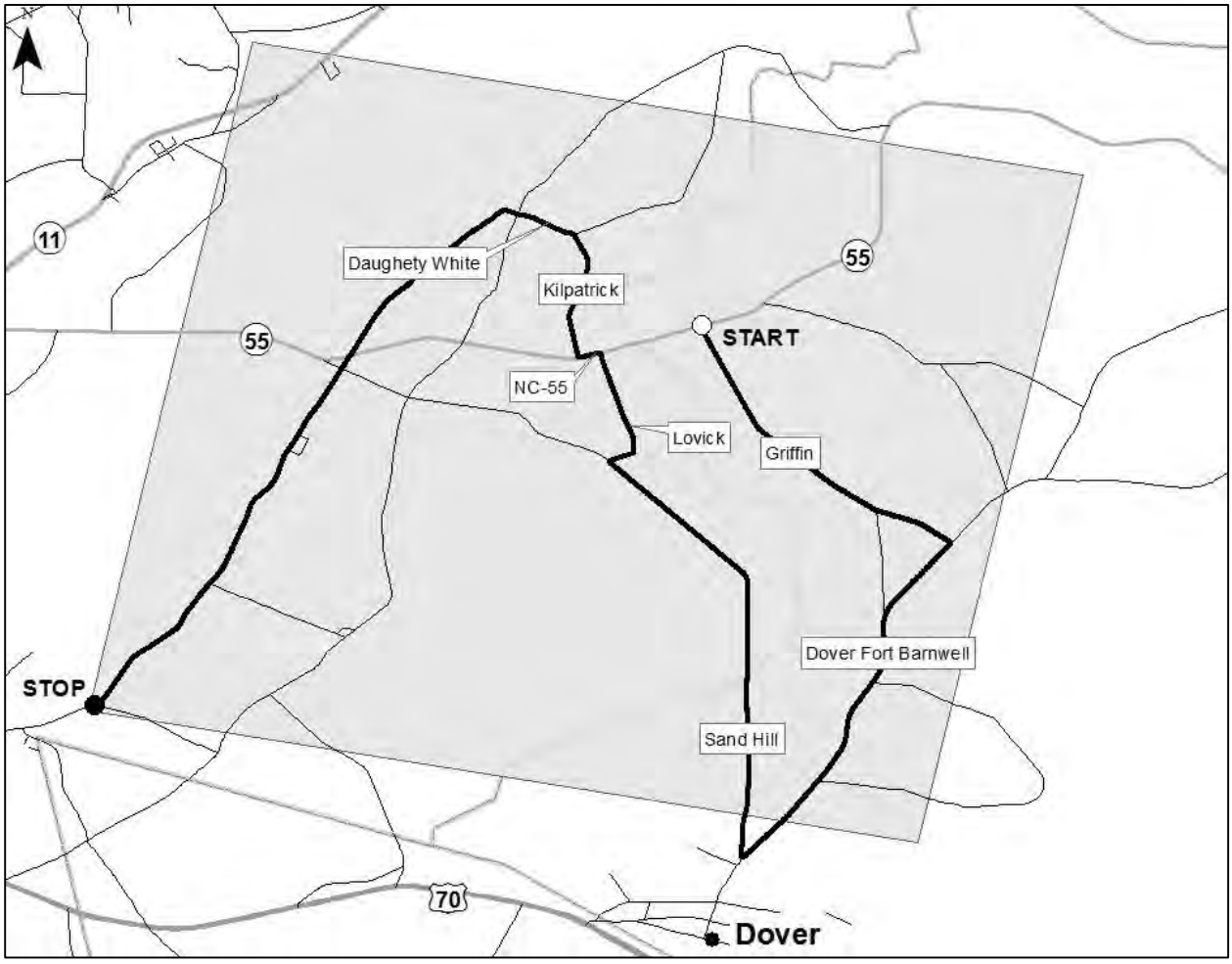


Grid 27 – Dover – Craven County

	Latitude	Longitude	Intersection
Start	35.296667	-77.438586	NC-55 and Griffin Rd.
Stop	35.247195	-77.518377	Neuse Rd. and Gray-Tilghman Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T No stop sign	Griffin Rd.	SR-1271	2.8
Right	T Stop sign	Dover Fort Barnwell Rd.	SR-1262	3.3
Right	Y No stop sign	Sand Hill Rd. ** after crossing county line changes to Seth West Rd. (SR-1807)*	SR-1264	1.6
Right	T No stop sign	Lovick Rd.	SR-1806	1.1
Left	T Stop sign	NC-55	NC-55	<0.2
Right	T No stop sign	Kilpatrick Rd.	SR-1805	1.2
Left	T Stop sign	Daughety White Rd. ** road name changes to Neuse Rd.*	SR-1804	6

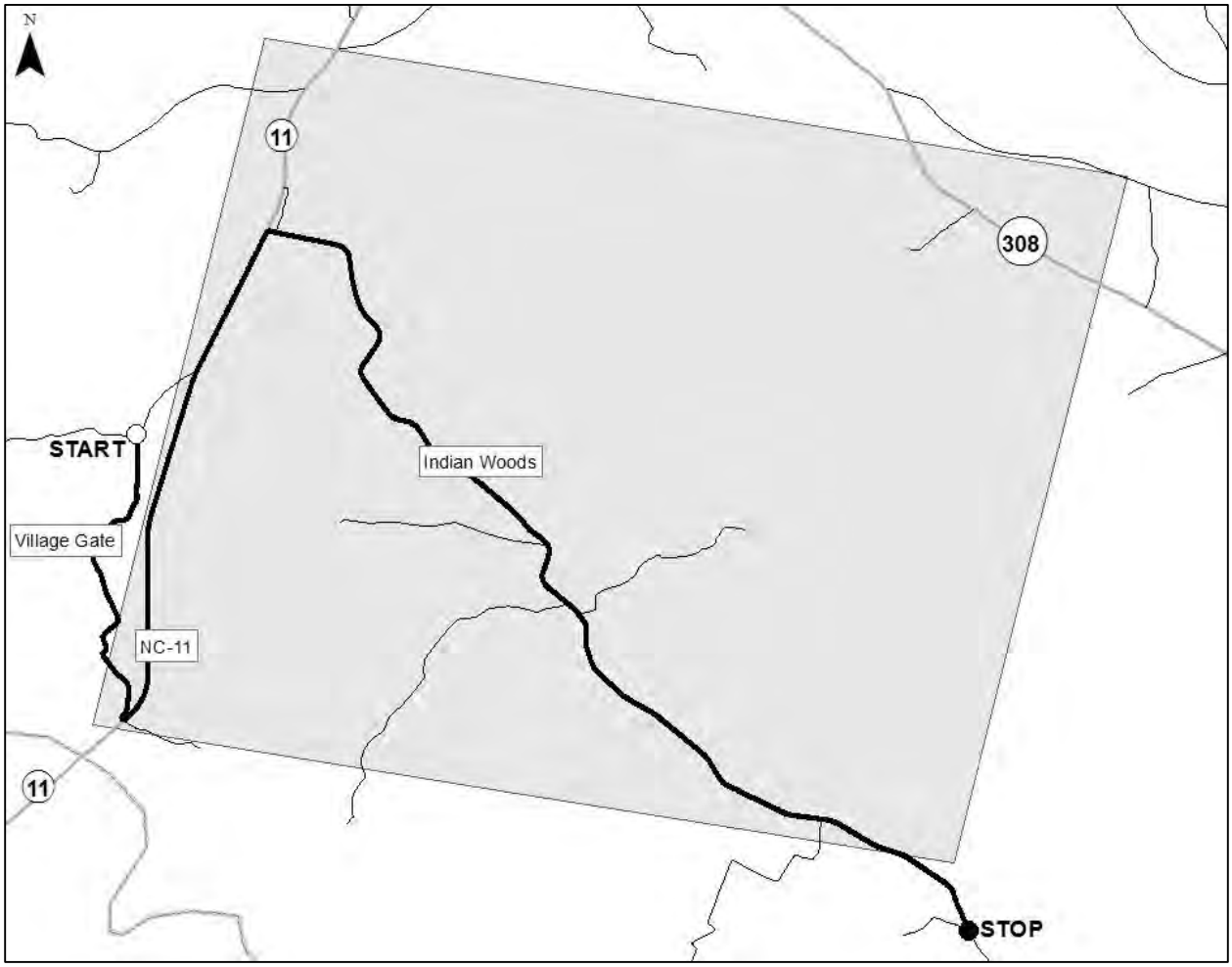
Grid 27 – Dover



Grid 29 – Windsor – Bertie County

	Latitude	Longitude	Intersection
Start	36.053707	-77.208575	Village Gate Rd. (SR-1127) and State Road 1126
Stop	35.990810	-77.102483	Indian Woods Rd. and Rascoe Club House Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Straight	T No stop sign	Village Gate Rd. ** gravel road*	SR-1127	2.7
Left	T Stop sign	NC-11 North	NC-11	4.4
Right	T No stop sign	Indian Woods Rd.	SR-1108	8.7

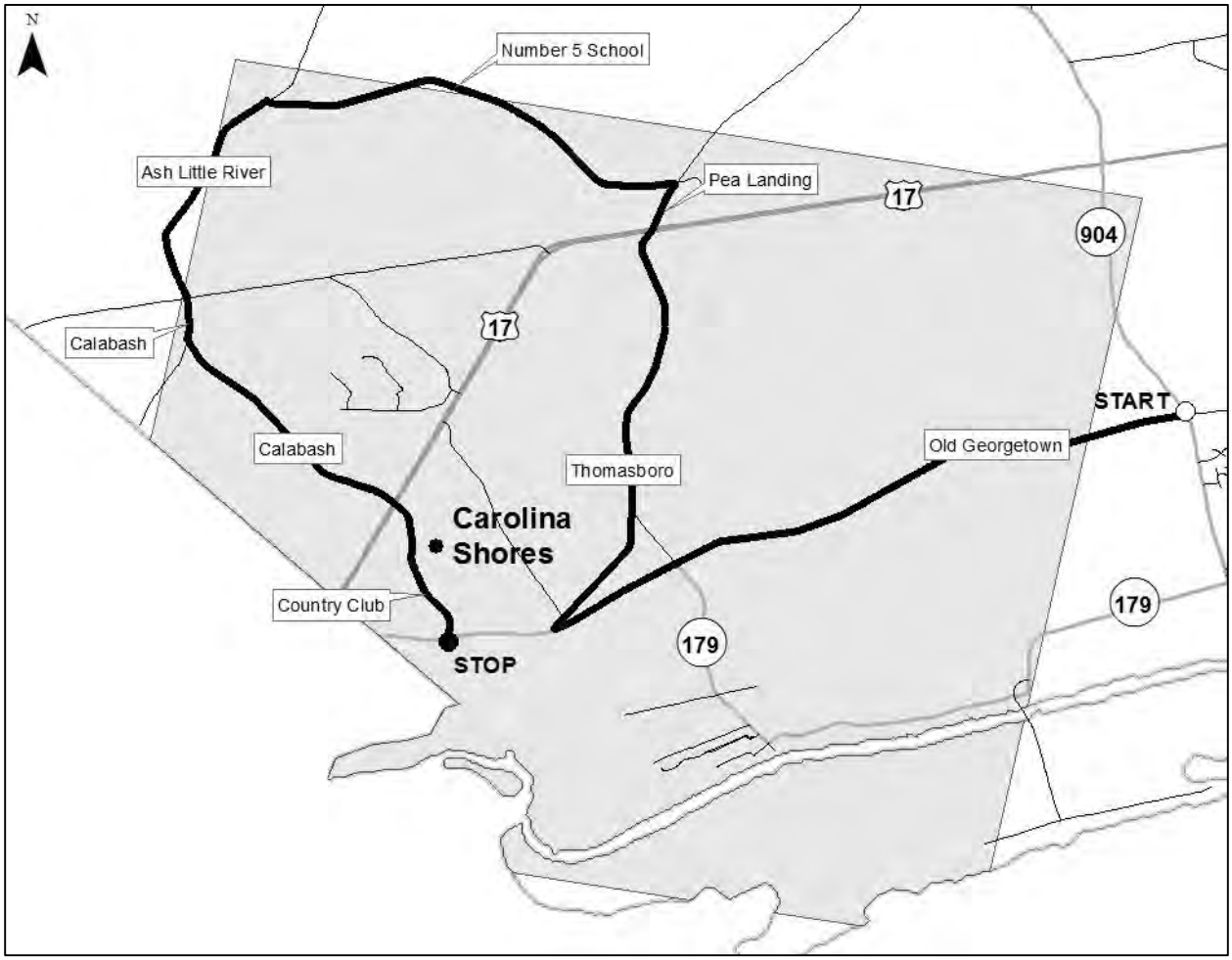


Grid 31 – Carolina Shores – Brunswick County

	Latitude	Longitude	Intersection
Start	33.916612	-78.492829	Seaside Rd. SW and Old Georgetown Rd. SW
Stop	33.890635	-78.579428	Country Club Rd. and Beach Dr. SW

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	+ Traffic light	Old Georgetown Rd. SW ** road name changes to Beach Dr. SW*	NC-179	4.7
Right	Y No stop sign	Thomasboro Rd. SW	SR-1165	3.5
Straight	+ Traffic light	Pea Landing Rd. NW	SR-1304	0.4
Left	T No stop sign	Number 5 School Rd. NW	SR-1305	3.1
Left	T Stop sign	Ash Little River Rd. NW	SR-1300	1.9
Straight	+ Stop sign	Calabash Rd. NW	SR-1300	0.3
Left	Y No stop sign	Calabash Rd. NW	SR-1300	2
Straight	+ Traffic light	Country Club Rd.	SR-1168	1.2

Grid 31 – Carolina Shores

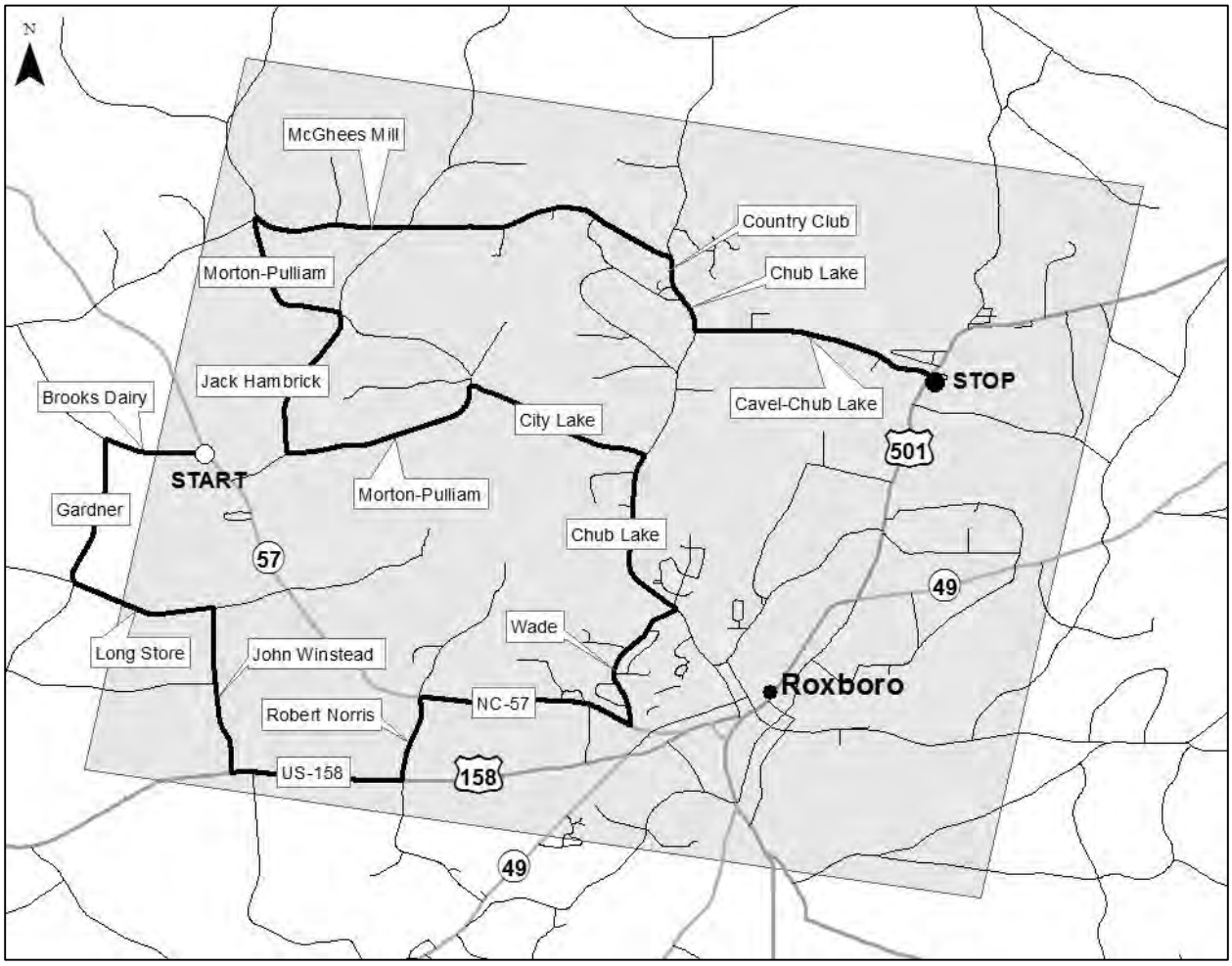


Grid 1 – Roxboro - Person County

	Latitude	Longitude	Intersection
Start	36.427826	-79.053308	Brooks Dairy Rd. and NC-57 North
Stop	36.437555	-78.963426	Cavel-Chub Lake Rd. and US-501 N

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T No stop sign	Brooks Dairy Rd.	SR-1309	0.7
Left	T No stop sign	Gardner Rd.	SR-1305	1.3
Left	+ Stop sign	Long Store Rd.	SR-13433	1
Right	T No stop sign	John Winstead Rd.	SR-1307	1.4
Left	T Stop sign	Leasburg Rd. ** if necessary drive normal highway speed*	US-158 E	1.2
Left	T No stop sign	Robert Norris Rd.	SR-1308	0.7
Right	+ Stop sign	Semora Rd. ** if necessary drive normal highway speed*	NC-57 S	1.5
Left	T No stop sign	Wade Rd.	SR-1346	1.2
Left	T Stop sign	Chub Lake Rd.	SR-1333	1.5
Left	T No stop sign	City Lake Rd.	SR-1336	1.4
Left	T Stop sign	Morton-Pulliam Rd.	SR-1342	1.5
Right	T No stop sign	Jack Hambrick Rd.	SR-1339	1.3
Left	+ Stop sign	Morton-Pulliam Rd.	SR-1336	1.1
Right	T Stop sign	Concord-Ceffo Rd.	SR-1337	< 0.1 miles
Right	T Stop sign	McGhees Mill Rd. ** road name change to Community House Rd.*	SR-1337 *SR-1371*	2.4 *0.6*
Right	T Stop sign	Country Club Rd.	SR-1333	0.2
Left	T Stop sign	Chub Lake Rd.	SR-1333	0.3
Left	T No stop sign	Cavel-Chub Lake Rd.	SR-1351	1.7

Grid 1 – Roxboro

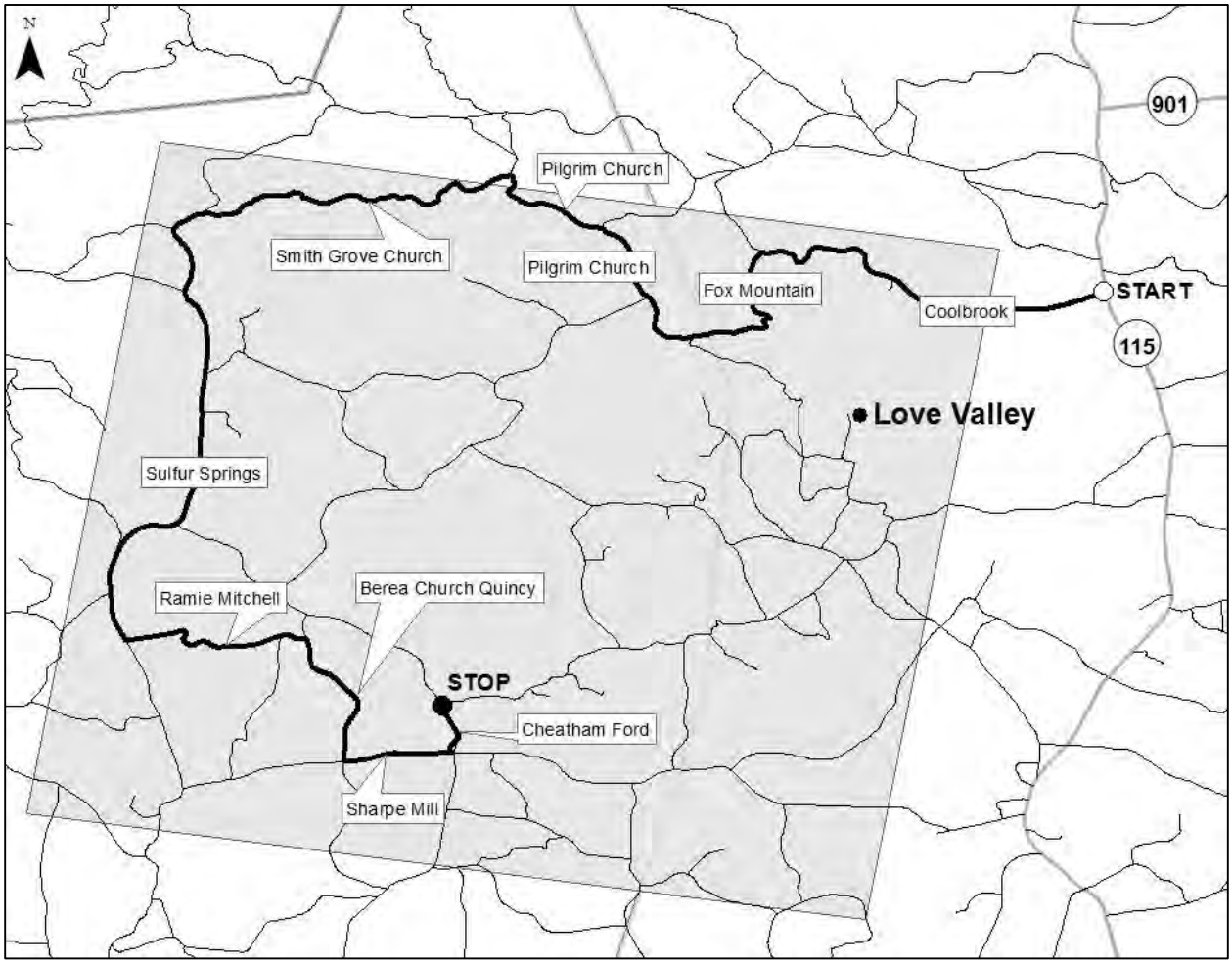


Grid 9 – Love Valley – Iredell/Alexander County

	Latitude	Longitude	Intersection
Start	36.005929	-80.956342	NC-115 and Coolbrook Rd.
Stop	35.951967	-81.043378	Center Church Rd. and Cheatham Ford Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T No stop sign	Coolbrook Rd.	SR-1595	2.9
Left	T No stop sign	Fox Mountain Rd. ** starts as gravel road* ** at 1.4 miles paved road, road name and number changes to Pilgrim Church Rd. (SR-1449)* ** at 2 miles the road changes back to gravel*	SR-1600	3.4
Left	Y No stop sign	Pilgrim Church Rd. ** 2nd Pilgrim Church Rd. on left, why there are 2 roads with the same name??*	SR-1447	1
Left	T Stop sign	Smith Grove Church Rd. ** gravel road changes to paved road*	SR-1443	2.7
Left	T Stop sign	Sulfur Springs Rd.	SR-1001	4.4
Left	T No stop sign	Ramie Mitchell Rd.	SR-1458	1.5
Right	T Stop sign	Berea Church Quincy Rd.	SR-1460	1.3
Left	+ Stop sign	Sharpe Mill Rd.	SR-1461	0.8
Left	+ Stop sign	Cheatham Ford Rd.	SR-1456	0.5

Grid 9 – Love Valley

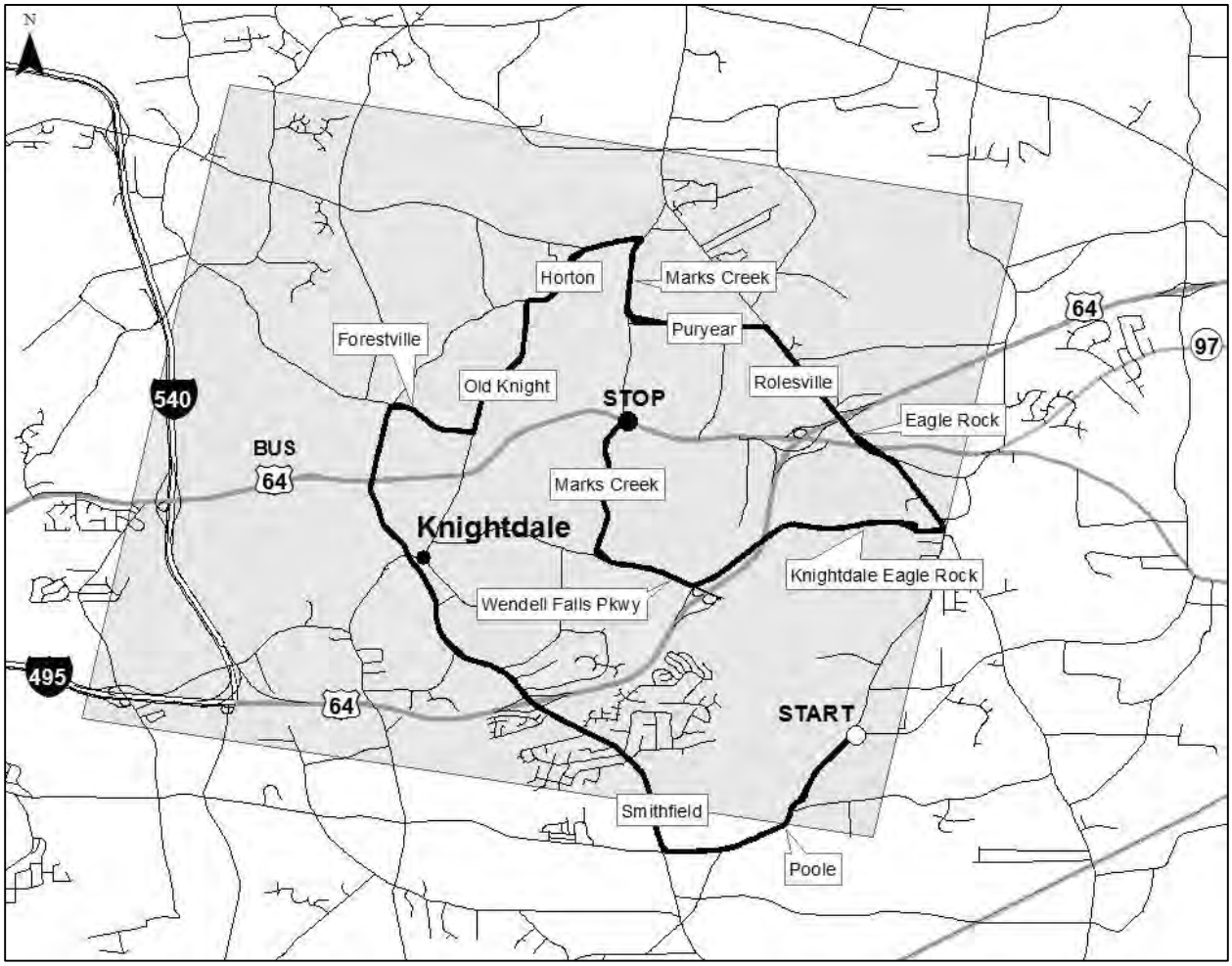


Grid 12 – Knightdale – Wake County

	Latitude	Longitude	Intersection
Start	35.762444	-78.421354	Martin Pond Rd. and Poole Rd.
Stop	35.806463	-78.452704	US-64 BUS and Marks Creek Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	Y Stop sign	Poole Rd.	SR-1007	2
Right	+ Traffic light	Smithfield Rd.	SR-2233	5.2
Right	+ Traffic light	Forestville Rd.	SR-2049	0.7
Left	T Stop sign	Old Knight Rd.	SR-2232	1.4
Right	T Stop sign	Horton Rd. ** at 0.8 mile there is a Y-intersection stay to the right the road number changes to SR-2215*	SR-2231	1.2
Right	Y Stop sign	Marks Creek Rd.	SR-2234	0.8
Left	T No stop sign	Puryear Rd.	SR-2235	1.1
Right	T Stop sign	Rolesville Rd.	SR-1003	1.3
Straight	+ Traffic light	Eagle Rock Rd. ** at 0.3 mile there is a Y-intersection veer right staying on Eagle Rock Rd.*	SR-1003	1.1
Right	T No stop sign	Knightdale Eagle Rock Rd. ** at 0.2 mile turn right and at 0.3 mile turn left staying on Knightdale Eagle Rock Rd.*	SR-2501	2.2
Right	+ Traffic light	Wendell Falls Parkway	SR-2501	0.9
Right	T No stop sign	Marks Creek Rd.	SR-2500	1.3

Grid 12 – Knightdale

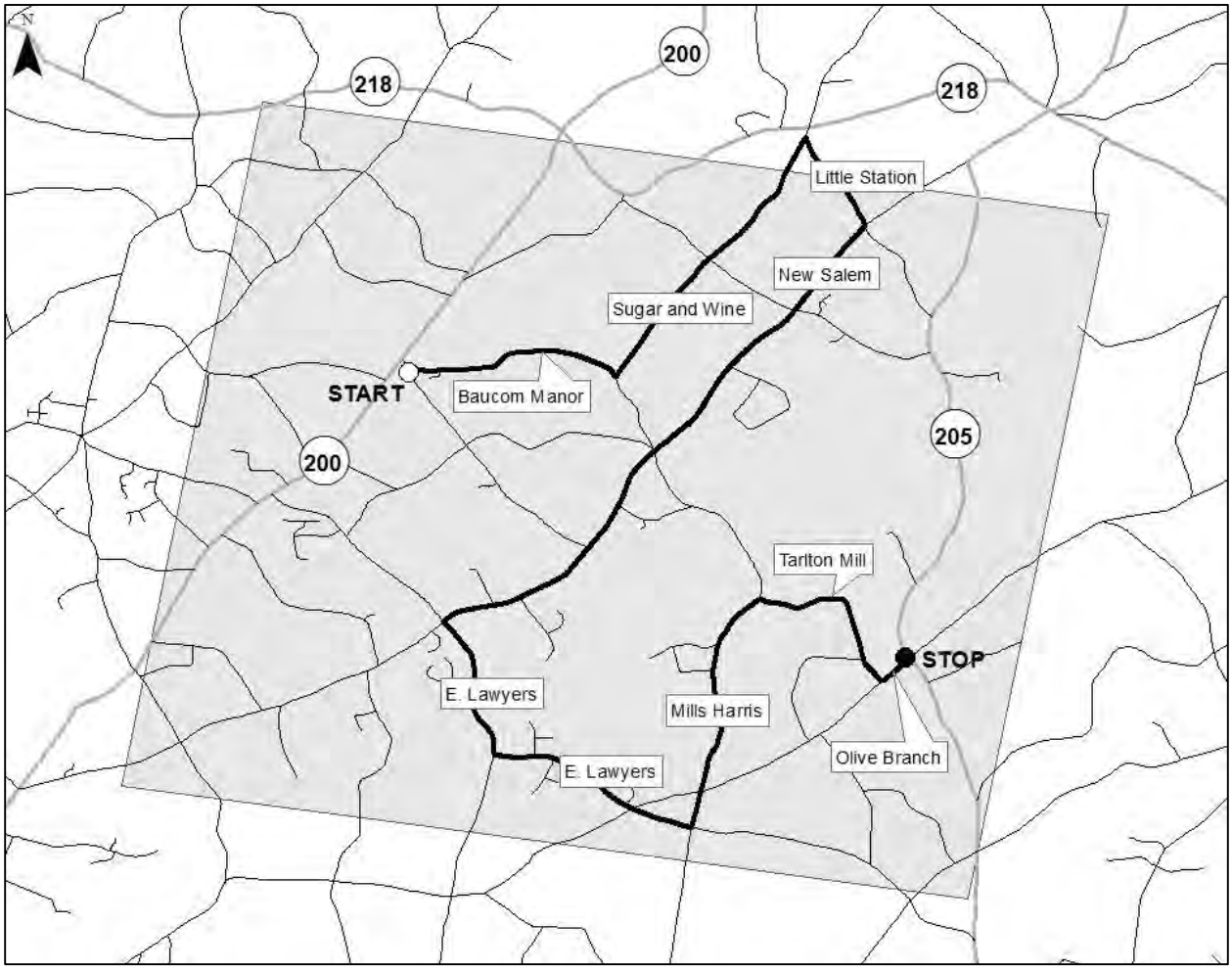


Grid 15 – Monroe – Union County

	Latitude	Longitude	Intersection
Start	35.097884	-80.451743	Baucom Manor Rd. and Watson Church Rd.
Stop	35.060056	-80.387801	Fairfield Baptist Church parking lot, right off of Olive Branch Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	Y No stop sign	Baucom Manor Rd.	SR-1652	1.6
Left	T Stop sign	Sugar and Wine Rd.	SR-1649	2.6
Right	T No stop sign	Little Station Rd.	SR-1659	0.9
Right	+ Stop sign	New Salem Rd.	SR-1627	4.8
Left	+ No stop sign	E. Lawyers Rd.	SR-1631	1.3
Left	T No stop sign	E. Lawyers Rd.	SR-1632	1.6
Left	+ Stop sign	Mills Harris Rd.	SR-1645	2.2
Right	T No stop sign	Tarlton Mill Rd.	SR-1649	1.4
Left	T Stop sign	Olive Branch Rd.	SR-1006	0.2

Grid 15 – Monroe

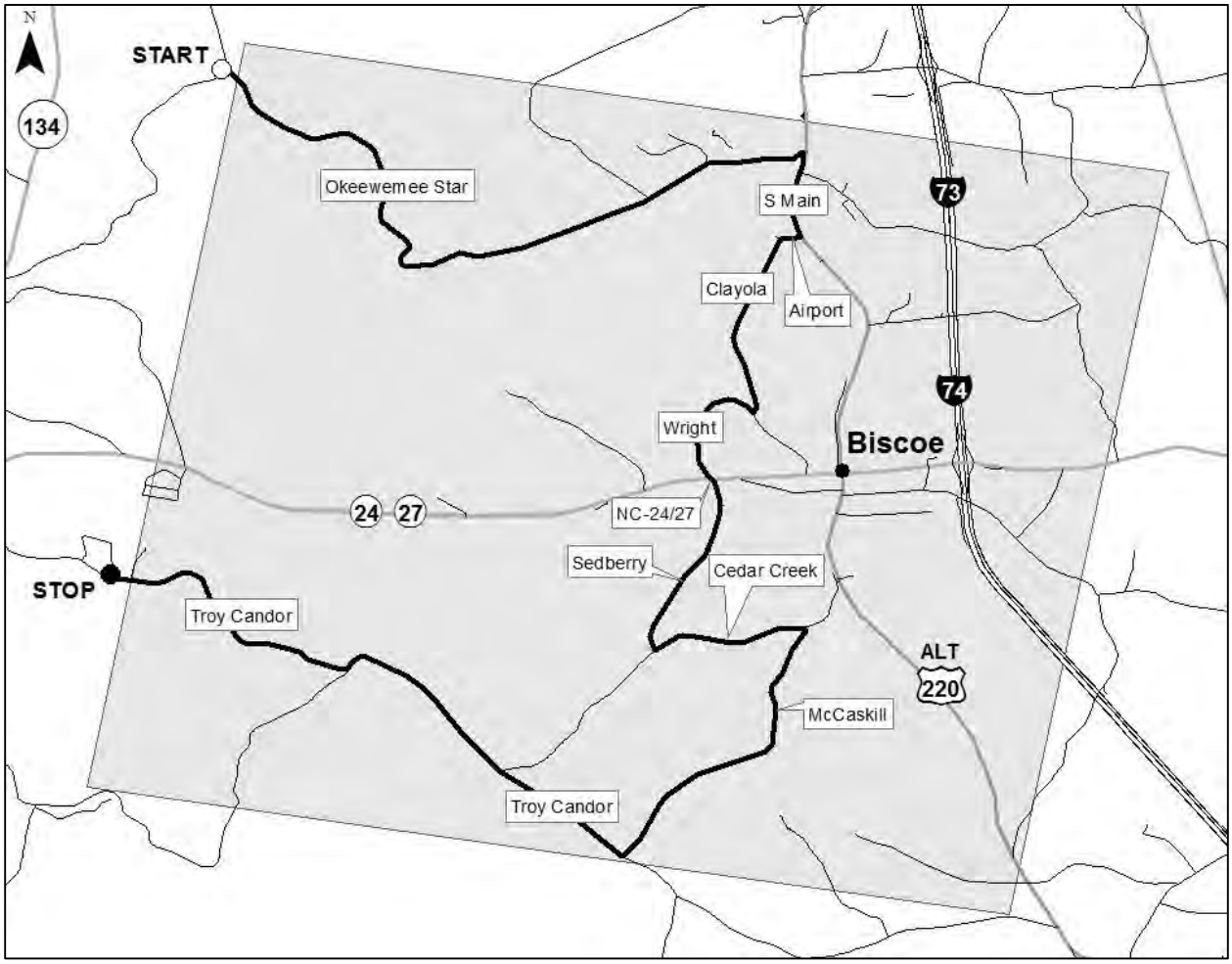


Grid 20 – Biscoe – Montgomery County

	Latitude	Longitude	Intersection
Start	35.407387	-79.852706	Okeewemee Rd. and Okeewemee Star Rd.
Stop	35.347305	-79.866537	2 miles after making last turn onto Troy Candor Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	Y No stop sign	Okeewemee Star Rd.	SR-1340	5.2
Right	+ Traffic light	S Main St.	US-220 BUS	0.7
Right	T No stop sign	Airport Rd.	SR-1376	0.1
Left	T No stop sign	Clayola Dr.	SR-1396	1.5
Right	T Stop sign	Wright Rd.	SR-1337	1
Left	T Stop sign	NC-24/ 27	NC-24/ 27	<0.1
Right	T No stop sign	Sedberry Rd.	SR-1556	1.5
Left	T Stop sign	Cedar Creek Rd.	SR-1557	1
Right	Y Stop sign	McCaskill Rd.	SR-1558	2.4
Right	T Stop sign	Troy Candor Rd.	SR-1519	2.5
Right	Y No stop sign	Troy Candor Rd.	SR-1554	2

Grid 20 – Biscoe

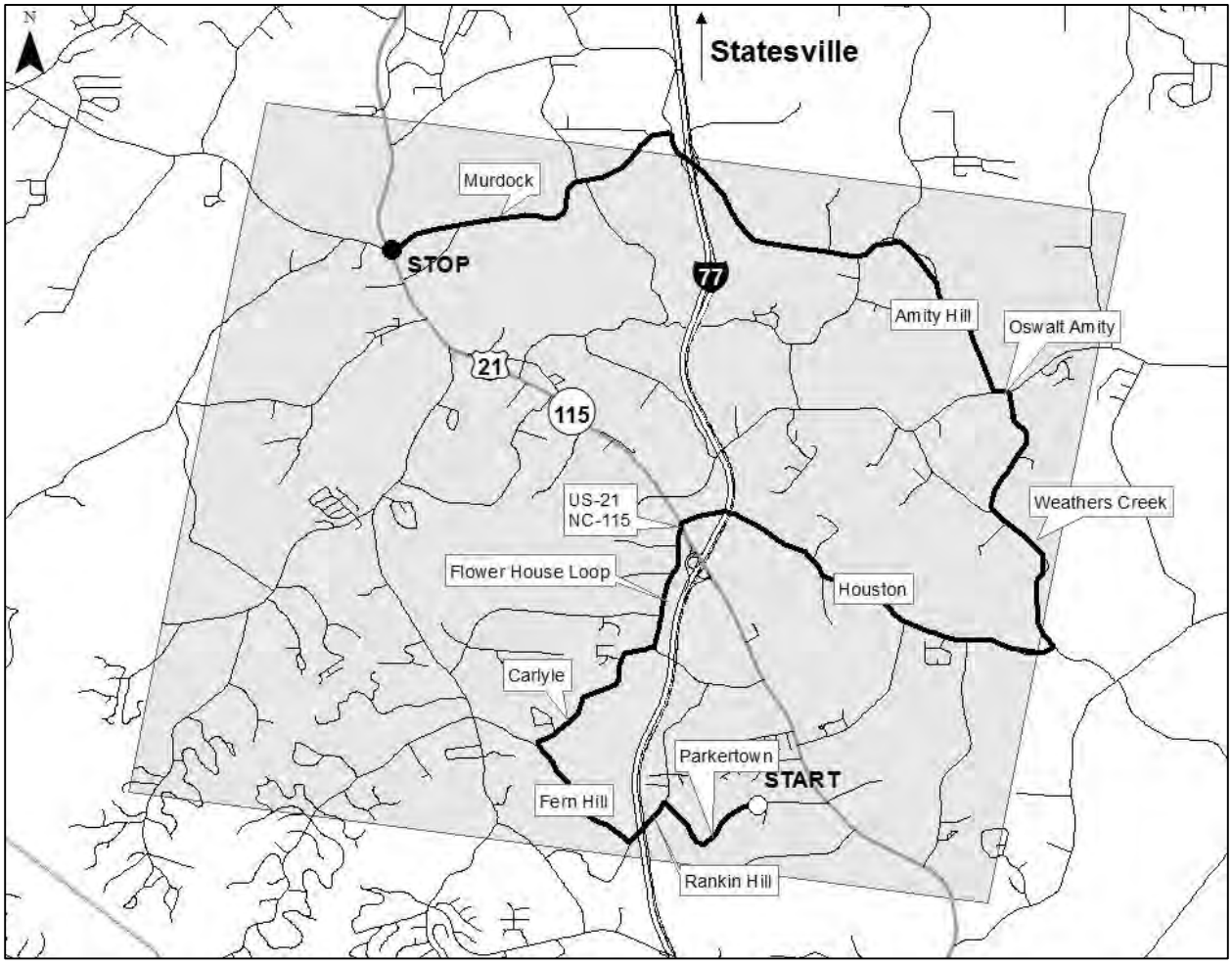


Grid 21 – Statesville – Iredell County

	Latitude	Longitude	Intersection
Start	35.642915	-80.851181	US-21/ NC-115 and Parkertown Rd. drive 0.5 mile then start
Stop	35.714402	-80.895755	Murdock Rd. and US-21/ NC-115 (traffic light)

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T No stop sign	Parkertown Rd. ** sections of Parkertown Rd. are gravel*	SR-1310	1.1
Left	T Stop sign	Rankin Hill Rd.	SR-1311	0.4
Right	T No stop sign	Fern Hill Rd.	SR-1300	1.1
Right	T No stop sign	Carlyle Rd.	SR-1313	1.3
Left	T Stop sign	Flower House Loop	SR-1312	1.1
Left	T Stop sign	US-21/ NC-115	US-21/ NC-115	<0.1
Right	T No stop sign	Houston Rd.	SR-2375	3.1
Left	T Stop sign	Weathers Creek Rd.	SR-2379	1.4
Right	Y No stop sign	Weathers Creek Rd.	SR-2379	1.2
Left	T Stop sign	Oswalt Amity Rd.	SR-1001	0.1
Right	T No stop sign	Amity Hill Rd.	SR-2342	3.8
Left	T No stop sign	Murdock Rd.	SR-2350	2.5

Grid 21 – Statesville

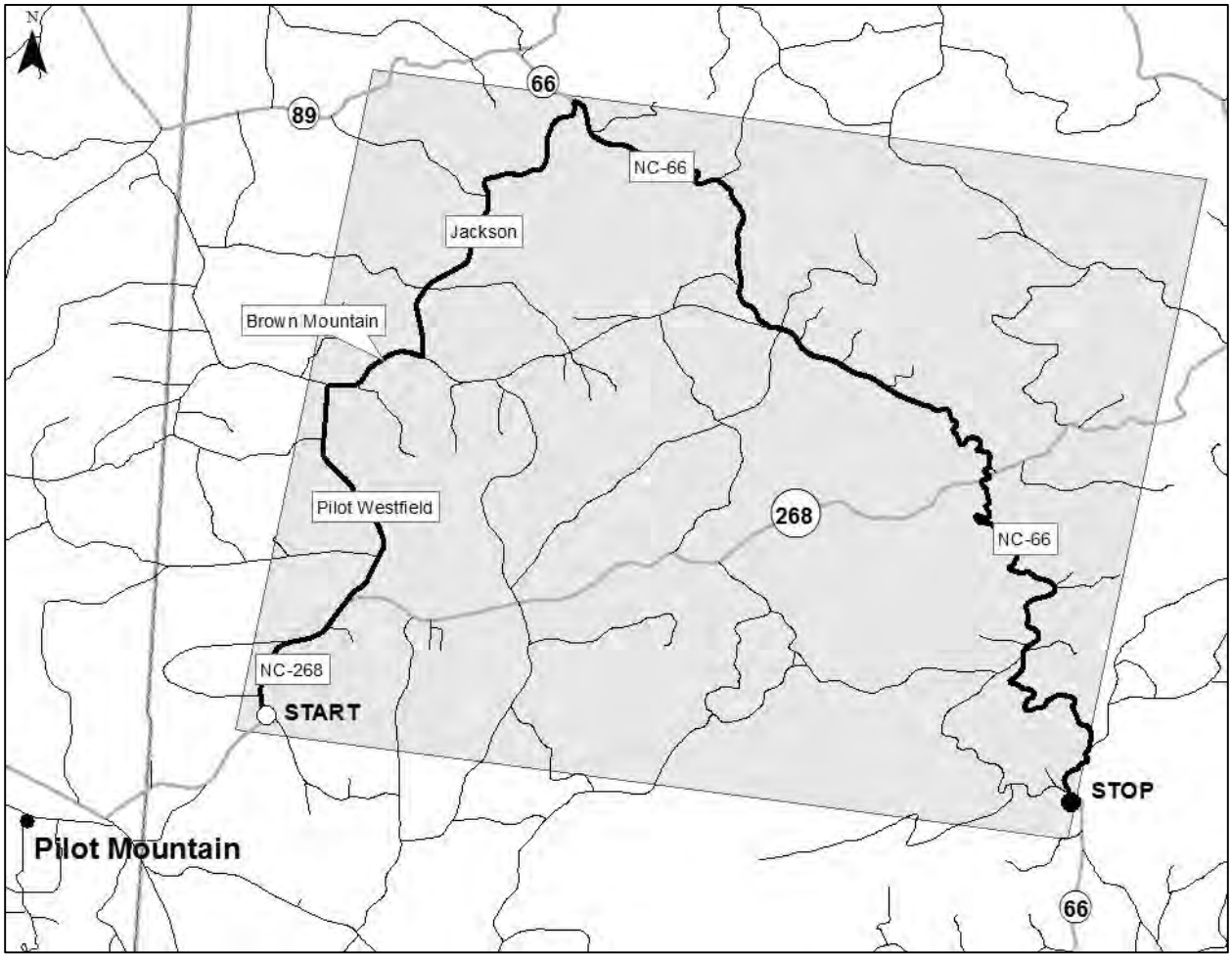


Grid 24 – Pilot Mountain – Stokes County

	Latitude	Longitude	Intersection
Start	36.397382	-80.432849	NC-268 and Venable Rd.
Stop	36.386767	-80.325402	NC-66 and Taylor Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Straight	T No stop sign	NC-268	NC-268	1.3
Left	Y No stop sign	Pilot Westfield Rd.	SR-1199	2.2
Right	T Stop sign	Brown Mountain Rd.	SR-1210	0.8
Left	T No stop sign	Jackson Rd. ** SR-1212 changes to SR-1214*	SR-1212	3
Right	T Stop sign	NC-66	NC-66	10.2

Grid 24 – Pilot Mountain

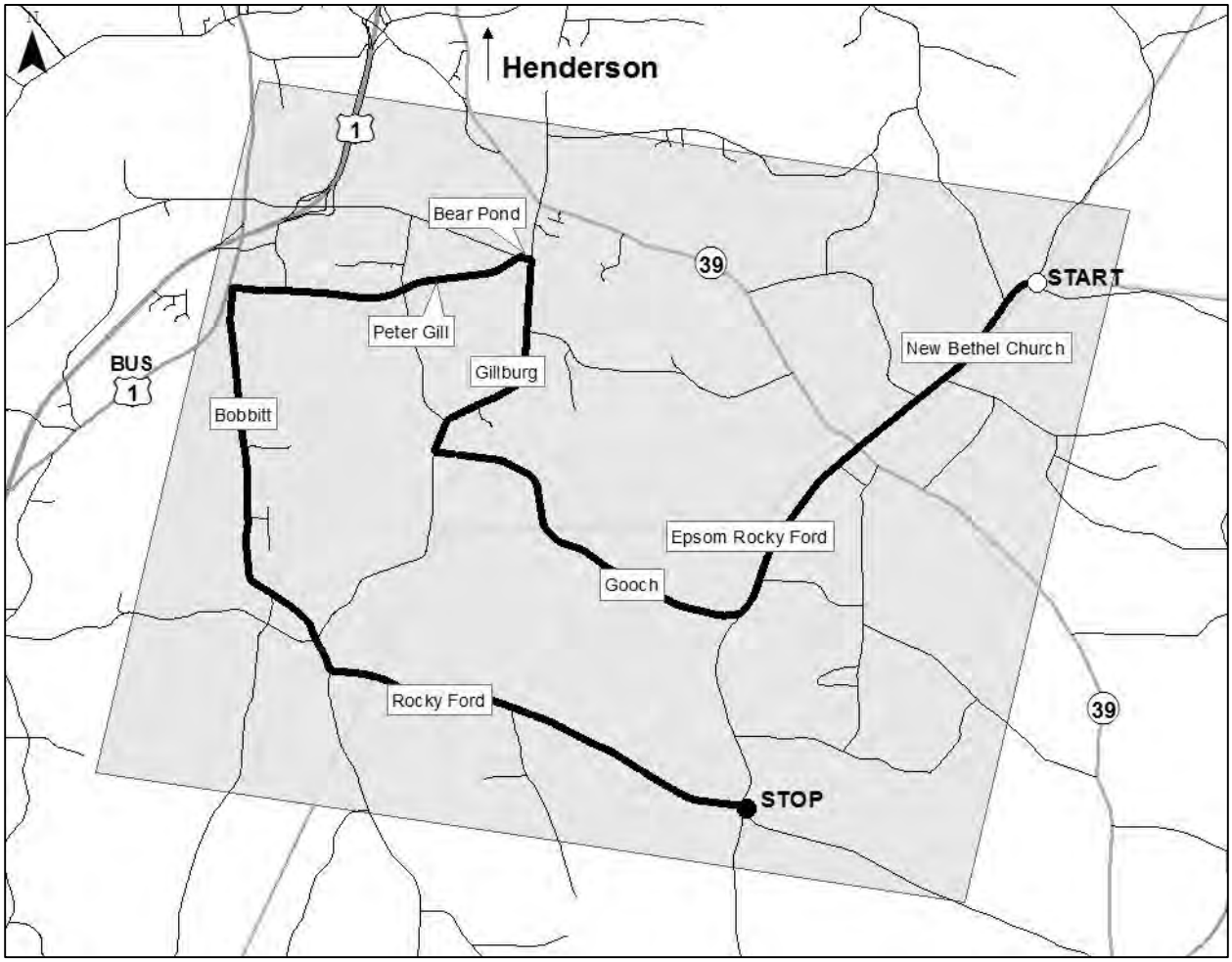


Grid 26 – Henderson – Vance/Rankin County

	Latitude	Longitude	Intersection
Start	36.266187	-78.306902	New Bethel Church Rd. and Cheeks Quarter Rd.
Stop	36.199786	-78.343015	Rocky Ford Rd. and Sims Bridge Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T Stop sign	New Bethel Church Rd.	SR-1523	1.9
Straight	+ Stop sign	Epsom Rocky Ford Rd. ** after crossing county line SR-1523 changes to SR-1003*	SR-1523	1.7
Right	T No stop sign	Gooch Rd. ** after crossing county line Gooch Rd. (SR-1252) changes to Julian Smith Rd. (SR-1542)*	SR-1252	2.8
Right	T Stop sign	Gillburg Rd.	SR-1519	2
Left	T No stop sign	Bear Pond Rd.	SR-1115	<0.1
Left	T No stop sign	Peter Gill Rd.	SR-1546	2.1
Left	T No stop sign	Bobbitt Rd.	SR-1549	3.6
Left	T No stop sign	Rocky Ford Rd. ** after crossing county line SR-1550 changes to SR-1239*	SR-1550	3.2

Grid 26 – Henderson

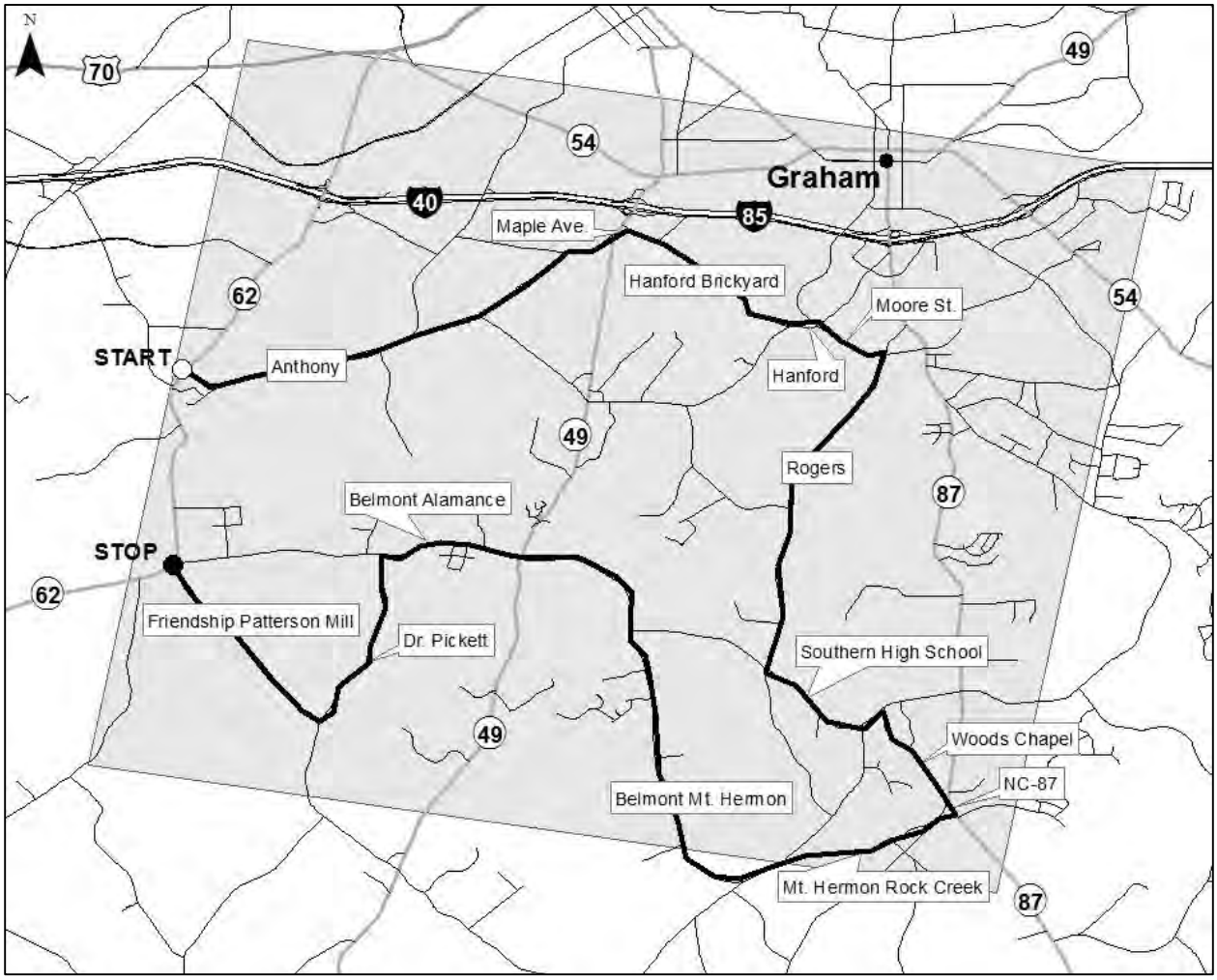


Grid 30 – Graham – Alamance County

	Latitude	Longitude	Intersection
Start	36.043614	-79.485523	NC-62 and Anthony Rd.
Stop	36.020111	-79.487039	Friendship Patterson Mill Rd. and NC-62

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T No stop sign	Anthony Rd.	SR-1147	0.3
Left	Y No stop sign	Anthony Rd.	SR-1148	1.3
Right	Y No stop sign	Anthony Rd.	SR-1148	1.3
Right	Y No stop sign	Anthony Rd.	SR-1148	0.4
Left	T Traffic light	Maple Ave.	NC-49	<0.1
Right	+ Traffic light	Hanford Brickyard Rd.	SR-2304	1.4
Left	Y No stop sign	Hanford Rd.	SR-2312	0.2
Right	+ Traffic light	Moore St.	SR-2433	0.5
Right	+ Traffic light	Rogers Rd.	SR-2309	2.8
Left	+ Traffic light	Southern High School Rd.	SR-2387	1.1
Right	T No stop sign	Woods Chapel Rd.	SR-2324	0.9
Right	T Stop sign	NC-87	NC-87	0.1
Right	+ No stop sign	Mt. Hermon Rock Creek Rd.	SR-2327	1.6
Right	Y No stop sign	Belmont Mt. Hermon Rd.	SR-1136	3.6
Straight	+ Traffic light	Belmont Alamance Rd.	SR-1136	0.9
Left	T No stop sign	Dr. Pickett Rd.	SR-1131	1.5
Right	Y Stop sign	Friendship Patterson Mill Rd.	SR-1130	1.7

Grid 30 – Graham

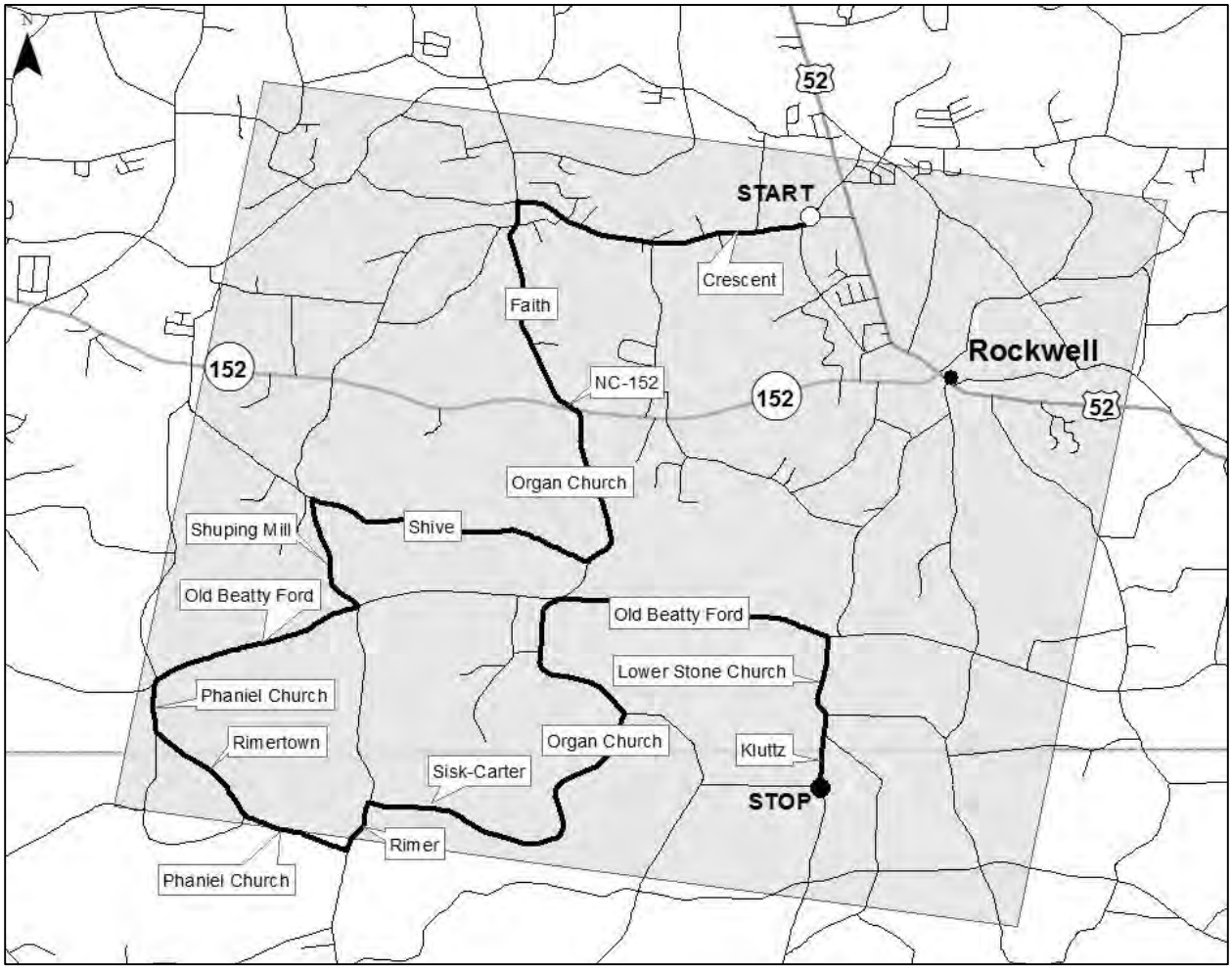


Grid 32 – Rockwell – Rowan County

	Latitude	Longitude	Intersection
Start	35.571544	-80.423104	US-52 N and Crescent Rd.
Stop	35.490163	-80.423719	Kluttz Rd. and Emanuel Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	5-way No stop sign	Crescent Rd.	SR-2319	2.5
Left	+ Stop sign	Faith Rd.	SR-1006	1.8
Left	T Stop sign	NC-152	NC-152	<0.1
Right	T No stop sign	Organ Church Rd.	SR-1006	1.3
Right	T No stop sign	Shive Rd.	SR-2564	2
Left	Y No stop sign	Shuping Mill Rd.	SR-2559	1
Right	+ Stop sign	Old Beatty Ford Rd.	SR-1221	1.5
Left	+ No stop sign	Phaniel Church Rd.	SR-2569	0.4
Left	Y No stop sign	Rimertown Rd.	SR-2568	0.9
Left	Y No stop sign	Phaniel Church Rd.	SR-2433	0.8
Left	T Stop sign	Rimer Rd.	SR-2429	0.4
Right	T No stop sign	Sisk-Carter Rd. ** at county line road number changes from SR-2434 to SR-2566*	SR-2434	2.6
Left	T Stop sign	Organ Church Rd.	SR-1006	1.3
Right	T Stop sign	Old Beatty Ford Rd.	SR-1221	1.9
Right	T No stop sign	Lower Stone Church Rd. ** at county line road number changes from SR-2464 to SR-2436*	SR-2464	2.1
Left	+ No stop sign	Kluttz Rd.	SR-2435	1.2

Grid 32 – Rockwell

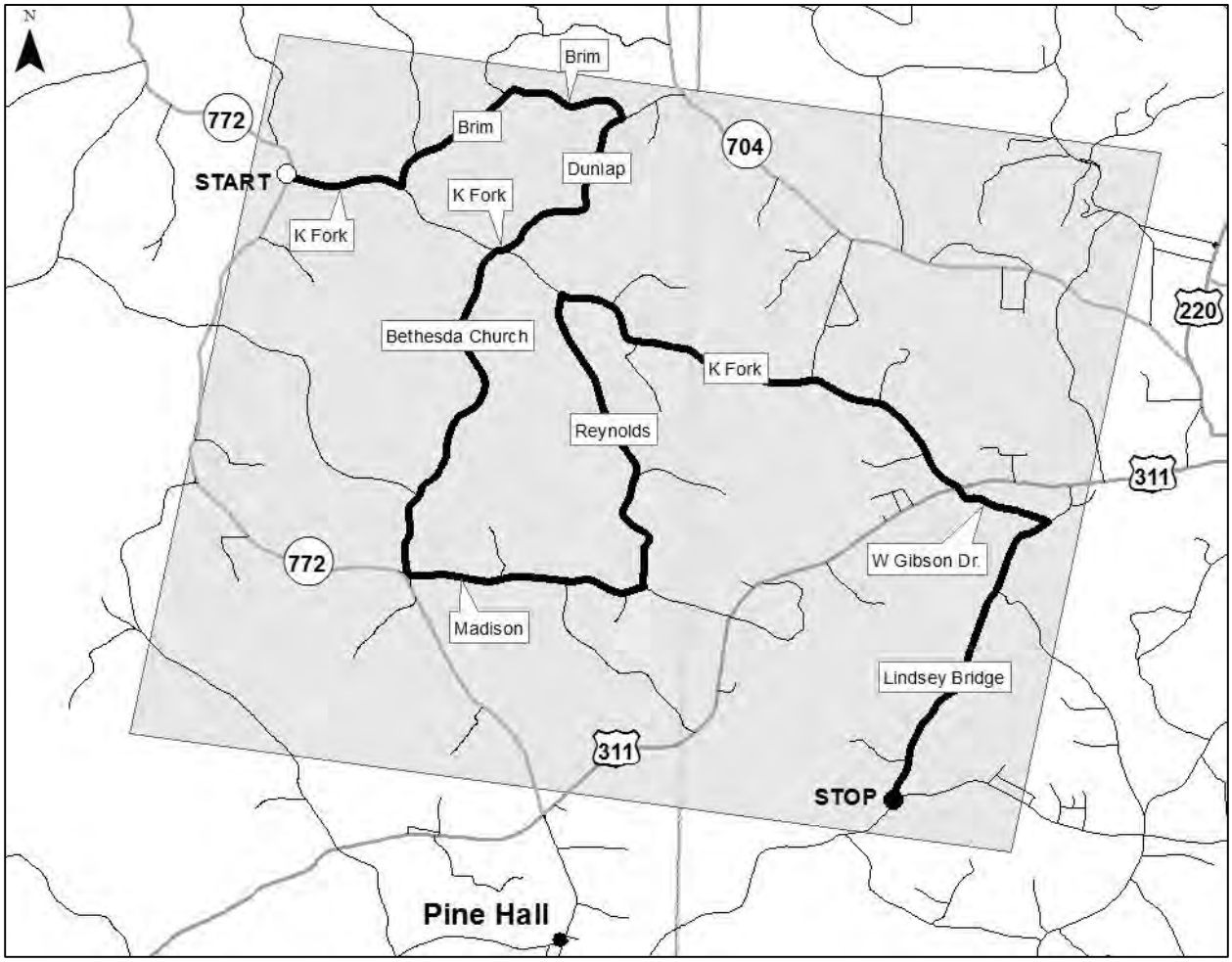


Grid 33 – Pine Hall – Stokes/Rockingham County

	Latitude	Longitude	Intersection
Start	36.420824	-80.083397	NC-772 and K Fork Rd.
Stop	36.343134	-80.007410	Lindsey Bridge Rd. and Eden Church Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T No stop sign	K Fork Rd.	SR-1686	0.8
Left	T No stop sign	Brim Rd.	SR-1693	1
Right	Y No stop sign	Brim Rd.	SR-1681	1
Right	T Stop sign	Dunlap Rd.	SR-1683	1.5
Right	T Stop sign	K Fork Rd.	SR-1686	<0.1
Left	T No stop sign	Bethesda Church Rd.	SR-1683	3.2
Left	+ Stop sign	Madison Rd.	SR-1729	1.7
Left	T No stop sign	Reynolds Rd.	SR-1688	2.7
Right	T Stop sign	K Fork Rd.	SR-1162	3.5
Straight	+ Traffic light	W Gibson Dr.	SR-1194	0.1
Left	Y No stop sign	W Gibson Dr.	SR-1194	0.6
Left	T Stop sign	Lindsey Bridge Rd.	SR-1138	3.2

Grid 33 – Pine Hall

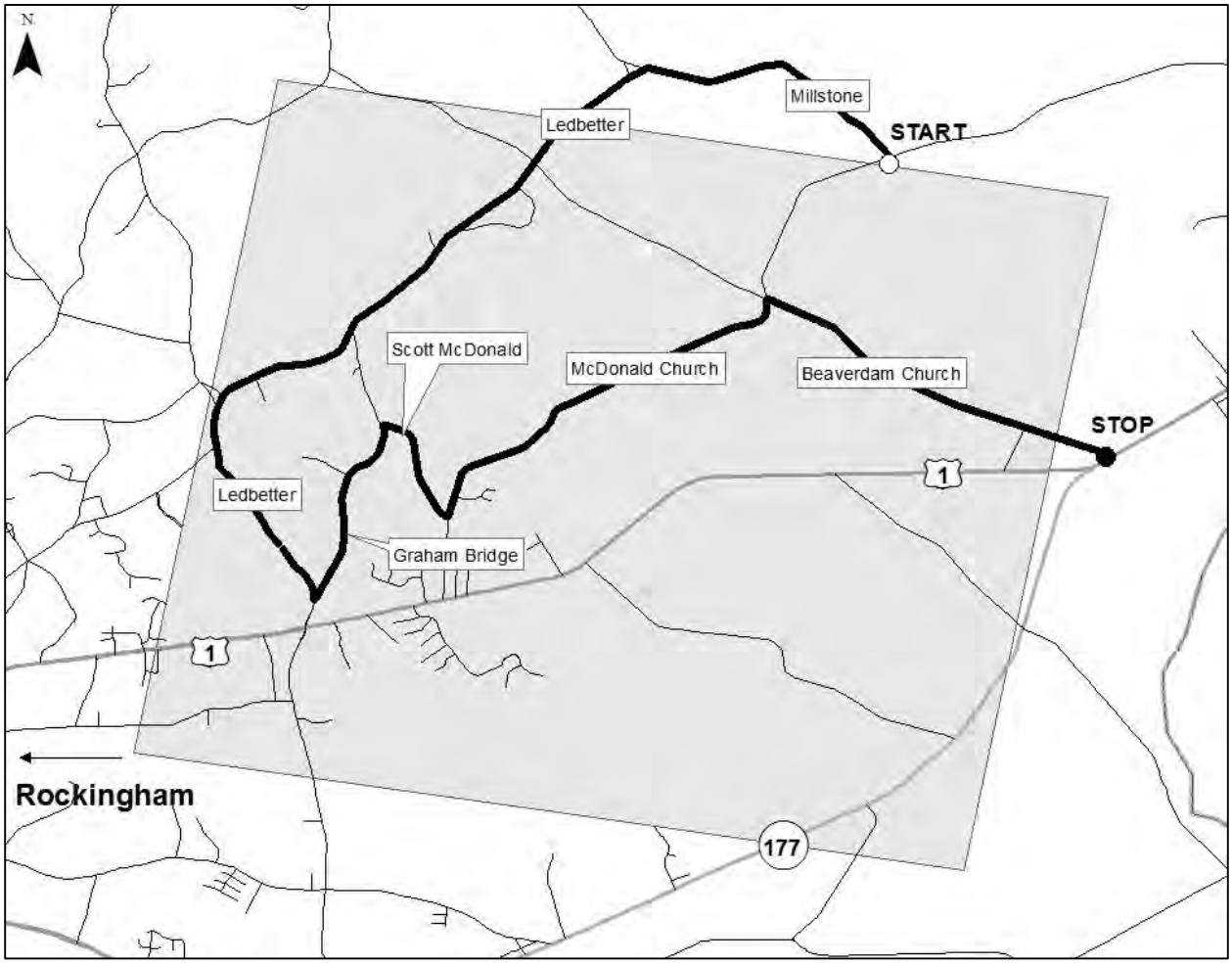


Grid 37 – Rockingham – Richmond County

	Latitude	Longitude	
Start	35.018382	-79.627483	McDonald Church Rd. and Millstone Rd.
Stop	34.978998	-79.598592	Beaverdam Church Rd. and US-1

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	T No stop sign	Millstone Rd.	SR-1487	2.2
Left	+ No stop sign	Ledbetter Rd.	SR-1442	6.5
Left	Y No stop sign	Graham Bridge Rd.	SR-1489	1.7
Right	T No stop sign	Scott McDonald Rd.	SR-1478	1
Left	T Stop sign	McDonald Church Rd.	SR-1475	3.3
Right	+ No stop sign	Beaverdam Church Rd.	SR-1486	3

Grid 37 – Rockingham

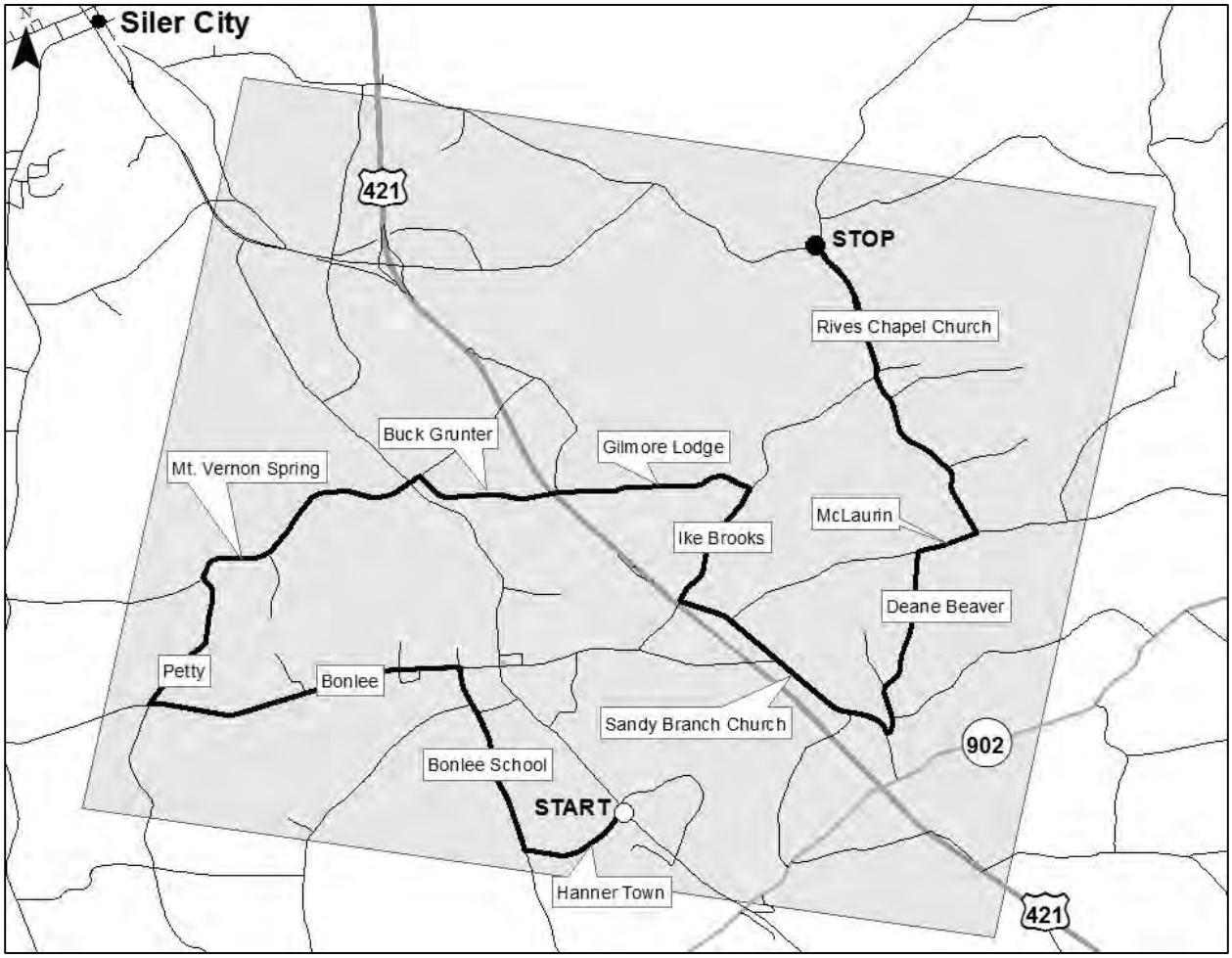


Grid 47 – Siler City – Chatham County

	Latitude	Longitude	Intersection
Start	35.628143	-79.400127	Old US-421 South and Hanner Town Rd.
Stop	35.696232	-79.375927	Rives Chapel Church Rd. and Alston Bridge Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T No stop sign	Hanner Town Rd.	SR-1142	0.8
Right	T Stop sign	Bonlee School Rd.	SR-1139	1.6
Left	+ Stop sign	Bonlee Rd.	SR-1005	2.2
Right	+ No stop sign	Petty Rd.	SR-1136	1.2
Right	T Stop sign	Mt. Vernon Spring Rd. ** after crossing Old US-421 road name changes to Foust Rd.*	SR-1134	1.9
Right	T No stop sign	Buck Grunter Rd.	SR-2119	1
Straight	+ Stop sign	Gilmore Lodge Rd.	SR-2119	1.4
Right	T Stop sign	Ike Brooks Rd.	SR-2120	1
Left	T No stop sign	Sandy Branch Church Rd.	SR-2207	1.8
Left	T No stop sign	Deane Beaver Rd.	SR-2180	1.6
Right	T Stop sign	McLaurin Rd.	SR-2175	0.4
Left	T No stop sign	Rives Chapel Church Rd.	SR-2170	2.6

Grid 47 – Siler City

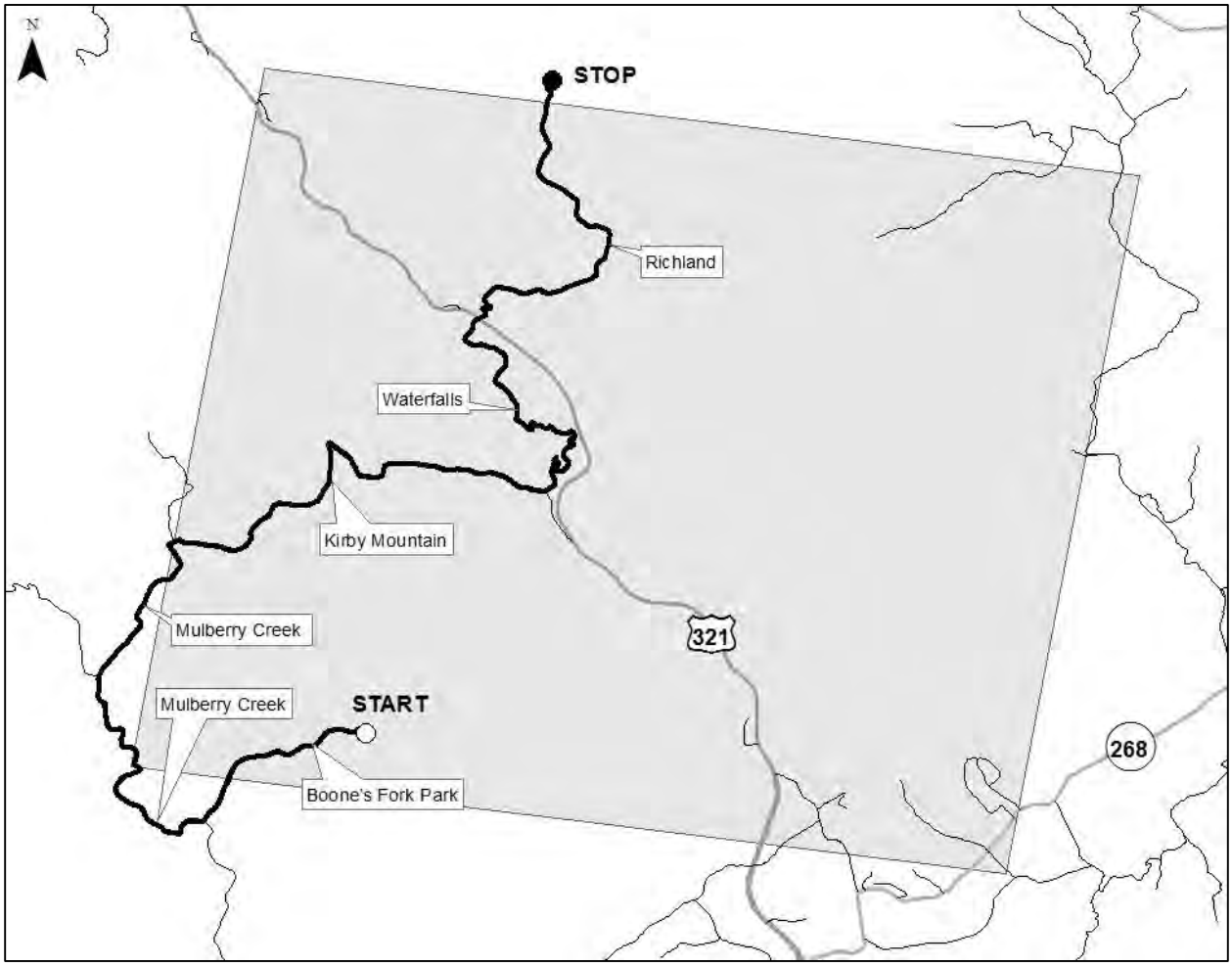


Grid 5 – Lenoir – Caldwell County

	Latitude	Longitude	Intersection
Start	36.010371	-81.622159	Boone's Fork primitive campground gate *nearest intersection is Mulberry Creek Rd. and Boone's Fork Park Rd.
Stop	36.091634	-81.597795	Hines Branch Rd. and Richland Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
--	--	Boone's Fork Park Rd. ** gravel road*	FR-2055	1.5
Right	T Stop sign	Mulberry Creek Rd.	SR-1368	2.2
Right	T No stop sign	Mulberry Creek Rd. ** gravel road*	SR-1369	1.5
Straight	T No stop sign	Kirby Mountain Rd.	SR-1370	3.6
Left	T No stop sign	Waterfalls Rd. ** sections of road are gravel*	SR-1371	2.7
Straight	+ Stop sign	Richland Rd.	SR-1372	3.5

Grid 5 – Lenoir

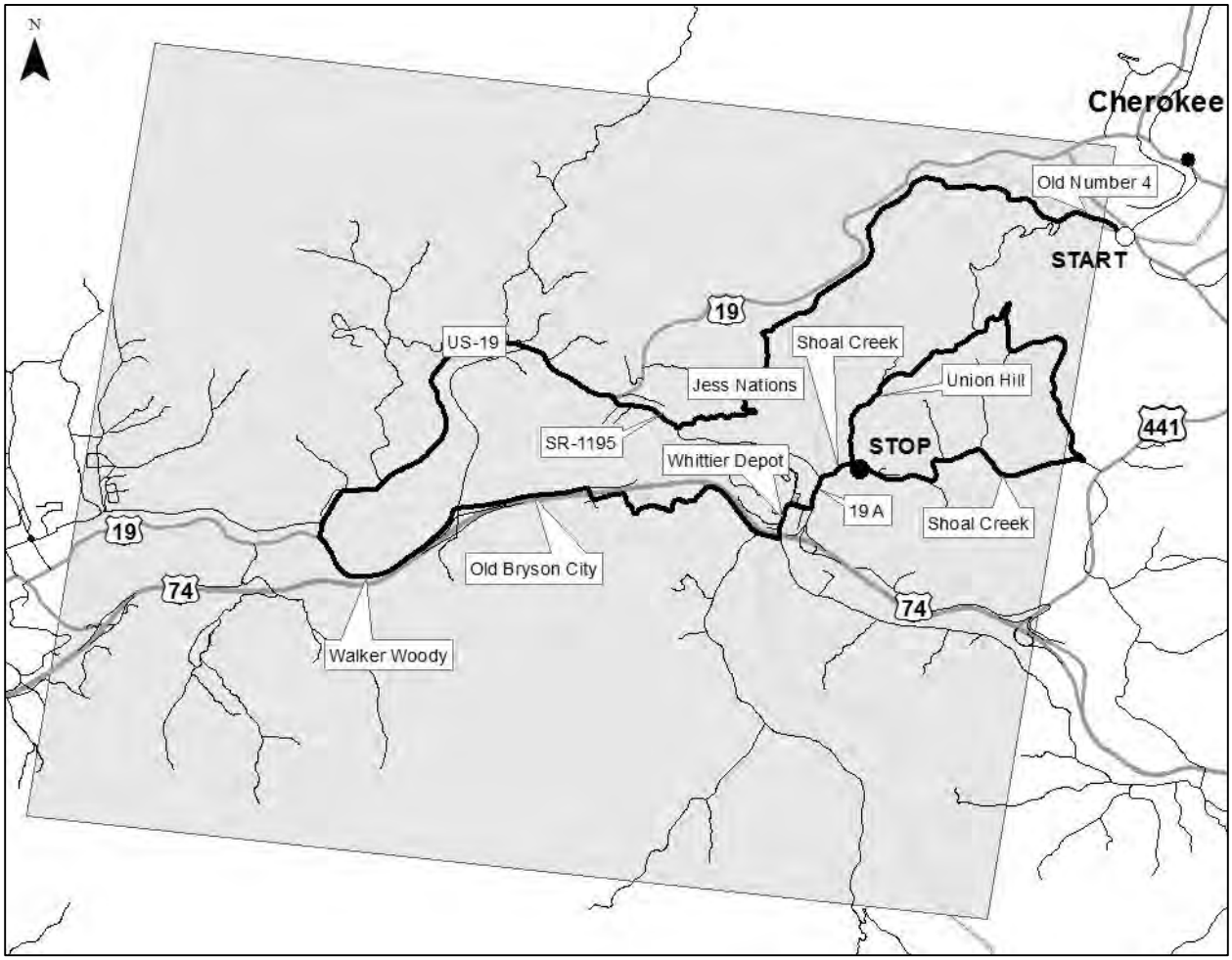


Grid 6 – Cherokee – Swain County

	Latitude	Longitude	Intersection
Start	35.466011	-83.321494	US-441 (heading south) and Old Number 4 Rd.
Stop	35.439306	-83.352917	Shoal Creek Rd. and Union Hill Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	+ No stop sign	Old Number 4 Rd.	SR-1236	2.8
Straight	T No stop sign	Jess Nations Rd. ** after 1.1 miles road changes to gravel*	SR-1236	1.8
Right	T Stop sign	State Road 1195 ** follow road right and cross bridge*	SR-1195	0.5
Left	T Stop sign	Ela Rd.	US-19 S	3.2
Left	T No stop sign	Walker Woody Rd.	SR-1168	1.3
Straight	+ Stop sign	Old Bryson City Rd.	SR-1192	0.9
Right	T No stop sign	Old Bryson City Rd.	SR-1192	<0.1
Left	T No stop sign	Old Bryson City Rd.	SR-1192	1.7
Left	T Stop sign	Whittier Depot St.	SR-1175	0.3
Right	T No stop sign	Whittier Depot Rd. ** cross railroad tracks*	SR-1175	0.1
Left	T Stop sign	19 A	SR-1531	0.3
Right	T No stop sign	Shoal Creek Rd.	SR-1416	0.2
Left	T No stop sign	Union Hill Rd.	SR-1411	1.9
Right	T No stop sign	Union Hill Rd.	SR-1411	0.4
Left	Y No stop sign	Union Hill Rd.	SR-1411	1.4
Right	T Stop sign	Shoal Creek Rd.	SR-1416	1.9

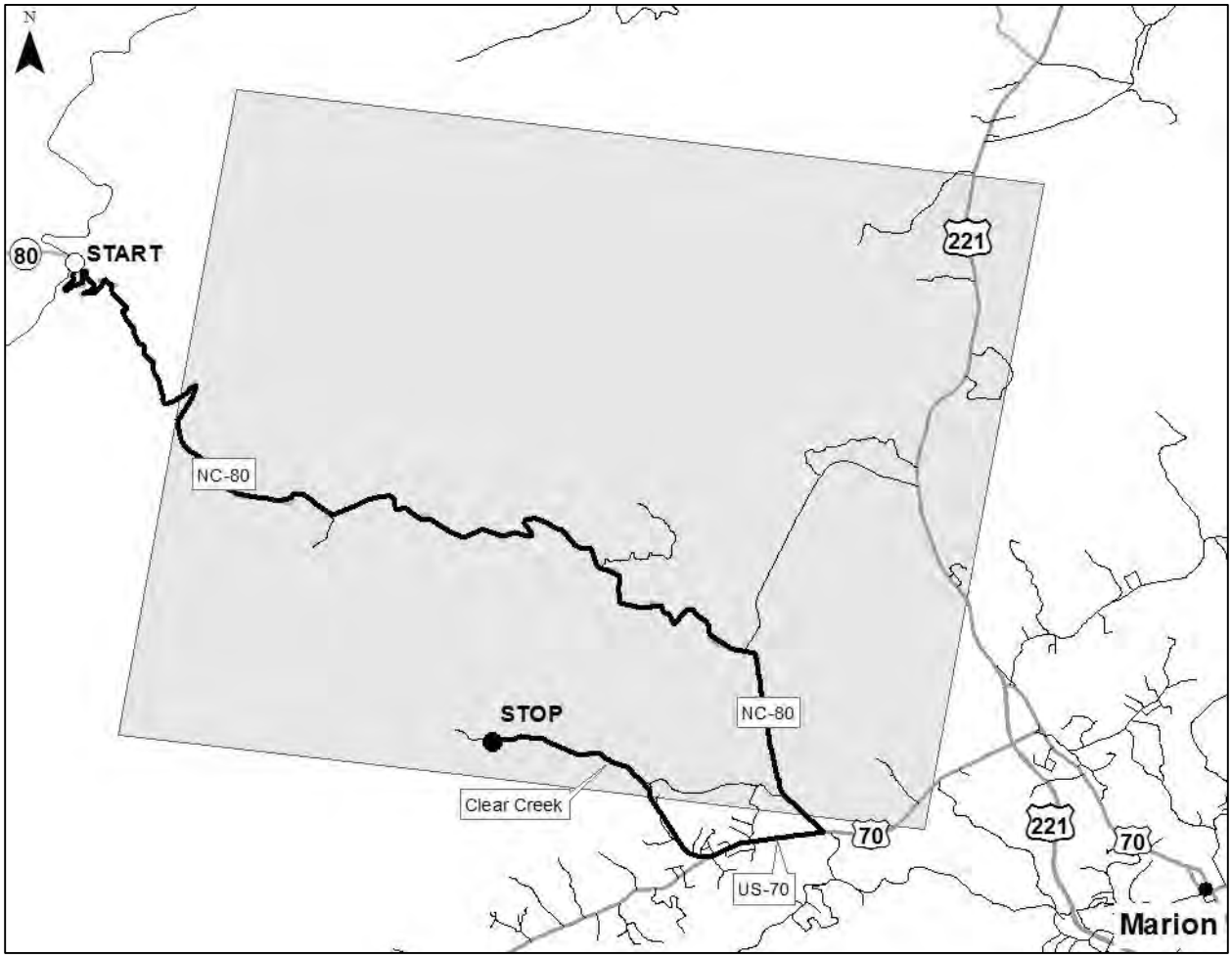
Grid 6 – Cherokee



Grid 39 – Marion – McDowell County

	Latitude	Longitude	
Start	35.770407	-82.164205	Blue Ridge Parkway and NC-80
Stop	35.704865	-82.110315	Stop after driving 2.3 miles on Clear Creek Rd. (dead end road)

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
U-turn	T Stop sign	NC-80 South	NC-80	12.2
Right	T Traffic light	US-70 West	US-70	0.9
Right	T No stop sign	Clear Creek Rd.	SR-1422	2.3

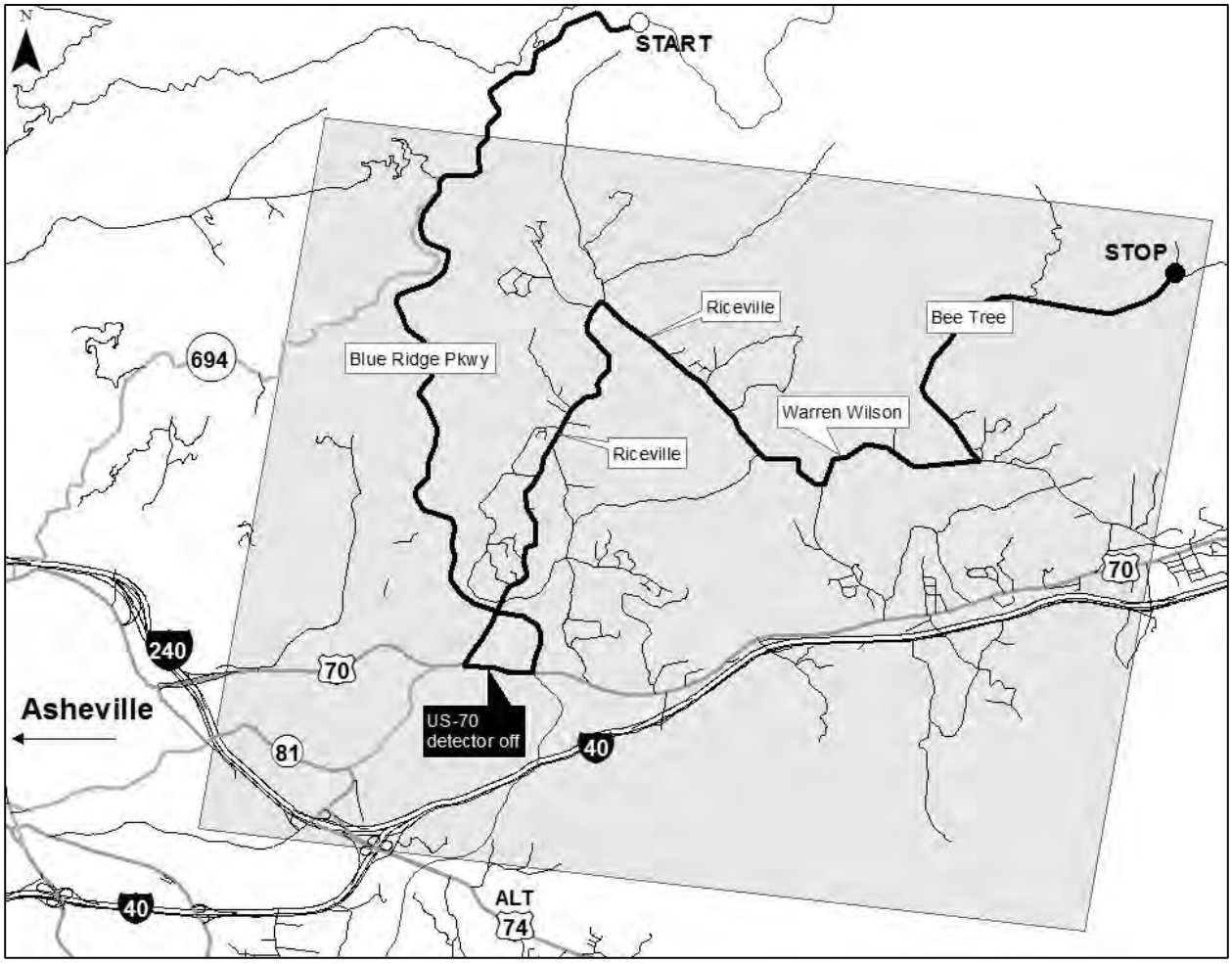


Grid 42 – Asheville – Buncombe County

	Latitude	Longitude	Intersection
Start	35.666913	-82.467172	2.4 miles North on Blue Ridge Parkway from the intersection of Blue Ridge Parkway and NC-694
Off	35.588796	-82.479117	
On	35.594319	-82.484118	
Stop	35.614613	-82.477310	Bee Tree Rd. and Summer Haven Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
U-turn	--	Blue Ridge Parkway South	FED-901	5
Right	Exit Stop sign	Tunnel Rd. **** turn off detector and drive normal speed***	US-70	0.5
Right	+ Traffic light	Riceville Rd. **** turn detector on <u>after</u> crossing under the Blue Ridge Parkway***	SR-2002	5.3
Left	+ Stop sign	Warren Wilson Rd.	SR-2416	1.3
Left	 No stop sign	Bee Tree Rd.	SR-2427	3

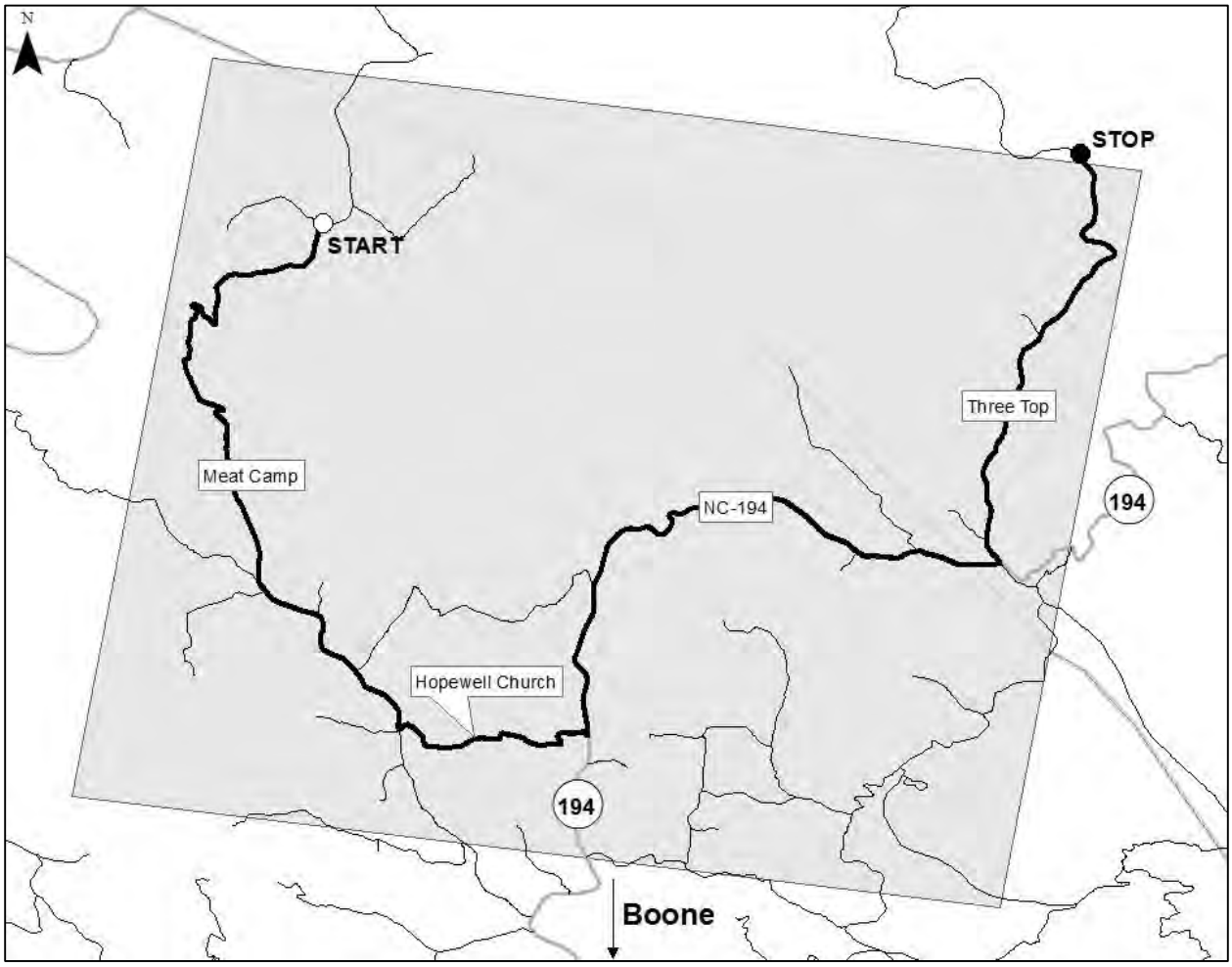
Grid 42 – Asheville



Grid 44 – Boone – Ashe/Watauga County

	Latitude	Longitude	
Start	36.352512	-81.680755	S Rd. and Meat Camp Rd.
Stop	36.360532	-81.590235	Three Top Rd. and William T. Calloway Rd.

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Right	T Stop sign	Meat Camp Rd.	SR-1340	5.8
Left	T No stop sign	Hopewell Church Rd.	SR-1339	1.6
Left	T Stop sign	NC-194 North	NC-194	4.8
Left	T No stop sign	Three Top Rd.	SR-1100	3.9

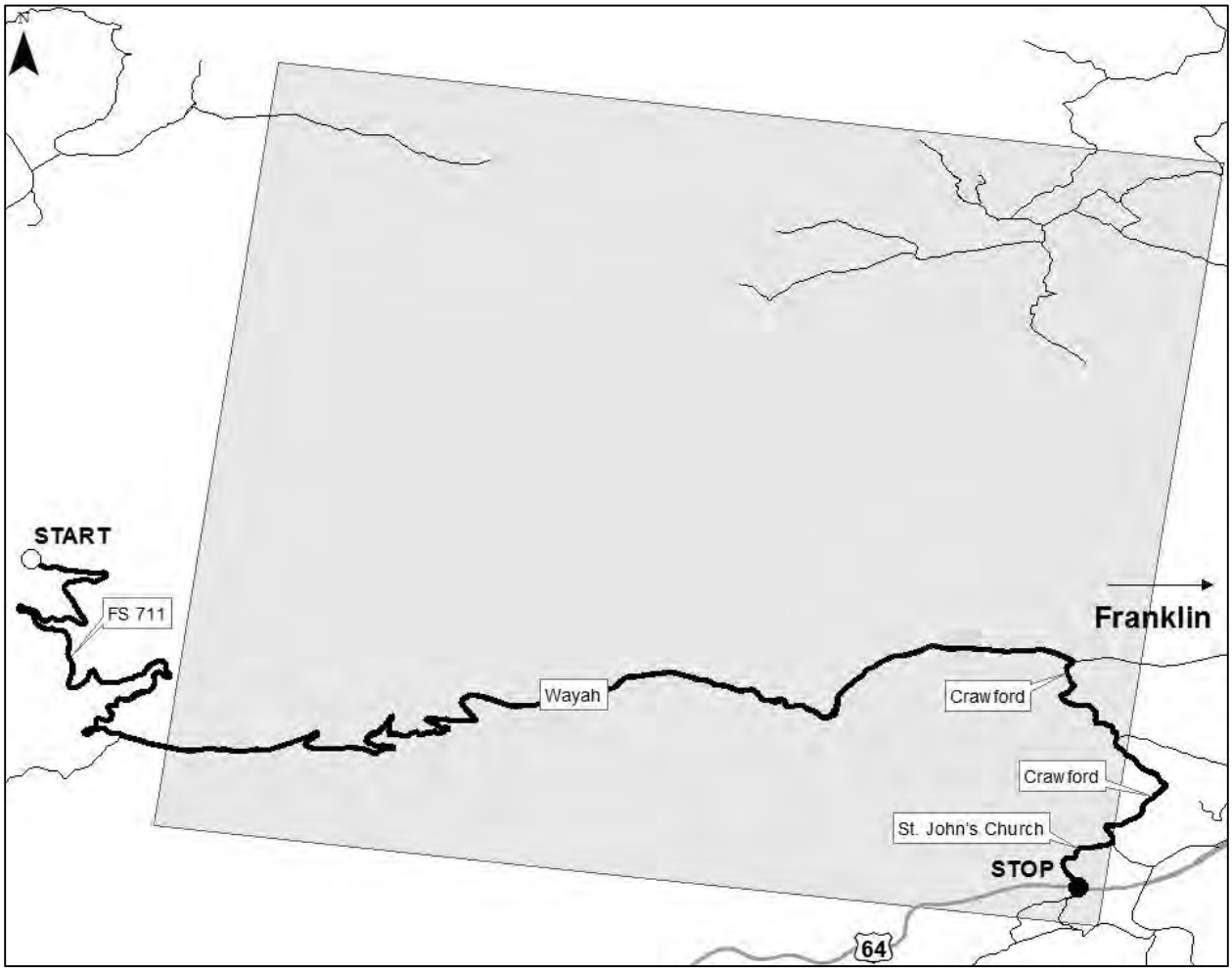


Grid 84 – Franklin - Macon County

	Latitude	Longitude	Intersection
Start	35.175662	-83.615702	Wayah Rd. and National Forest Service Rd. 711 (T-intersection), go 4.7 miles on National Forest Service Rd. 711 and make U-turn
Stop	35.137807	-83.494063	St. John's Church Rd. and US-64

Turn	Intersection Type	Street Name	SR-#	Distance (miles)
Left	--	National Forest Service Rd. 711	FED-711	4.7
Left	T Stop sign	Wayah Rd.	SR-1310	8.5
Right	T No stop sign	Crawford Rd.	SR-1309	1
Right	T Stop sign	Crawford Rd. ** gravel road*	SR-1308	1.3
Right	Y Stop sign	St. John's Church Rd.	SR-1308	0.6

Grid 84 – Franklin



NABat North Carolina

Mobile transect survey sheet

Please make sure your handwriting is legible

Surveyor(s): _____

Date (mm/dd/yyyy): _____

Sunset time: _____ Moonrise time: _____

Transect ID (grid cell ID): _____ Detector ID: _____

Transect name: _____

Start latitude (decimal degrees): N_____ longitude: W_____

End latitude (decimal degrees): N_____ longitude: W_____

	Time	Temp (F)	Wind speed	Humidity	Moon visible?	% cloud cover
Survey start						
Survey end						

	New	¼	Half	¾	Full
Moon phase					

Comments/notes (e.g. loud insects, traffics, wet roads/puddles, car stops, habitats):

NABat North Carolina

Stationary survey sheet (4 nights)

Please make sure your handwriting is legible

Grid cell ID: _____ Site name: _____

Latitude: N _____ Longitude: W _____

Elevation: _____ Distance to clutter: _____

Surveyor(s): _____

Microphone orientation: _____ Microphone height: _____

Photographic equipment: _____

Photos: _____

Date deployed:

Night 1	Sunset			Moonrise			Moonset			Sunrise		
Notes:												
	Temperature			Wind			RH					
	High	Low	Mean	High	Low	Mean	High	Low	Mean			

Night 2	Sunset			Moonrise			Moonset			Sunrise		
Notes:												
	Temperature			Wind			RH					
	High	Low	Mean	High	Low	Mean	High	Low	Mean			

Night 3	Sunset			Moonrise			Moonset			Sunrise		
Notes:												
	Temperature			Wind			RH					
	High	Low	Mean	High	Low	Mean	High	Low	Mean			

Night 4	Sunset			Moonrise			Moonset			Sunrise		
Notes:												
	Temperature			Wind			RH					
	High	Low	Mean	High	Low	Mean	High	Low	Mean			

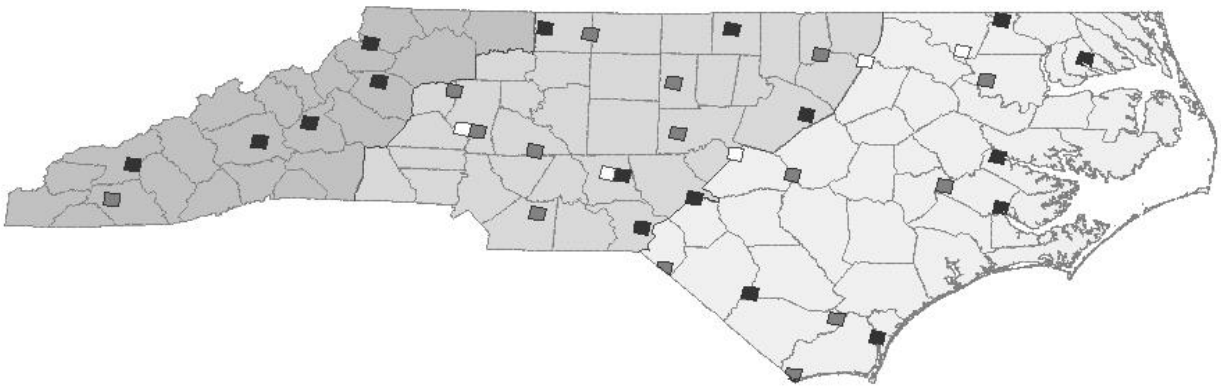
Date ended:

	New	¼	Half	¾	Full
Moon					

Comments/notes (e.g., recording mid-stop, recording resumed, significant weather):

Driving Transect Survey Training Manual

North American Bat Monitoring Program
North Carolina Division



Dr. Han Li
Ashley Matesson
Dr. Matina Kalcounis-Rueppell

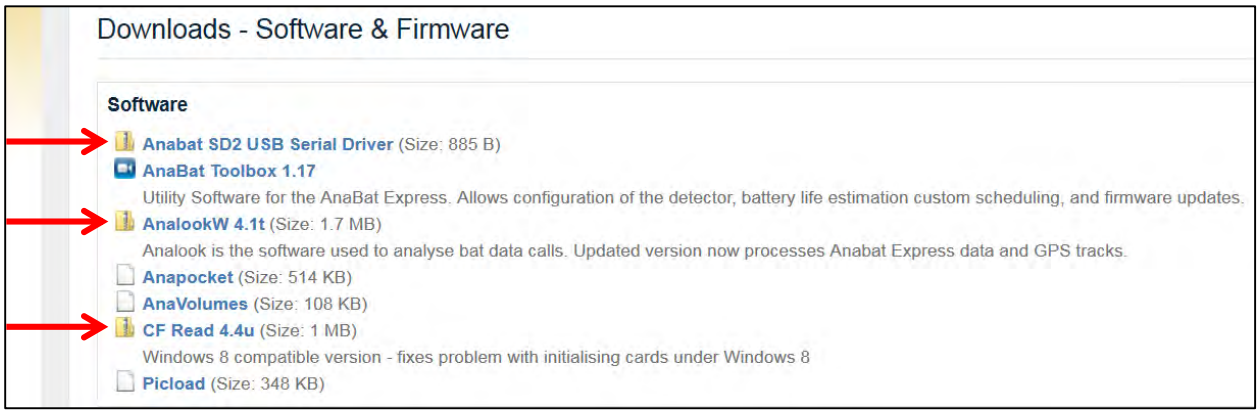
University of North Carolina at Greensboro
Department of Biology
321 McIver St.
312 Eberhart Building
Greensboro, NC 27412

September 2015

Table of Contents

Downloading Software

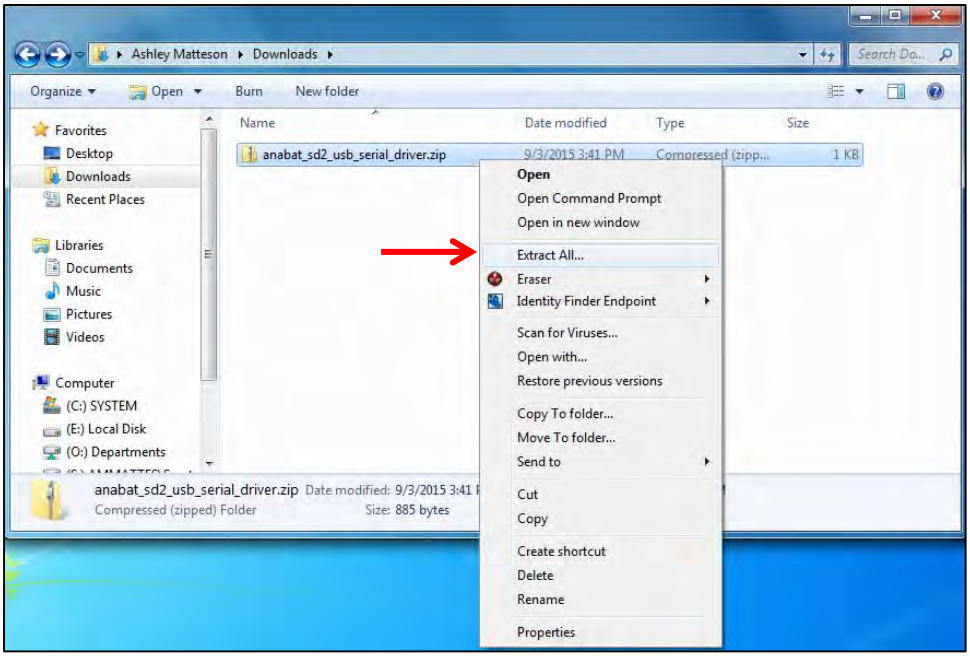
- AnaBat software can be downloaded from:
  - http://www.titley-scientific.com/us/index.php/software_firmware
- Three items are necessary:
  - AnaBat SD2 USB Serial Driver
  - AnaLookW 4.1t
  - CF Read 4.4u



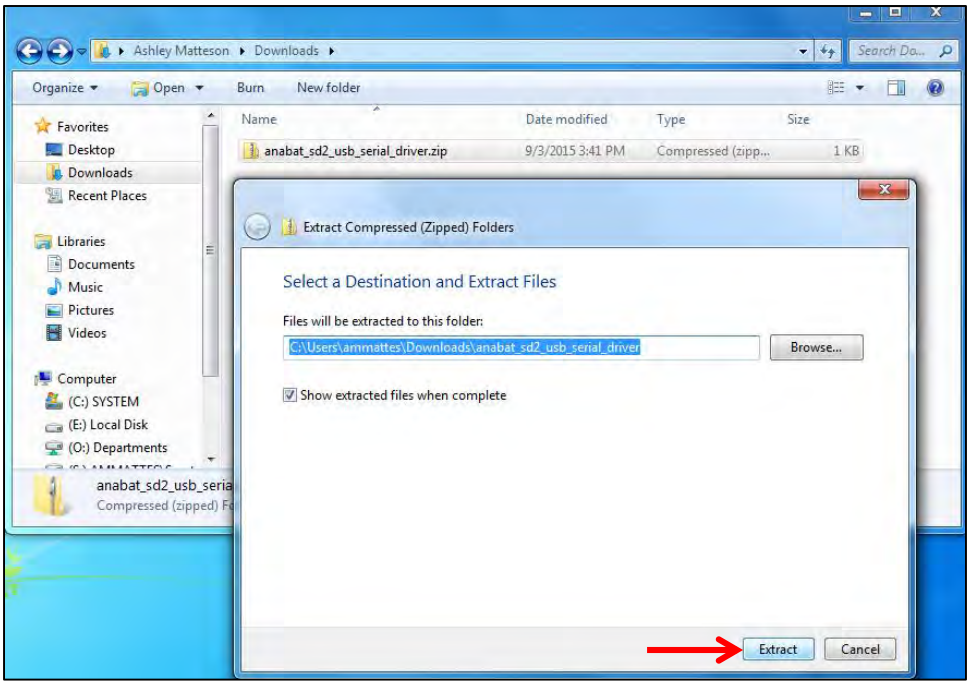
- Click on the link and a new window will open
  - Choose Save File
  - Click OK
  - Repeat for all three necessary items

Installing Serial Driver

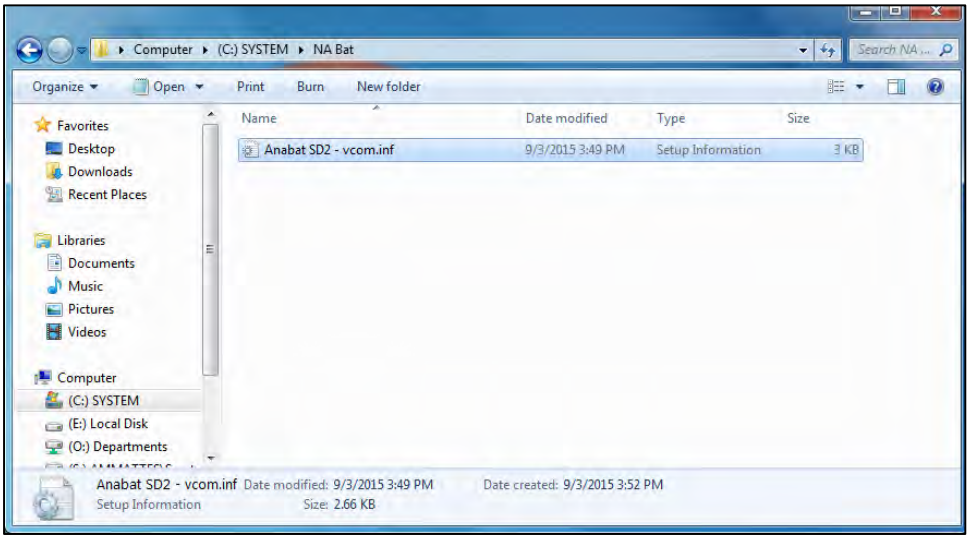
- AnaBat SD2 USB Serial Driver is needed to connect the AnaBat SD2 to a computer (PC only) using a USB cable
- Unzip the file by right clicking and selecting ‘Extract All…’



- New window will open click ‘Extract’

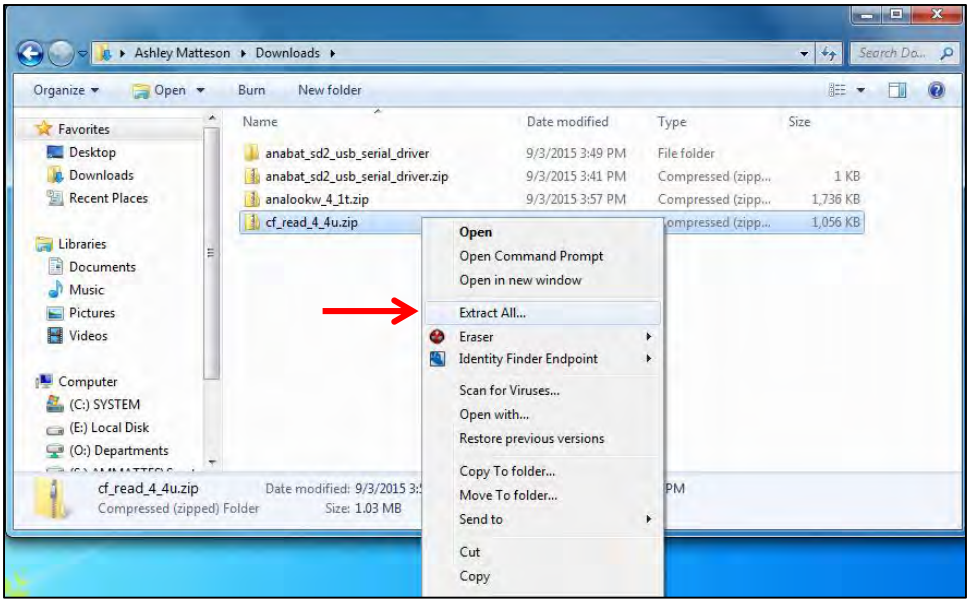


- Open the ‘anabat_sd2_usb_serial_driver’ folder
- Copy and paste the driver file ‘AnaBat SD2-vcom.inf’ to your computer

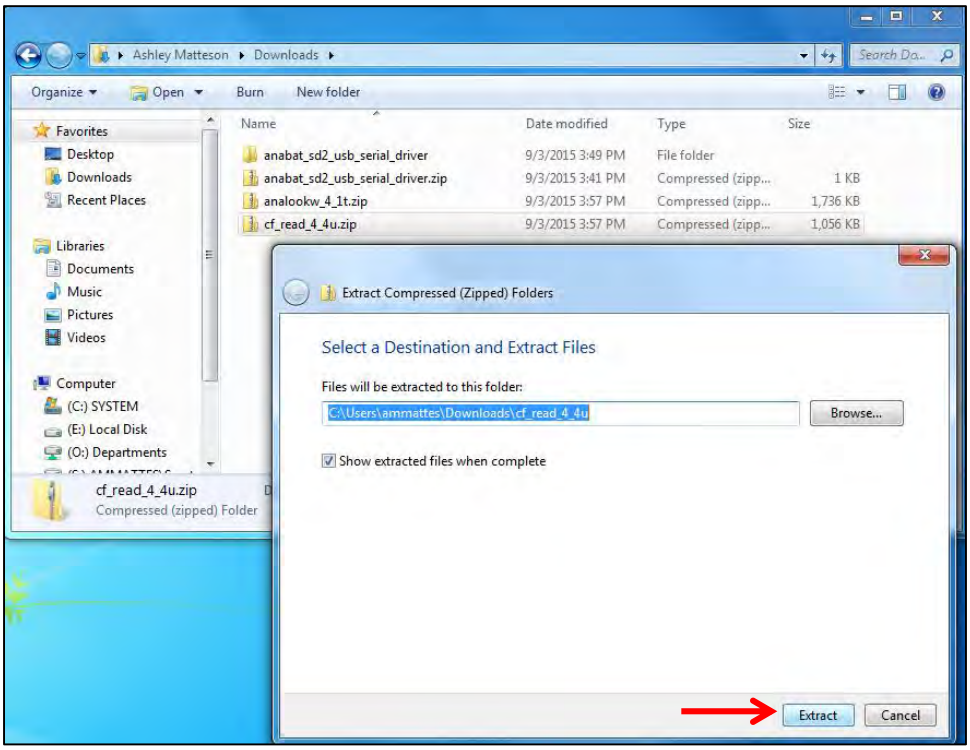


Installing CF Read 4.4

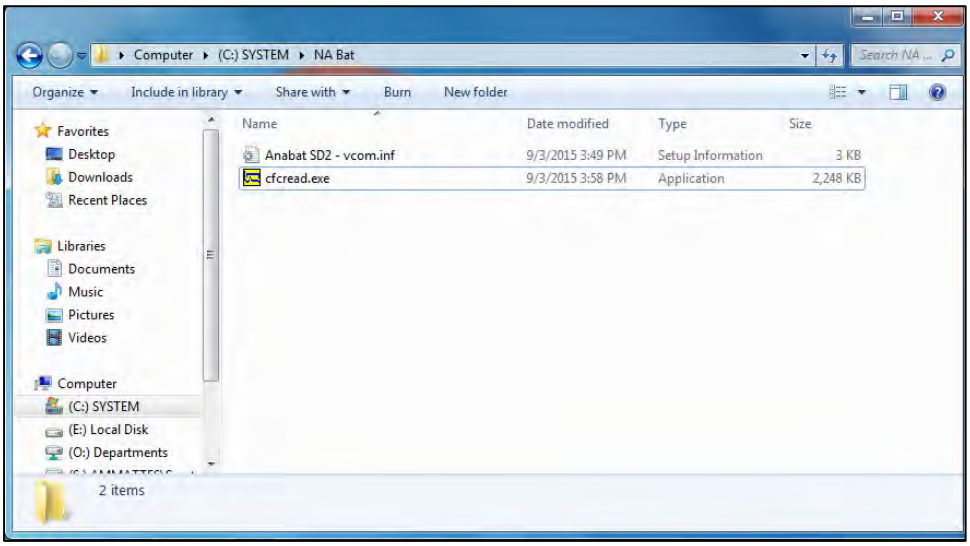
- CF Read 4.4 is used to Initialize CF Cards, Download Data, and Synchronize the internal detector clock
- Unzip the file by right clicking and selecting ‘Extract All…’



- New window will open click ‘Extract’

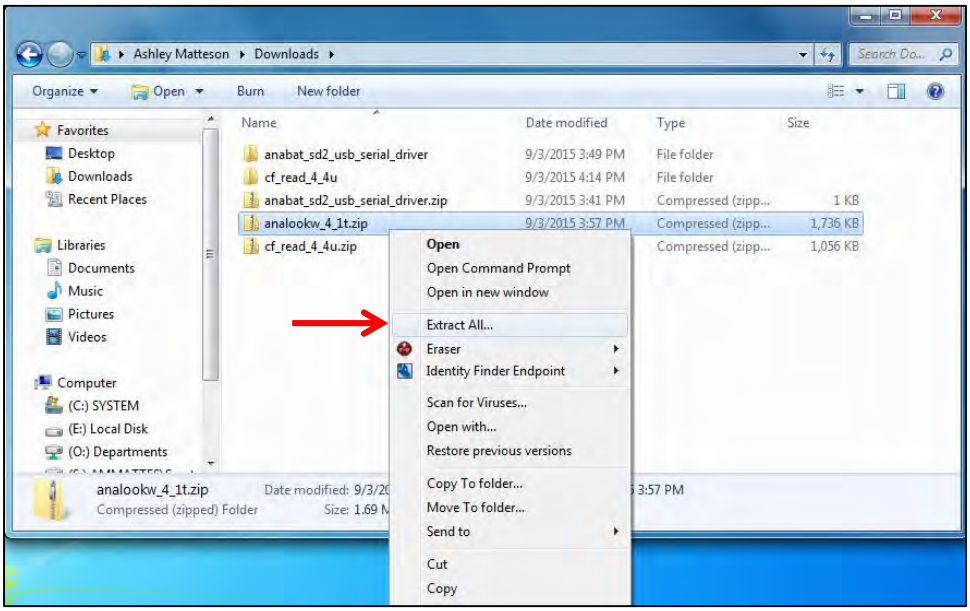


- Open the ‘cf_read_4_4u’ folder
- Copy and paste ‘cfcread.exe’ to your computer

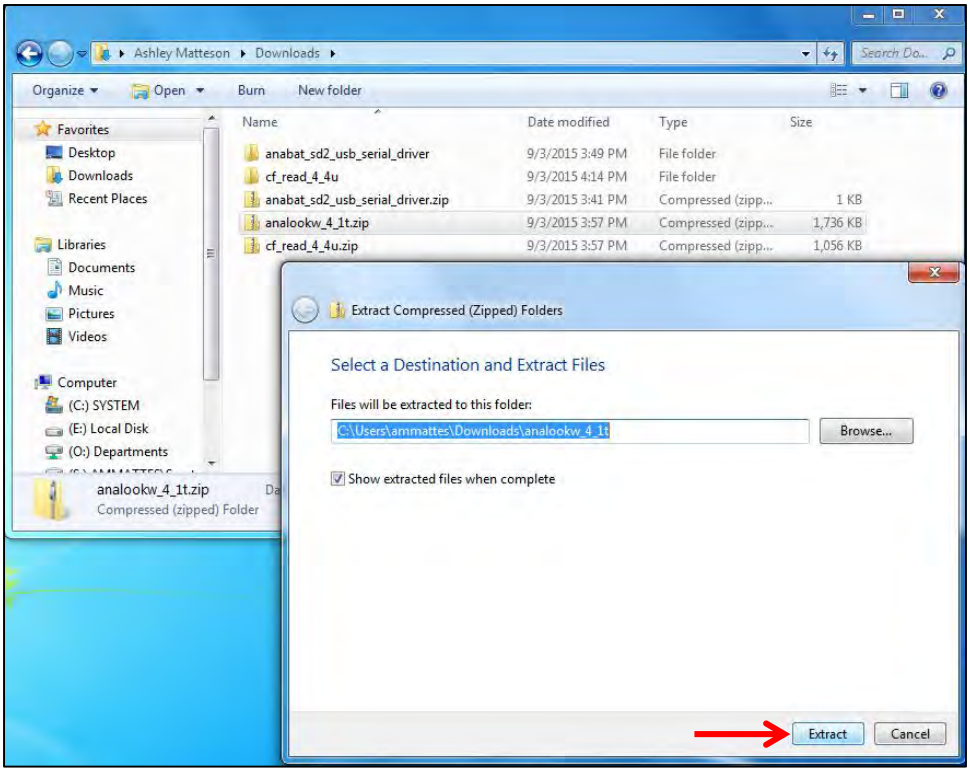


Installing AnaLookW 4.1t

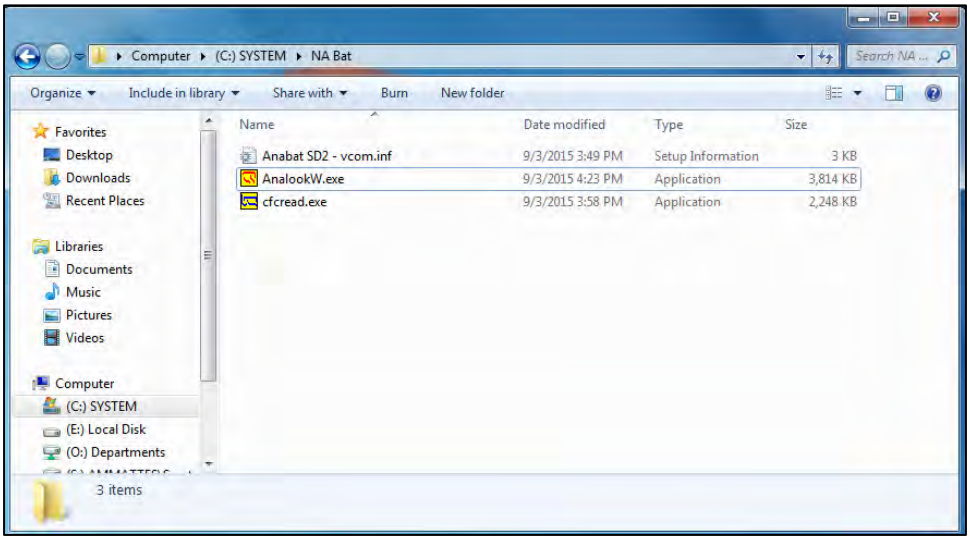
- AnaLookW 4.1t is used to view recordings
- Unzip the file by right clicking and selecting ‘Extract All…’



- New window will open click ‘Extract’

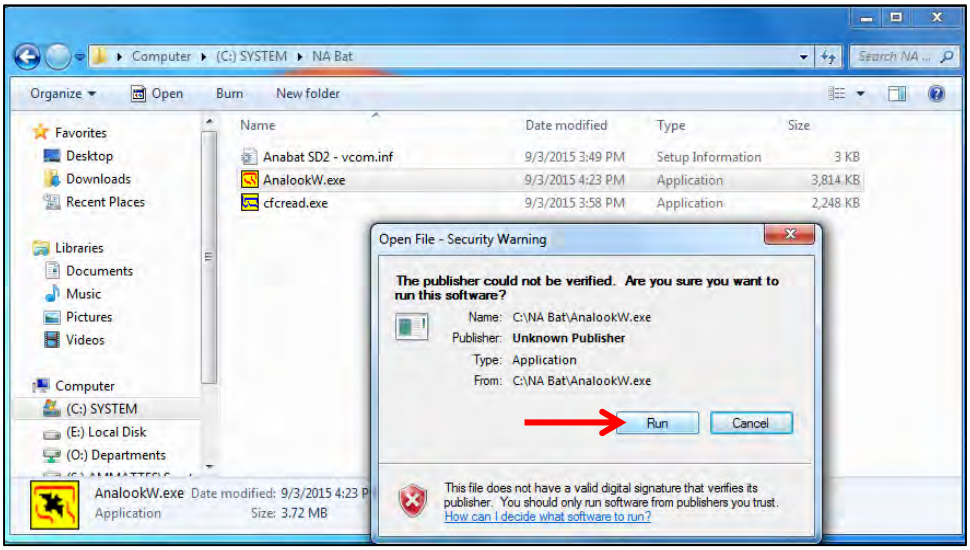


- Open the ‘analookw_4_1t’ folder
- Copy and paste ‘analookw.exe’ to your computer

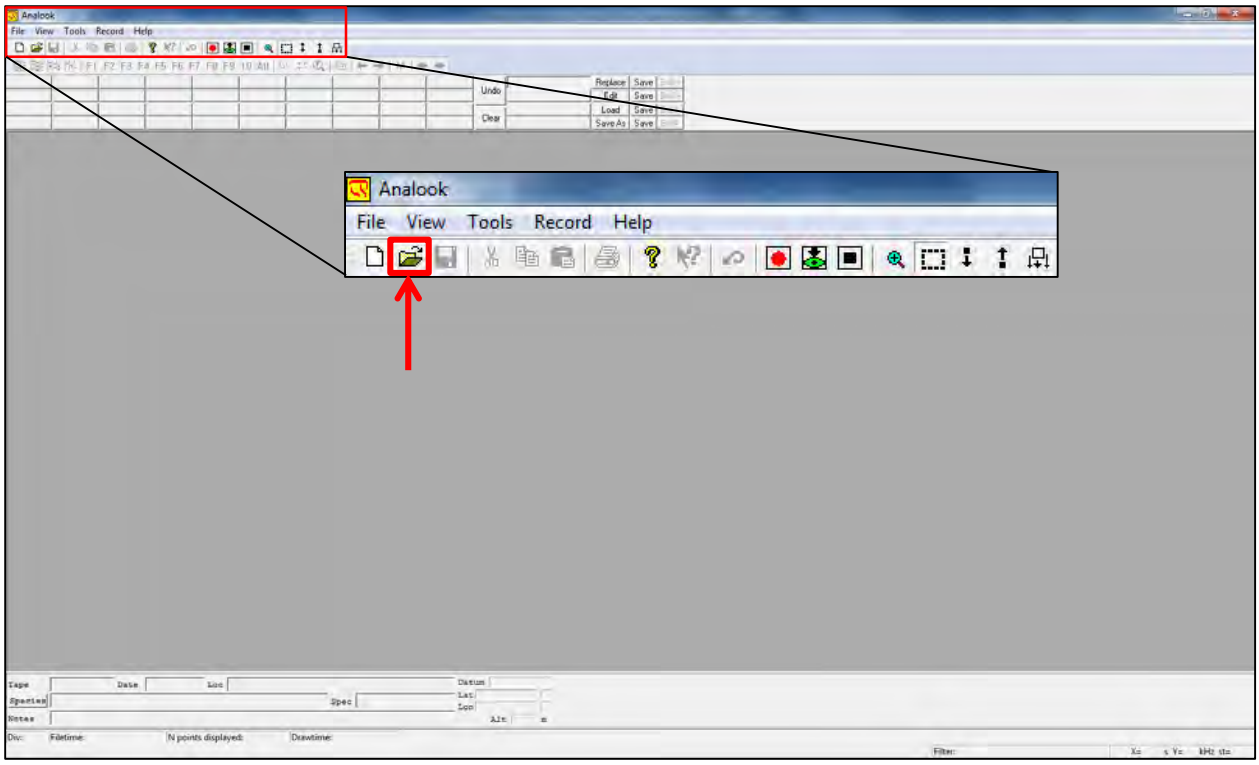


Using AnaLook W

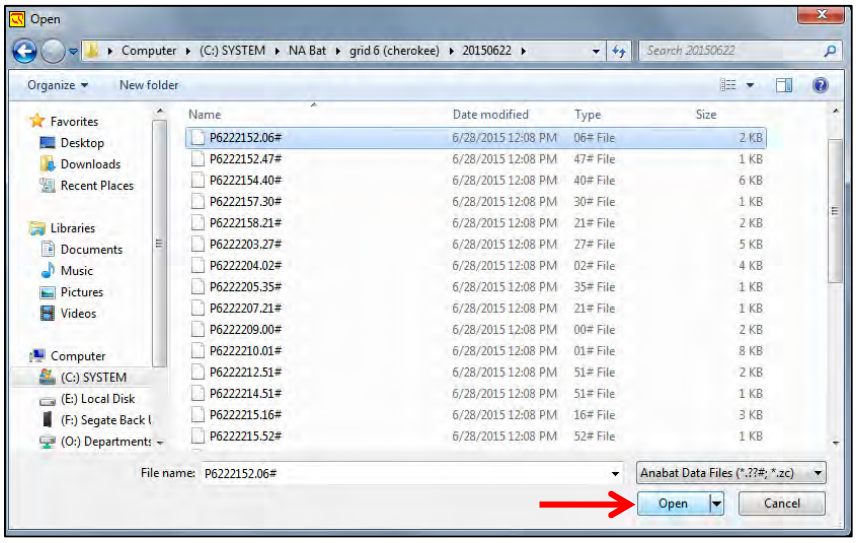
- If you choose to look at your recordings you can do this by clicking on ‘AnalookW.exe’
- New window will open click ‘Run’



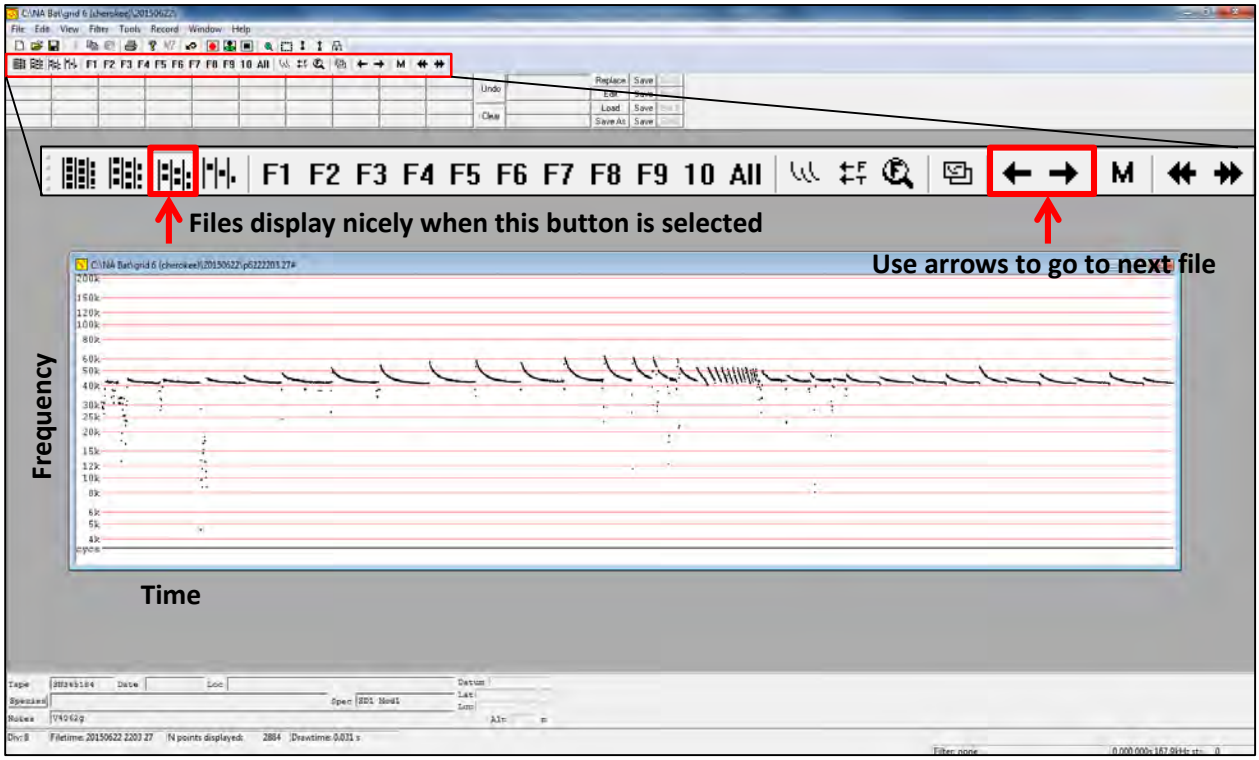
- Analook W will open
- Click on ‘Open’ icon



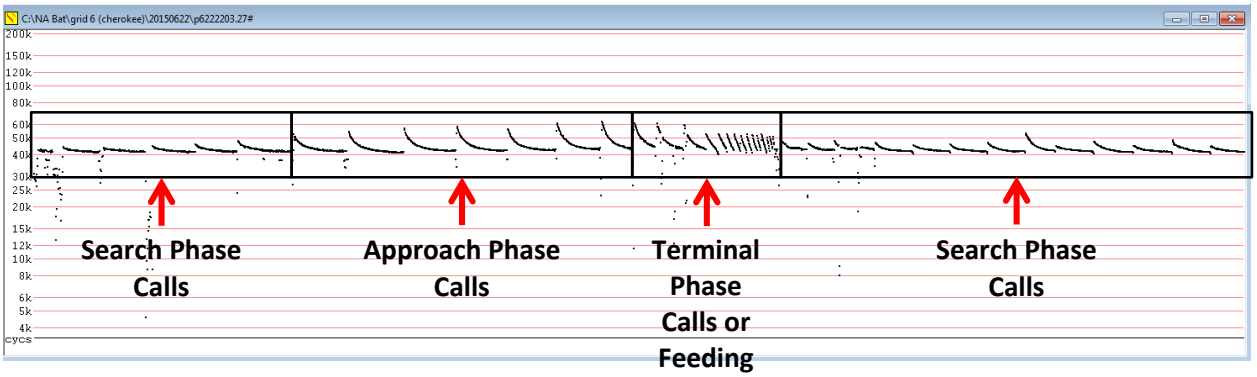
- New window will open
- Navigate to where you have backed up the data (your computer’s hard drive)
- Double click on the folder you are interested in
- Click on a file
- Click ‘Open’ (last step shown below)



- File will open and screen will look like this



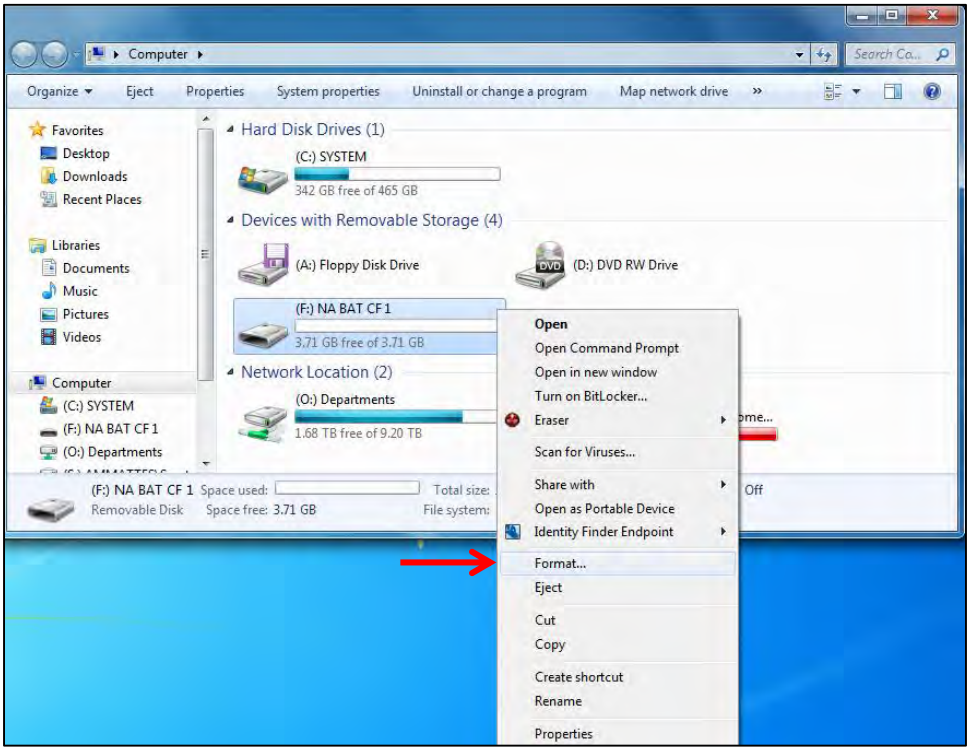
- What are you looking at???



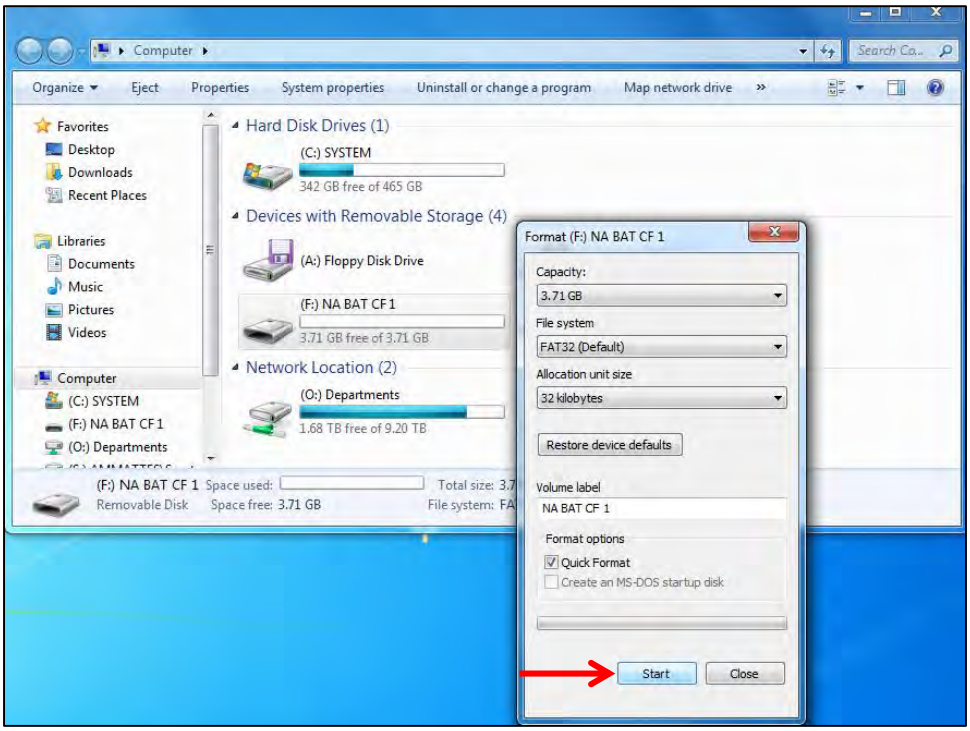
- Most of the calls you will record are search phase calls
- Bats use the search phase calls as they are flying looking for insects once an insect is detected the bat switches to approach phase calls, as the bat begins closing in on the insect it will switch to the terminal phase calls
- The different call types allow the bat to collect more precise information about where the insect is located

Formatting Compact Flash Cards

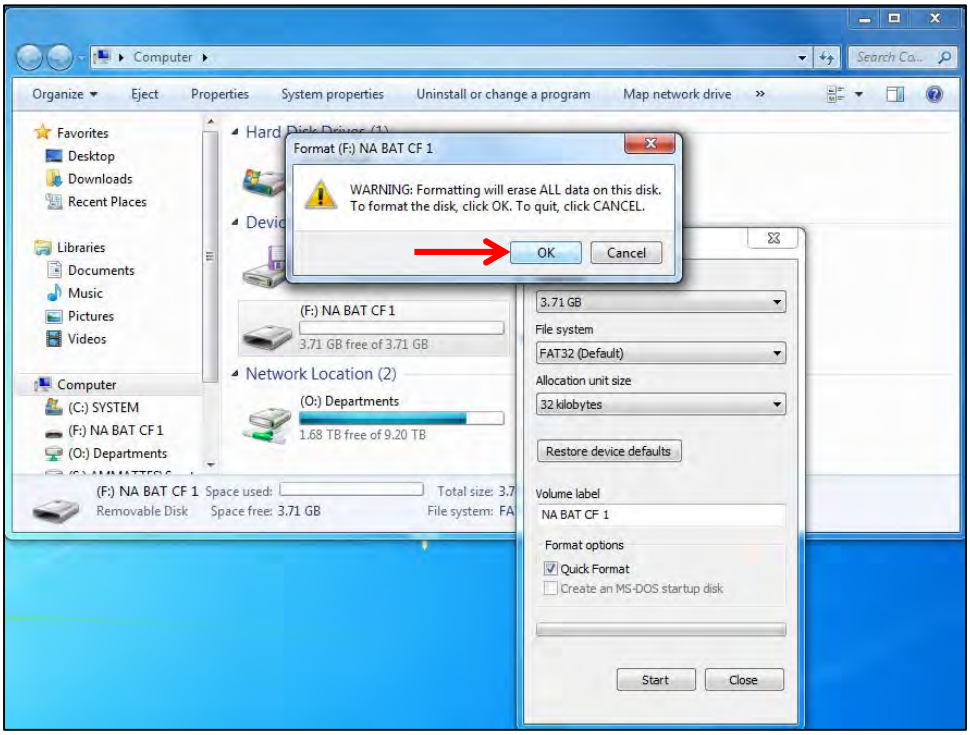
- Insert CF card into card reader
- Open ‘My Computer’
- Right click on the CF card
- Select ‘Format’



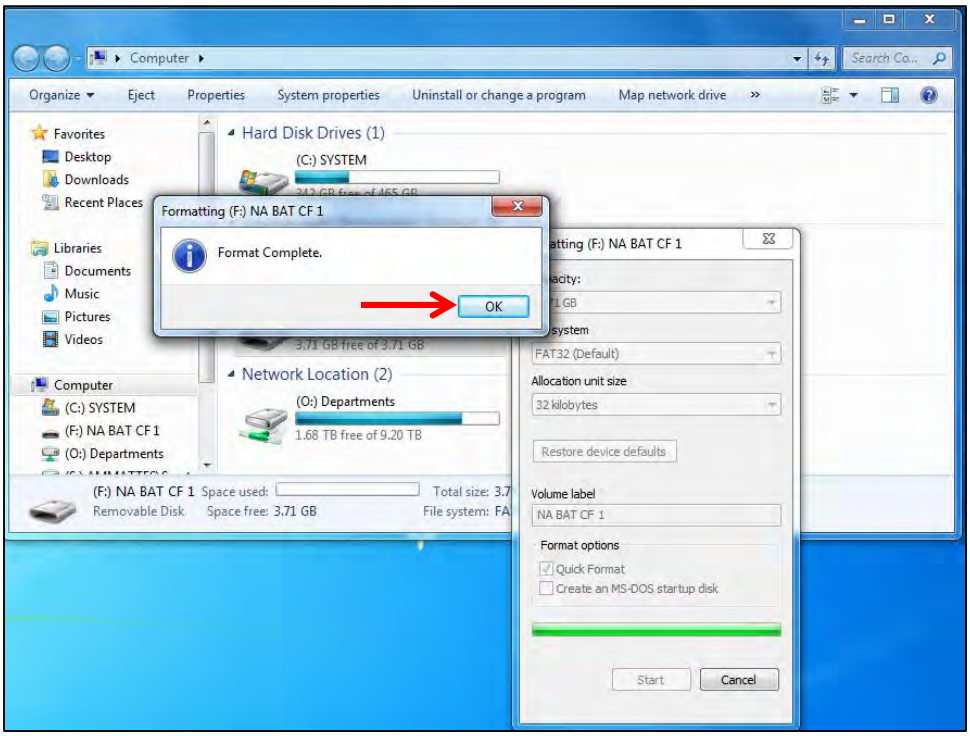
- New window opens
- Click ‘Start’



- New window opens
- Click ‘OK’

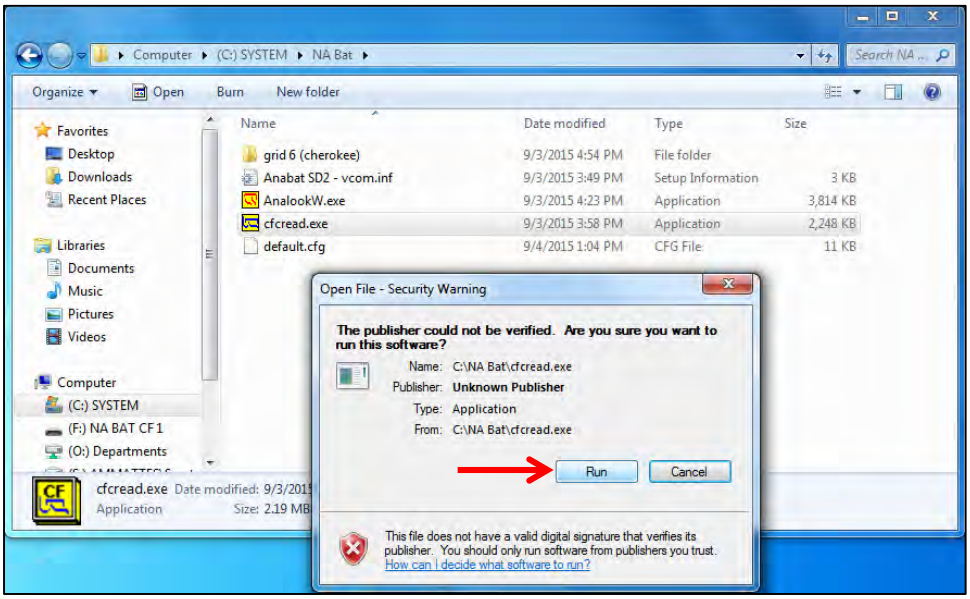


- Format Complete will appear in new window
- Click ‘OK’

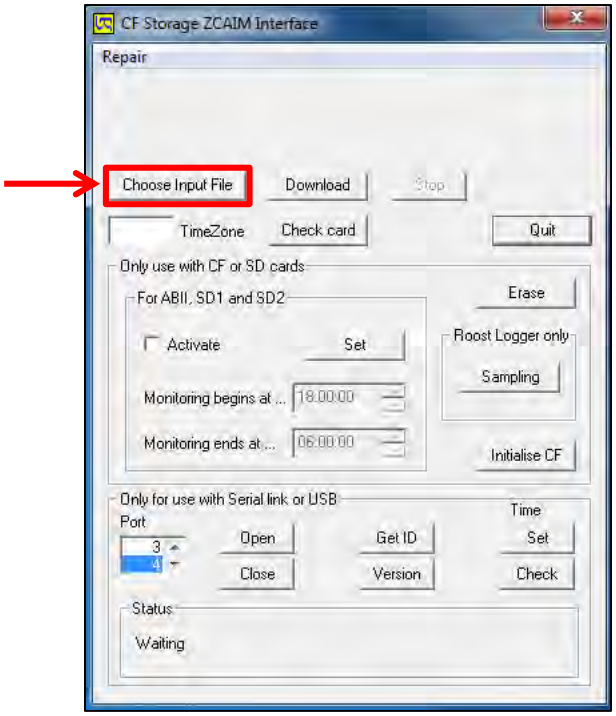


Initializing Compact Flash Cards

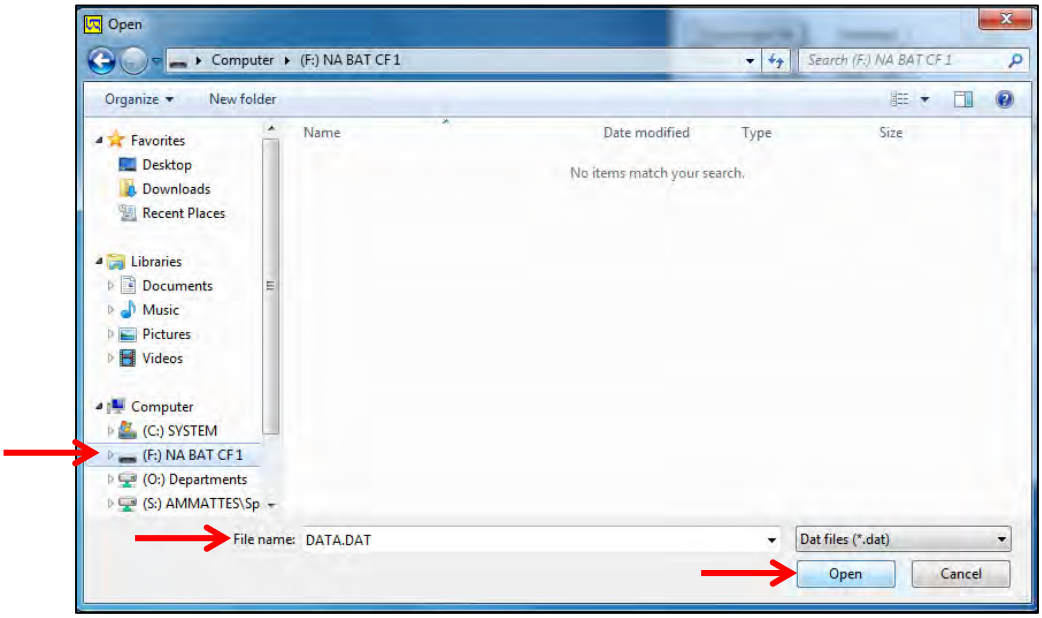
- Open cf_read.exe
- Click ‘Run’



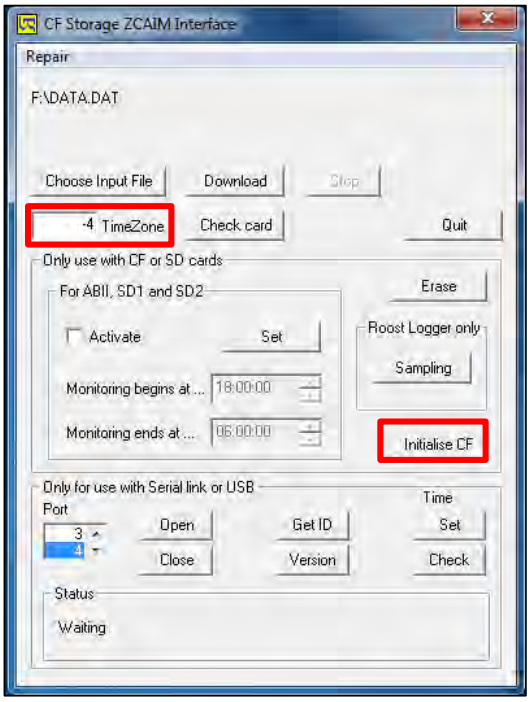
- CF Read opens
- Click ‘Choose Input File’

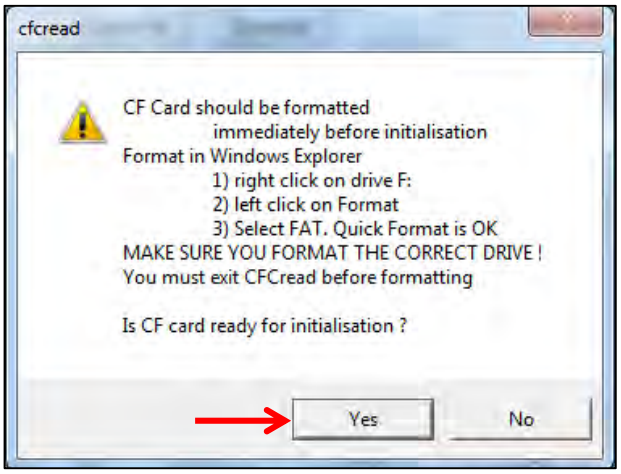


- New window opens
- Select the CF card
- DATA.DAT file is automatically created in the file name window
- Click ‘Open’

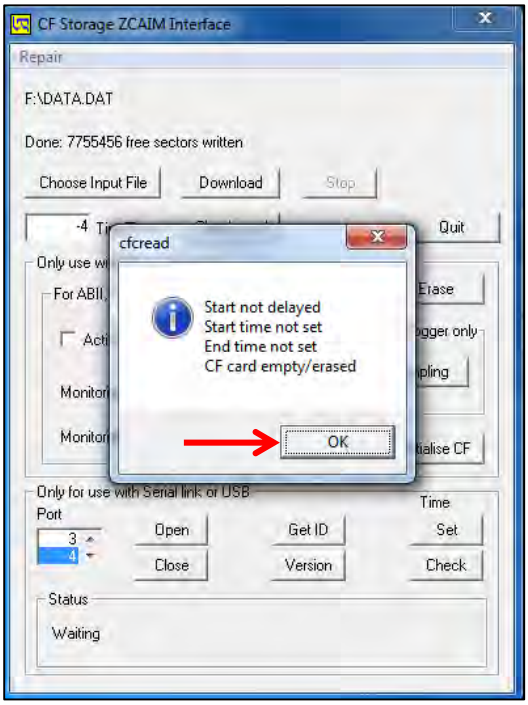


- Set Time Zone to -4 (for North Carolina)
- Click ‘Initialise CF’
- New window opens
- Click ‘Yes’





- Initialization process takes 5-10 minutes
- New window opens when initialization is complete
- Click ‘OK’



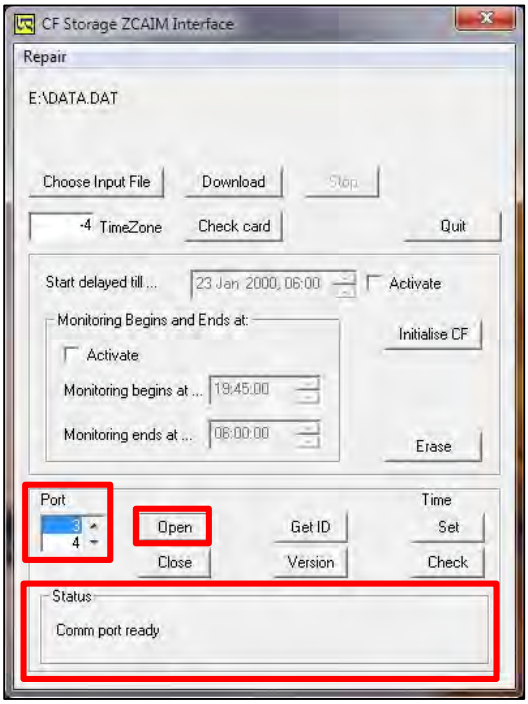
Synchronizing the AnaBat SD2 Internal Clock

- Before you can do this:
  1) Serial driver has to be installed
  2) CF card has to be removed from the AnaBat SD2
  3) Clock on your computer has to be correct
     - The internal clock in the AnaBat SD2 will sync to the computer's clock
- You will need:
  - USB cable
  - AnaBat SD2
  - Computer with proper software installed

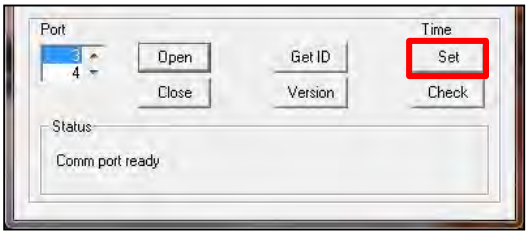
- Connect the USB cable to the AnaBat SD2

- Turn on the AnaBat SD2 BEFORE connecting it to the computer

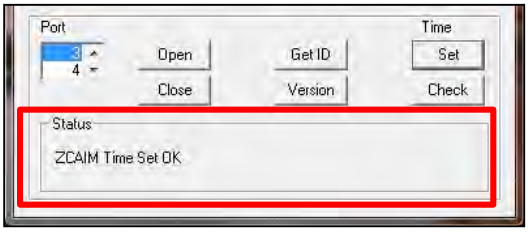
- Connect the AnaBat SD2 to the computer using the USB cable
- Open CF_read.exe
- Select ‘Port 3’
- Click ‘Open’
- It should display ‘Comm port ready’ in the status box
    - If error message displays try to repeat process with ‘Port 4’



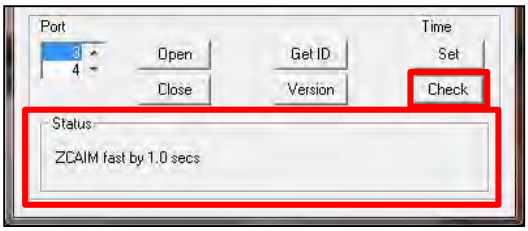
- Click ‘Set’



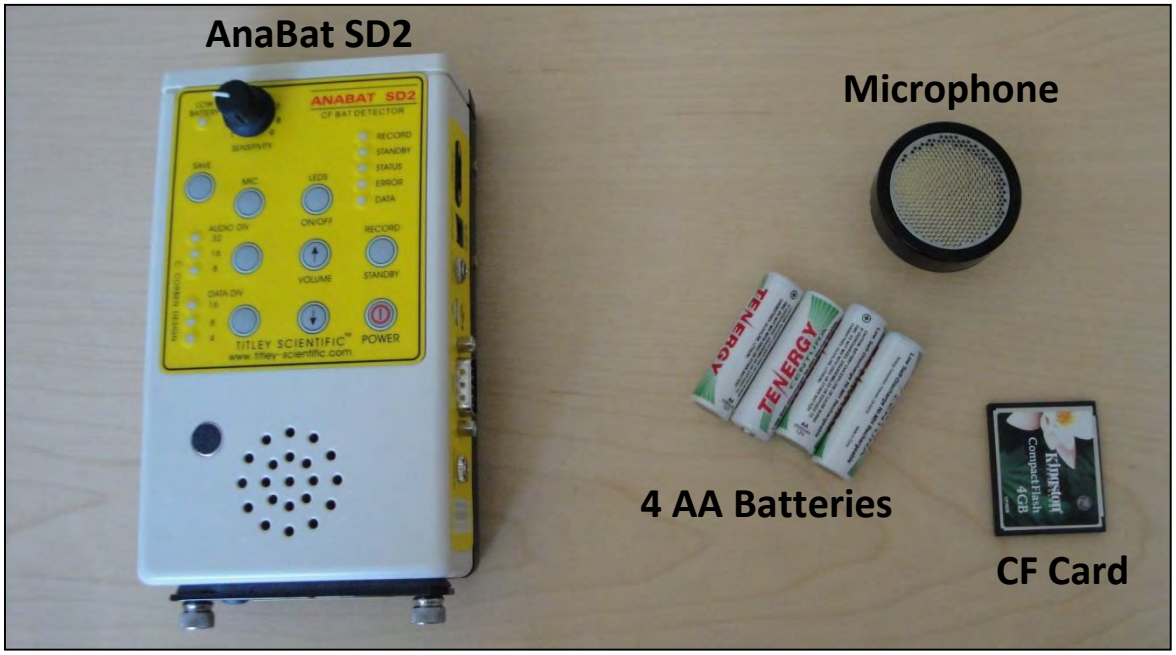
- In Status box ‘ZCAIM Time Set OK’ message will display



- Click ‘Check’
    - ZCAIM fast by 1.0 secs is good

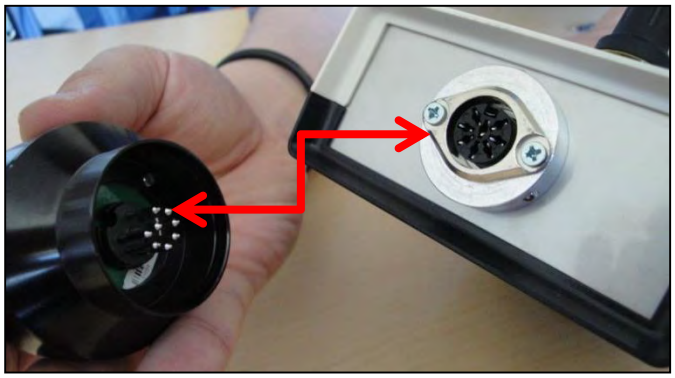


Equipment



Step 1: Connect Microphone

- Line up the pins on the microphone with the holes in the AnaBat SD2 and push microphone straight on until you hear a click



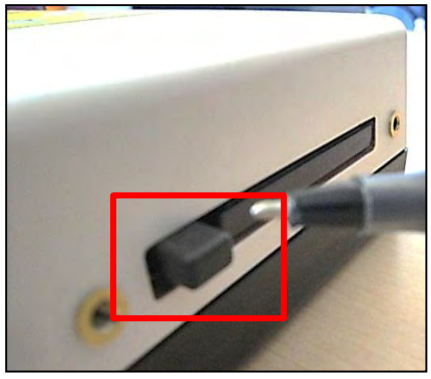
- To remove microphone pull straight off

Step 2: Install CF Card

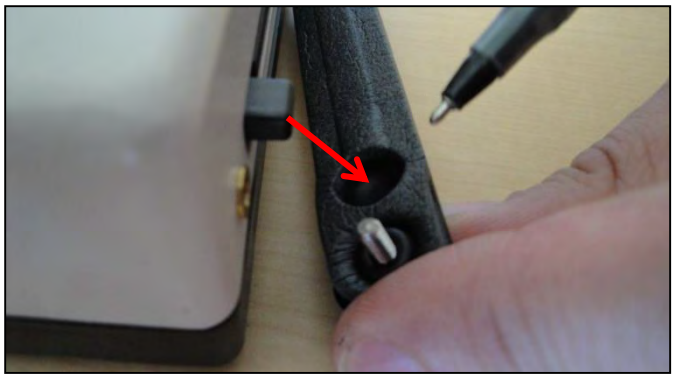
- Unscrew bottom panel

- Remove bottom panel

- Insert CF Card **<u>picture side up</u>**
    - Black button will pop out when card is inserted properly



- Replace bottom panel
    - Black button will line up with the hole in the bottom panel



- Remove CF Card by removing the bottom panel and pressing the black button

Step 3: Install Batteries

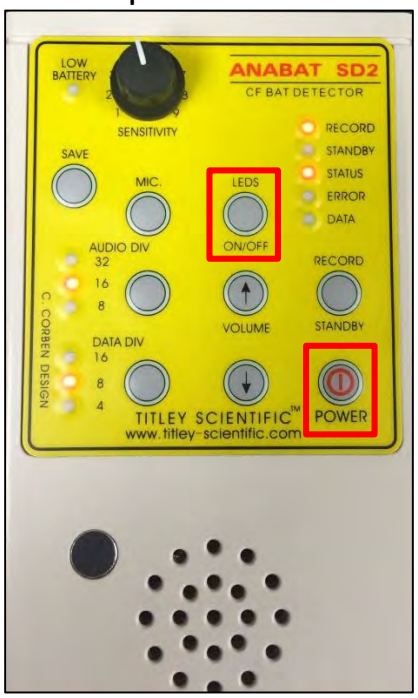
- Remove battery cover

- Install 4 AA batteries

- Replace battery cover

Step 4: Turn on AnaBat SD2

- Press POWER button
    - If no lights come on press the LEDS ON/OFF button

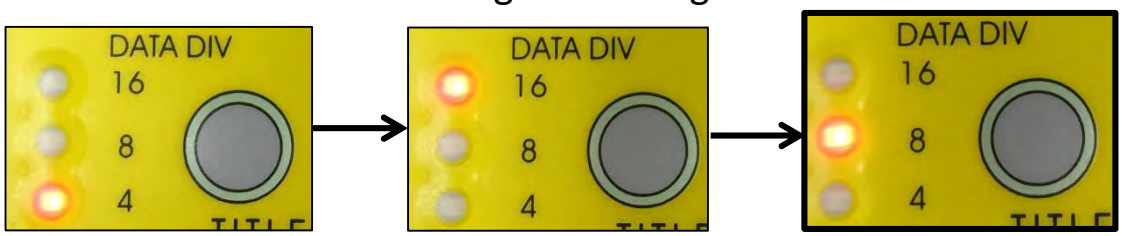


Step 5: Check the AnaBat SD2 Settings

- LOW BATTERY light should not be on

- AUDIO DIV light should indicate **<u>16</u>**
    - Press button to right to change

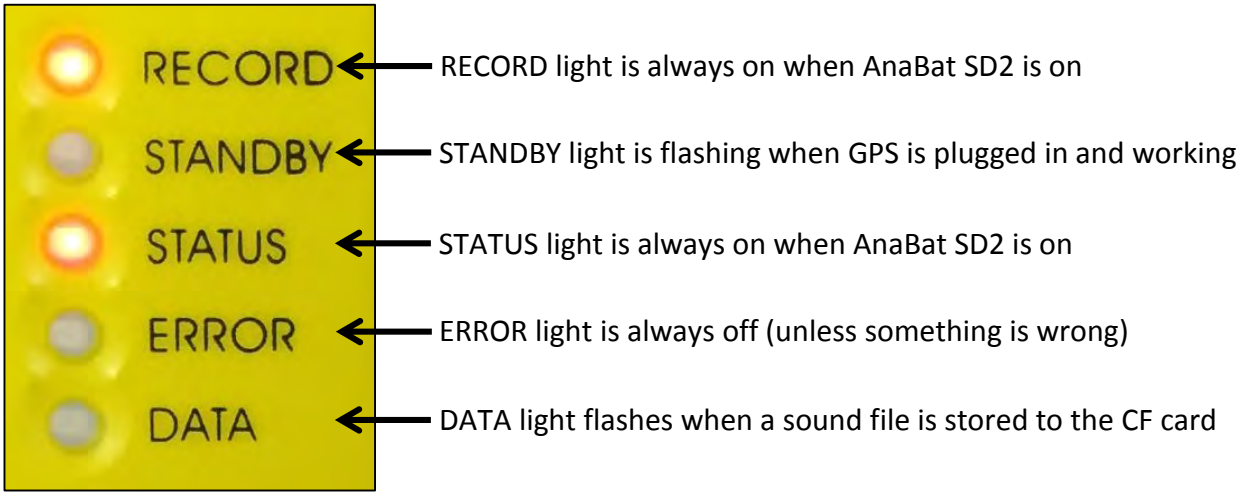
- DATA DIV light should indicate **<u>8</u>**
    - Press button to right to change



- SENSITIVITY Knob should be set between 4 and 5

- VOLUME up/down buttons
    - Make noise in front of the microphone you should hear noise from the AnaBat SD2
    - Adjust the volume up if you cannot hear noise

- RECORD and STATUS lights should be on when AnaBat SD2 is powered on
- STANDBY light will flash when GPS coordinates are recorded during driving transect
- DATA light will flash when sound is recorded to the CF card



- ERROR light will be illuminated if there is a problem
    - Common problems
        - Inserting CF card after AnaBat SD2 is on/ improperly
            - ✓ Turn off AnaBat SD2, remove CF card, insert CF card and turn on AnaBat SD2
        - CF card is not formatted and initialized correctly
            - ✓ Turn off AnaBat SD2, remove CF card, format CF card, initialize CF card, insert CF card and turn on AnaBat SD2

Equipment for Driving Transect

Inverter
****Will replace the GPS Battery Pack after 2015 season**

Step 1: Connect Cable

- Plug the silver end of the cable into the AnaBat SD2 by pushing straight on
    - Make sure to line up the pins on the silver end to the holes in the AnaBat SD2

- Plug the black end of the cable into the car mount by pushing straight on and screwing down the black piece
    - Loosen knob on the car mount so the head of the car mount moves freely

Step 2: Attach Microphone

- Remove microphone collar by turning left

- Plug microphone in by lining up pins and holes and pushing straight on

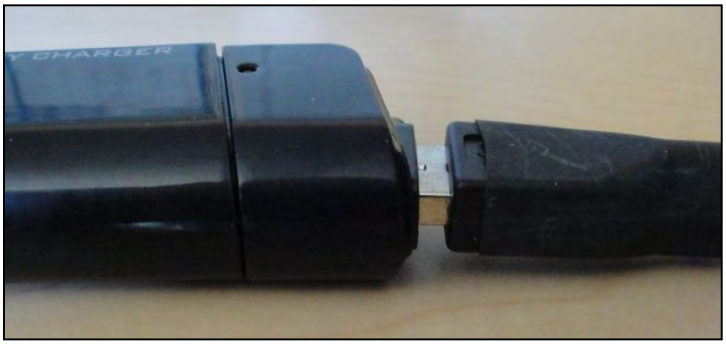
- Replace microphone collar by turning right

Step 3: Setting Up GPS

- GPS Battery Pack requires 2AA batteries
- Insert batteries

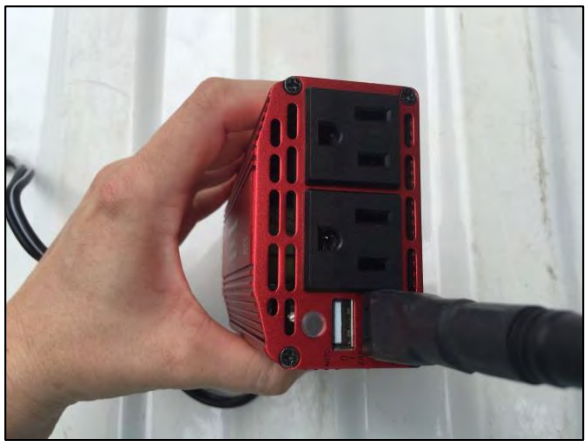
- Connect the USB end of the GPS cable to the USB port on the GPS battery pack

- This is what it looks like when it is plugged in all the way



- Alternatively to the GPS Battery Pack an Inverter with USB ports can be used

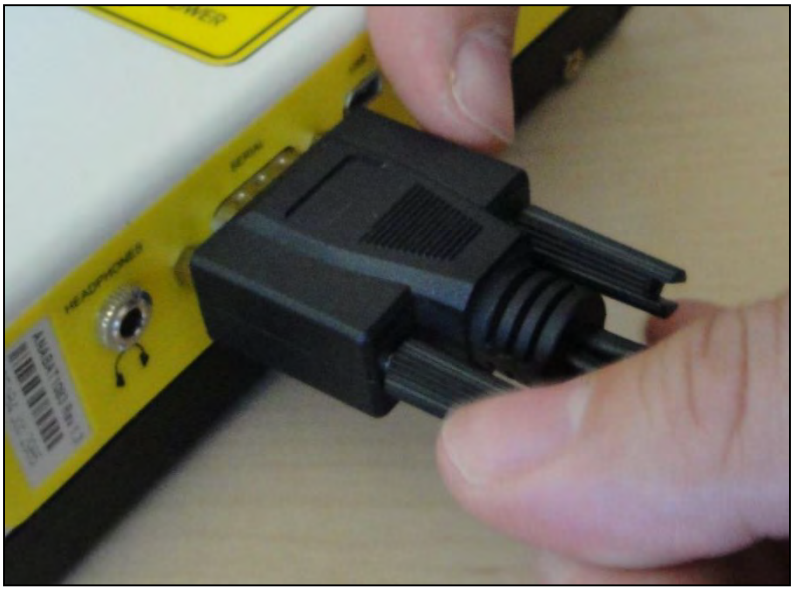
- Plug the USB end of the GPS cable to the USB port on the inverter



- Plug the inverter into the charging port in your vehicle

- Connect the serial end of the GPS cable to the serial port on the AnaBat SD2

- Secure connecting by tightening screws



- Check to make sure the GPS cable connection is secure

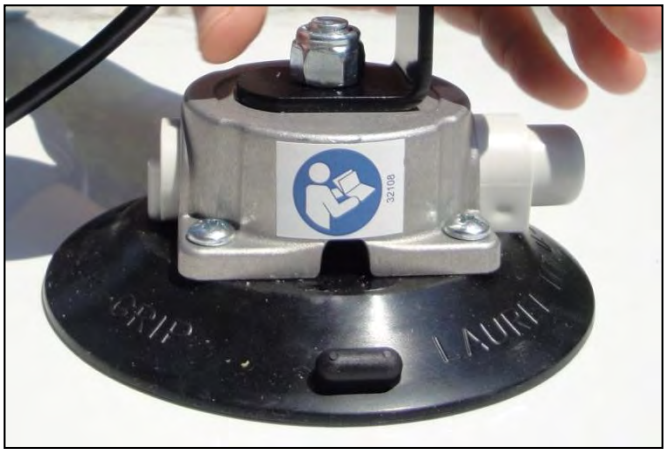
Step 4: Setting Up the Car Mount

- Clean vehicle top

- Remove suction cup cover by pulling off

- Place suction cup on vehicle top

- Secure suction cup to vehicle top by repeatedly pressing button until red line cannot be seen



- Test security by trying to move it

- Place GPS on vehicle top, there is a magnet on the bottom so it should secure itself to the vehicle

- Check orientation of microphone, the microphone should point straight up

- Loosen knob to adjust

- Roll down passenger side window and put the AnaBat SD2 and GPS battery pack or inverter inside vehicle

- If you want to you can roll up window part way

Step 5: Before Driving the Transect

- Turn on AnaBat SD2 by pressing POWER button
- Turn on GPS by moving switch to USB position
- Check microphone
    - DATA light should flash
- Check GPS
    - STANDBY light should flash
    - This is very important if the GPS is not working check the power source and the GPS cable connection to make sure nothing has come unplugged
- Fill out data sheet

Step 6: Drive Transect

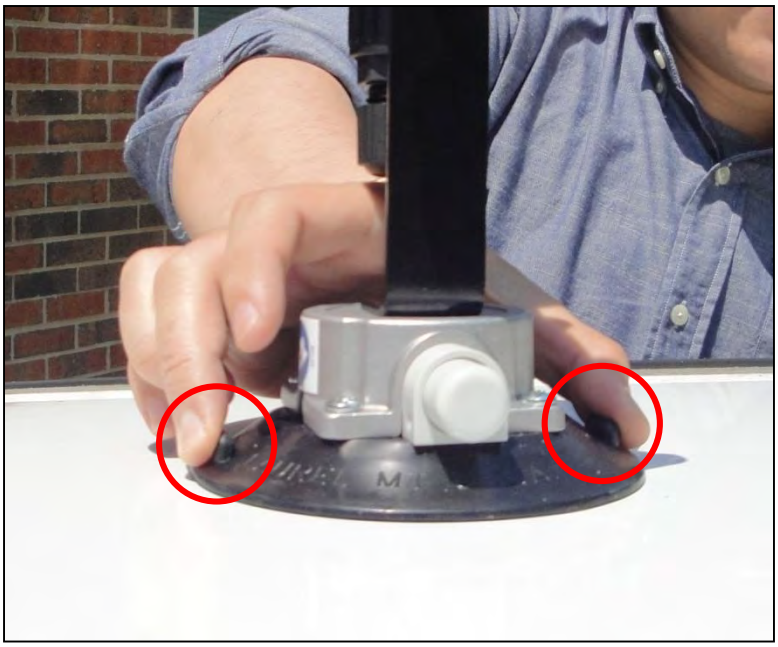
- Follow transect driving directions
- Monitor lights on the AnaBat SD2 throughout the transect
    - Make sure the LOW BATTERY light does not turn on
    - **Make sure the STANDBY light is flashing throughout the transect if it is not flashing GPS coordinates are not being recorded**
    - If the STANDBY light stops flashing pull over, when safe, check the GPS power source (either GPS battery pack or inverter) and the GPS cable connection

Step 7: After Driving the Transect

- Turn off AnaBat SD2 by pressing power button
- Turn off GPS by moving switch to off position
- Complete data sheet

Step 8: Remove Car Mount and GPS

- Squeeze black rubber tabs on both sides of suction cup to release



- Suction cup is released when red line appears

- Remove the GPS and car mount from vehicle top

North American Bat Monitoring Program – North Carolina Division

Driving Transect Safety and Instructions Document

Safety

1) Obey traffic laws
2) Keep hazard lights on while driving at 20mph
3) Use headlights
4) Pull over to take notes (if driver is alone)

Instructions

1) Stop AnaBat SD2 for…
    a. Any long unexpected stops
        i. For example
            - An accident
            - A train at railroad crossing
            - Construction zone
    b. When instructed to by transect directions
        i. For example
            - The transect uses a busy highway and it is unsafe to drive 20mph
2) Do NOT stop AnaBat SD2
    a. Traffic light
    b. Stop sign
    c. Pulling over to let other vehicles pass
    d. Short unexpected stop or speed change
        i. For example
            - A deer runs in front of your vehicle
            - A turning vehicle is in front of you
            - The road is curvy and you need to go slower than 20mph
3) Cancel the survey in the case of rain

North American Bat Monitoring Program – North Carolina Division

Driving Transect Data Sheet

Drivers Name: _____

Assistants Name (optional): _____

Date (mm/dd/yy): _____

Sunset time: _____ Moonrise time: _____

***Fill in before you drive the transect**

Grid Cell ID # : _____ AnaBat SD2 # : _____

Transect Name (on directions): _____

	Time	Temp. (°F)	Wind Speed	Humidity (%)	Moon Visible (Y/N)	Cloud Cover (estimate %)
Start						
End		***Fill in after you drive the transect**				

Moon Phase (circle one): New ¼ ½ ¾ Full

Notes:

***Fill while driving/after driving the transect**

North American Bat Monitoring Program – North Carolina Division

Driving Transect Data Sheet

Drivers Name: _____

Assistants Name (optional): _____

Date (mm/dd/yy): _____

Sunset time: _____ Moonrise time: _____

Grid Cell ID # : _____ AnaBat SD2 # : _____

Transect Name (on directions): _____

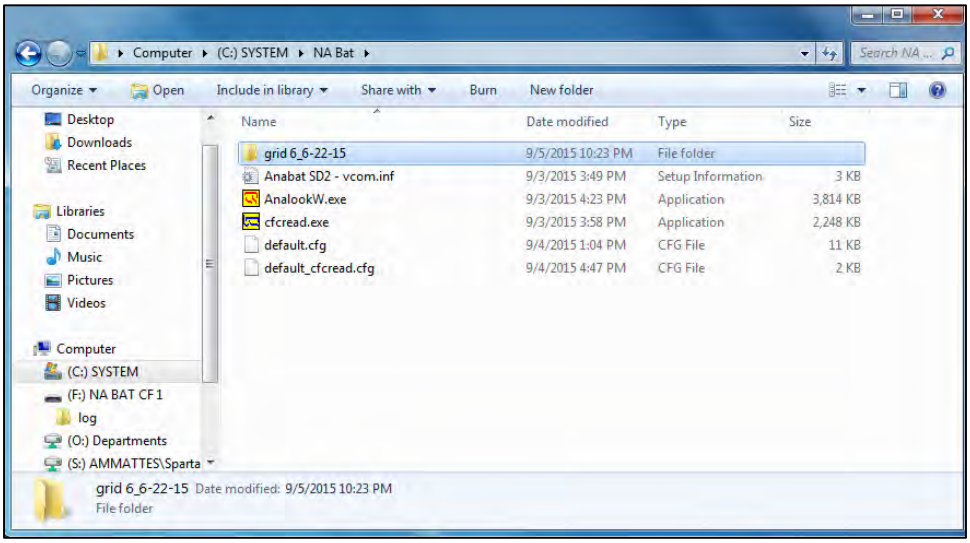
	Time	Temp. (°F)	Wind Speed	Humidity (%)	Moon Visible (Y/N)	Cloud Cover (estimate %)
Start						
End						

Moon Phase (circle one): New ¼ ½ ¾ Full

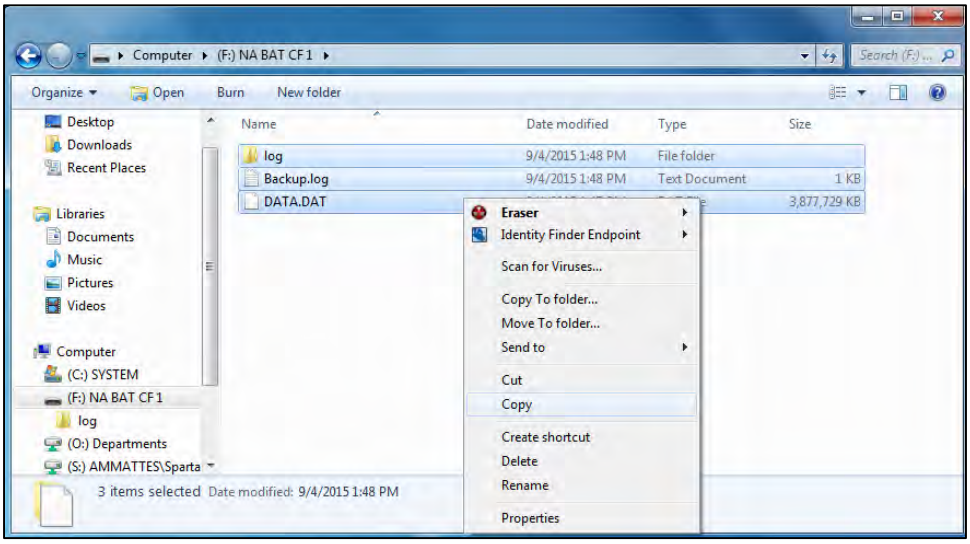
Notes:

Back-Up Raw Data

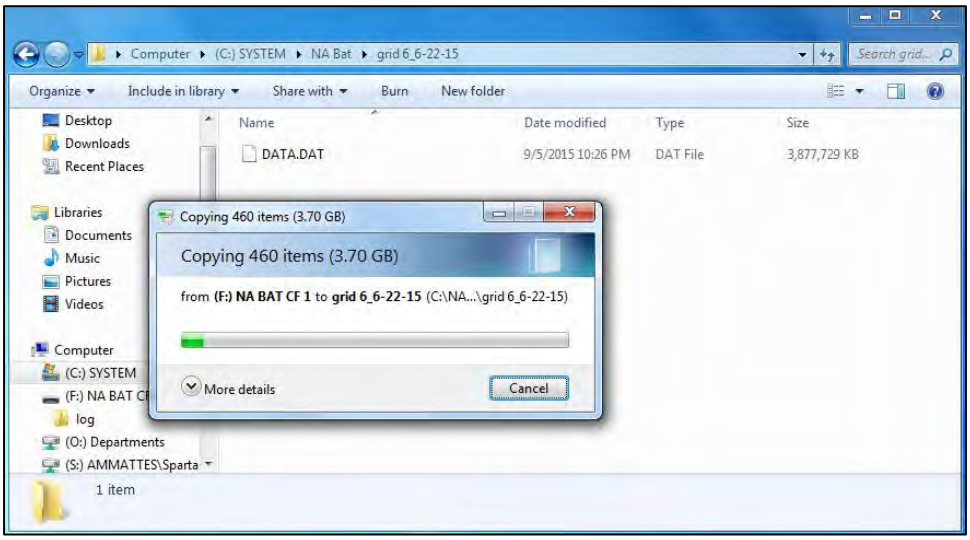
- Before you download data you should back up the raw data
- Use the card reader to plug the CF card into your computer
- Go to 'My Computer'
- On your computer's hard drive create a new folder
    - Name the folder grid cell ID number_date sampling occurred



- Open the CF card
- Select all of the files on the card
- Copy all files

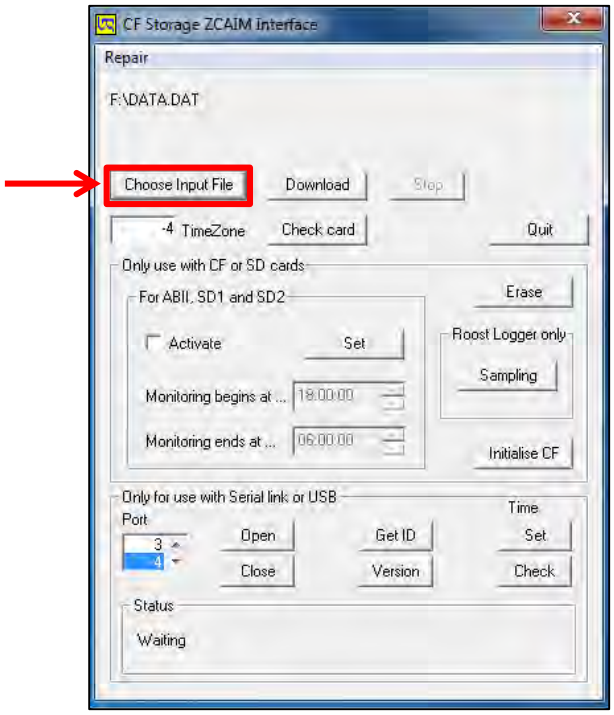


- Paste files into the folder you created on your computer's hard drive

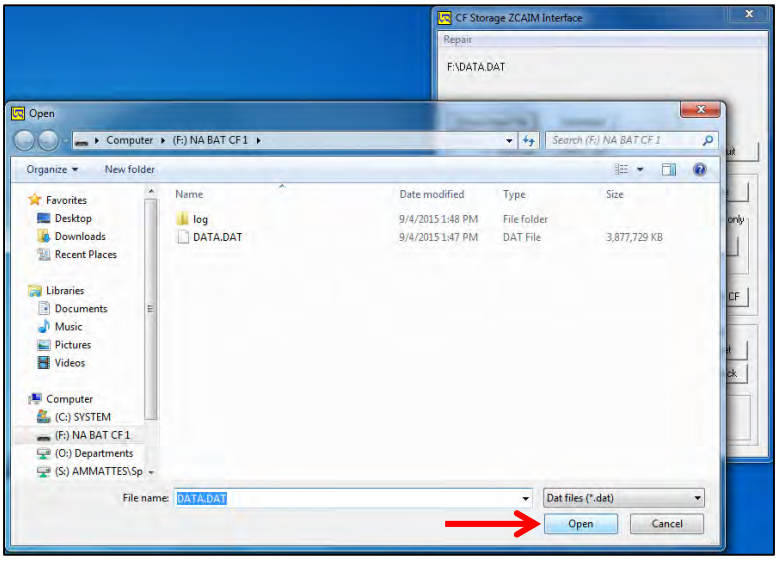


Downloading Data

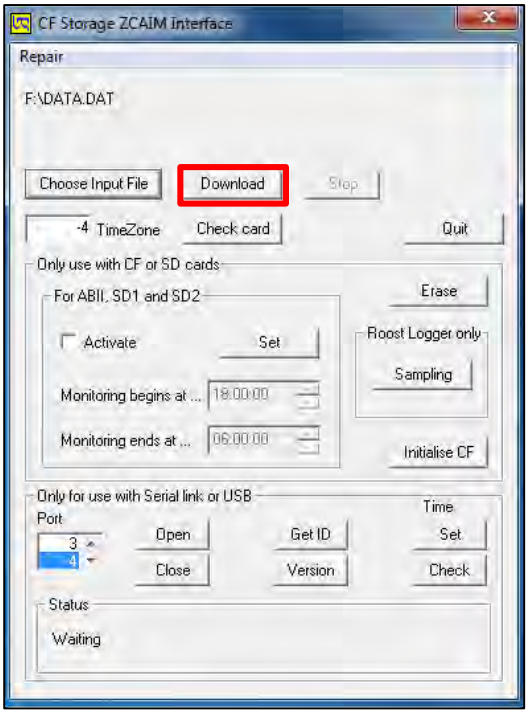
- Use the card reader to connect the CF card to the computer
- Open CF_read.exe



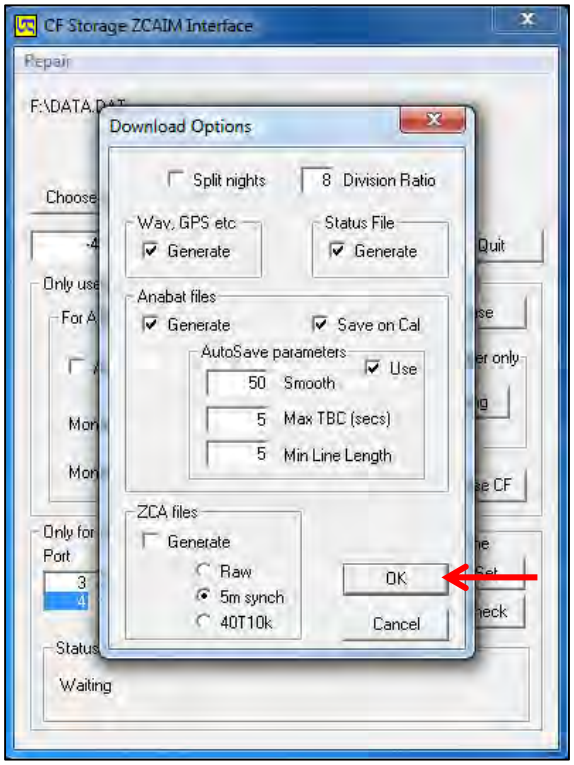
- Click ‘Choose Input File’
- New window opens
- Make sure DATA.DAT file is selected
- Click ‘Open’



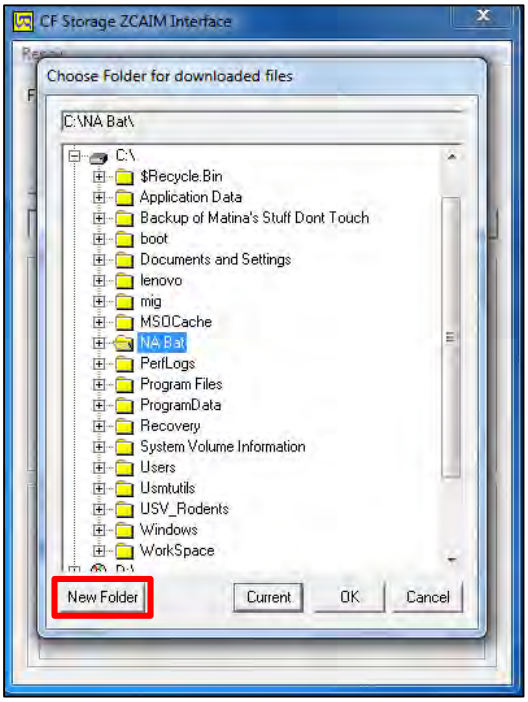
- Click ‘Download’



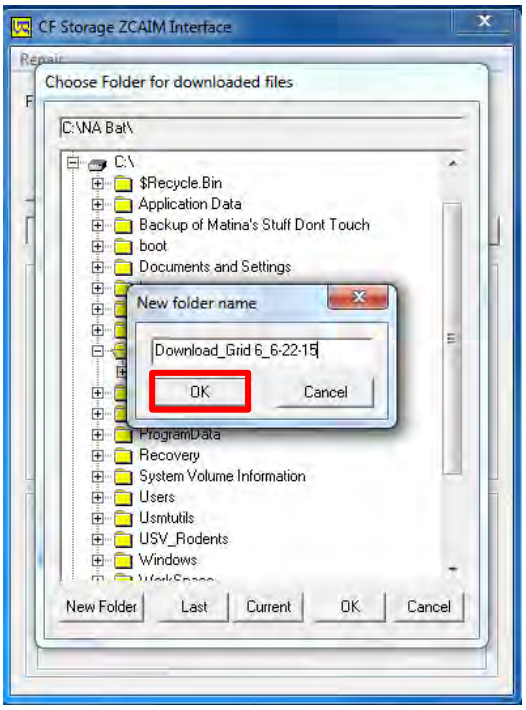
- New window opens
- Make sure the settings are as pictured below
- Click ‘OK’



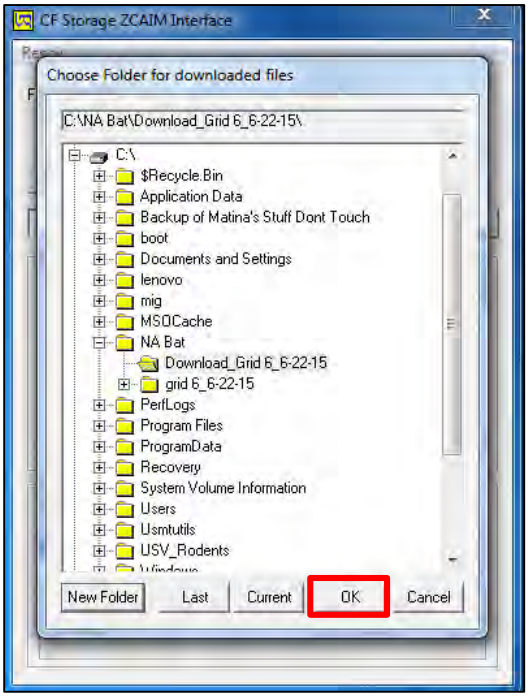
- New window opens
- Choose folder to save file on your computer's hard drive
- Click 'New Folder'



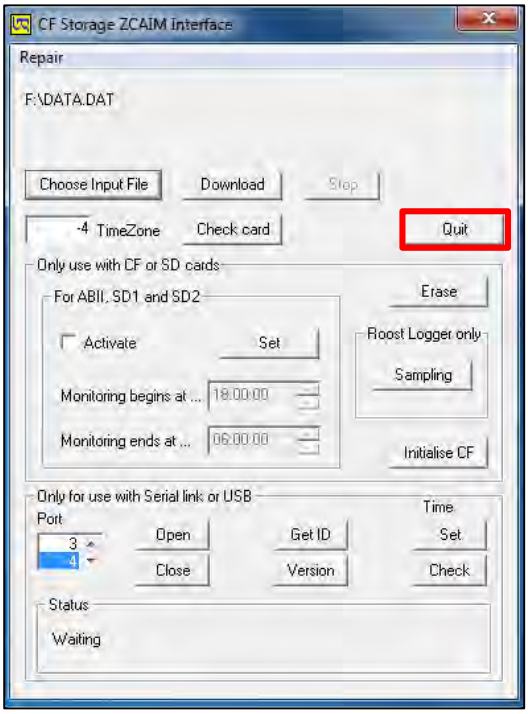
- New window opens
- Name the new folder Download_Grid Cell ID #_date
- Click 'OK'



- Click ‘OK’



- Download is complete when the Valid Anabat sectors read number stops increasing
- Click ‘Quit’



Stationary point survey guide

North American Bat Monitoring Program North Carolina Division

Han Li, PhD

University of North Carolina Greensboro

NABat stationary survey general protocol

- Within a pre-defined 10 km by 10 km grid cell, 2 -4 sites need to be sampled with stationary point acoustic detectors;
- All sites need to be monitored simultaneously from sunset to sunrise for 4 consecutive nights, regardless of weather;
- All monitoring should be conducted in June or July

Equipment

Please notice that any equipment suggested in this guide is based on the pilot study conducted by the University of North Carolina Greensboro and the North Carolina Wildlife Recourses Commission. No commercial interest is involved in equipment supplier choice.

Authors of this guide do not endorse any particular brand of equipment for the North American Bat Monitoring Program.

Prices listed in this guide are for budget guidance only.

AnaBat detector

- Current version: SD2
- Only available from Titley Scientific for $1998 per detector
  - Order 10 or more detectors receive 10% discount

Titley Scientific water proof setup

- Including: 1400 Pelican case configured for detector and 12 volt battery, internal cabling, battery connector lead, 3m microphone extension cable, weather proof weather head with stainless microphone
- Only available from Titley for $530 per setup

Battery and charger

- 12 volt 7.5 ah lead acid battery and charger
  - Fit into Titley Scientific water proof setup
  - Can last at least 10-15 nights of continuous recording
  - Take less than 24 hours to charge
  - Available from online merchandisers or local stores, including Titley Scientific
- AA battery
  - 4 batteries are needed for 1 detector
  - Can last a few nights of recording
  - NOT recommended for stationary survey

Memory card/card reader

- CF card/card reader
  - Most likely available from online merchandisers
  - A 4G CF card can hold at least 2 months of nightly AnaBat recording
  - A CF card reader is necessary for most newer computers

Microphone mounting gears

- Three common ways to mount the detector microphone for recording:
  - Tripod
  - Tree strap
  - Pole

Microphone mounting gears

- Tripod
  - Suitable for monitoring corridor, forest interior, water body, open field, etc.
  - Not suitable for tall undergrowth
  - Any intro level tripod (58” or taller) can be used
  - Available from online merchandisers or local stores
  - Titley Scientific water proof microphone has been modified with a screw hole and ready to use

Microphone mounting gears

- Tree strap
  - Suitable for monitoring corridor, water body, edge, etc.
  - Require a tree with a tall trunk, cannot be used on trees with low branches or too young
  - Only available from Titley Scientific for $50 per strap

Microphone mounting gears

- Pole
  - Suitable for monitoring canopy top, water body, open field, tall undergrowth, etc.
  - Require T- post mounting bracket, poles (PVC pipes), rope, and other securing gears
  - T- post mounting bracket available from Titley Scientific for $50 per bracket; the Dual Angle Weather Head Mount ($40) is not recommended
  - Other items available from online merchandisers or local stores

Miscellaneous

- Pad lock and python lock
  - Secure the equipment
  - Available from online merchandisers or local stores
- Volt meter
  - Check battery condition
  - Available from online merchandisers or local stores
- Additional microphone extension cables at different length
  - Longer microphone extension cables might be needed for certain circumstances
  - Only available from Titley Scientific for about $10 per meter of cable

Equipment summary

- For one NABat grid cell:
  - 4 AnaBat detectors
  - 4 Titley Scientific water proof setups
  - 4 12 volt 7.5 ah lead acid batteries and chargers
  - 2 tripods, 1 tree strap, and 2 T- post mounting bracket
  - 4 4G CF card
  - 1 card reader
  - 4 pad locks, 4 python locks
  - 1 additional backup 3m (5m) microphone extension cable
  - 6 1.5 m PVC pipes, 3 PVC pipe connectors, rope, hooks, etc.

Detector setup

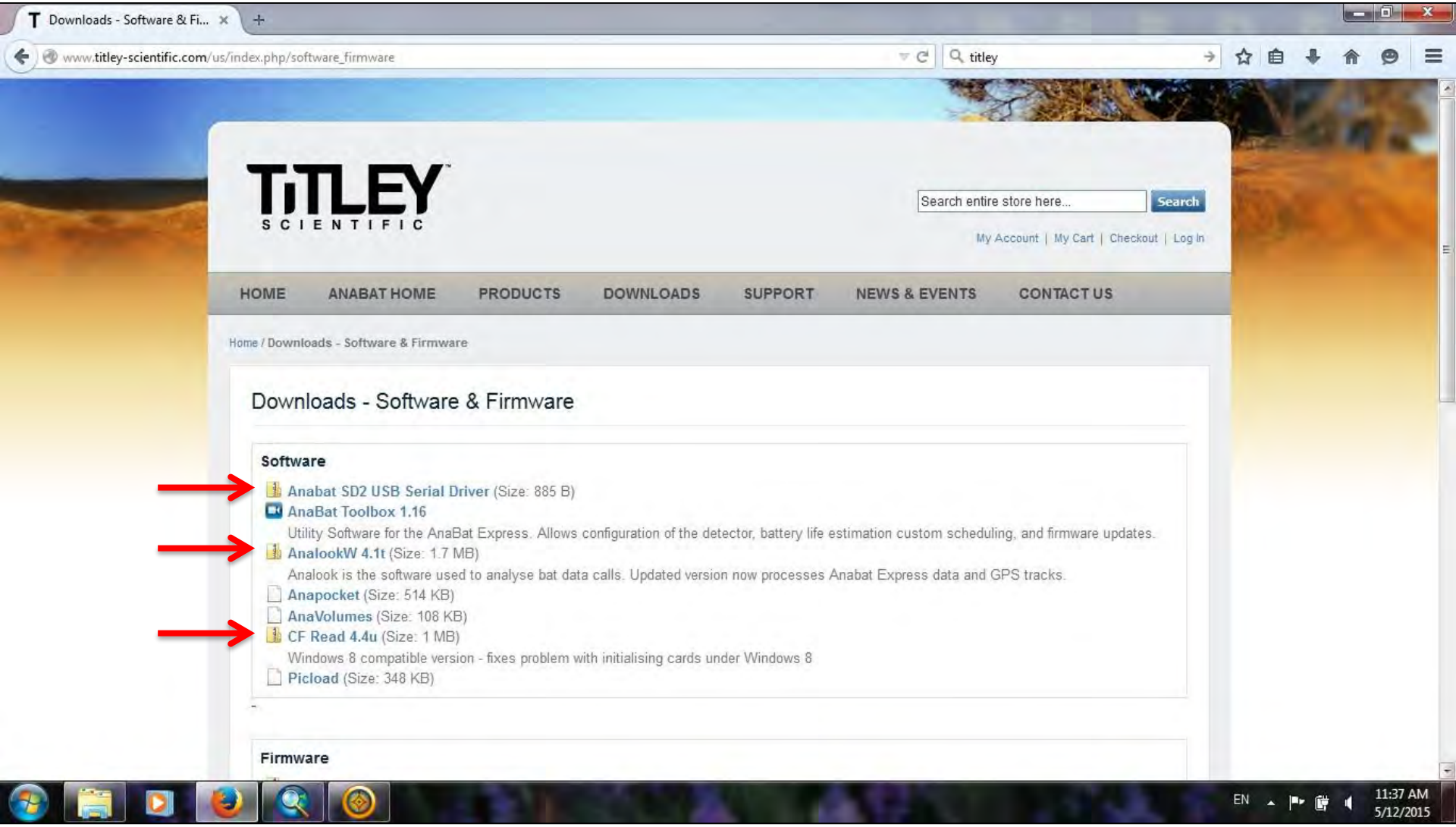
Downloading All Software

- AnaBat software can be downloaded from:

http://www.titleyscientific.com/us/index.php/software_firmware

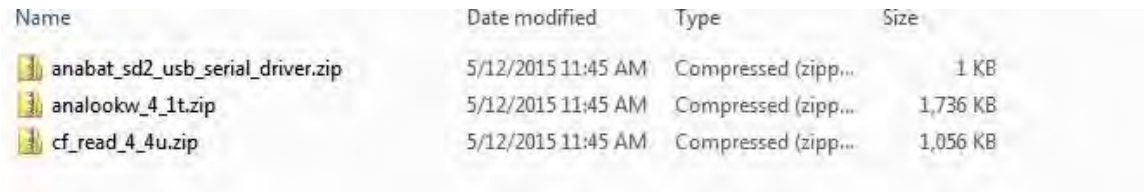
- Three items are necessary:
  - AnaBat SD2 USB Serial Driver
  - CF Read 4.4u
  - AnaLookW 4.1t

Downloading All Software

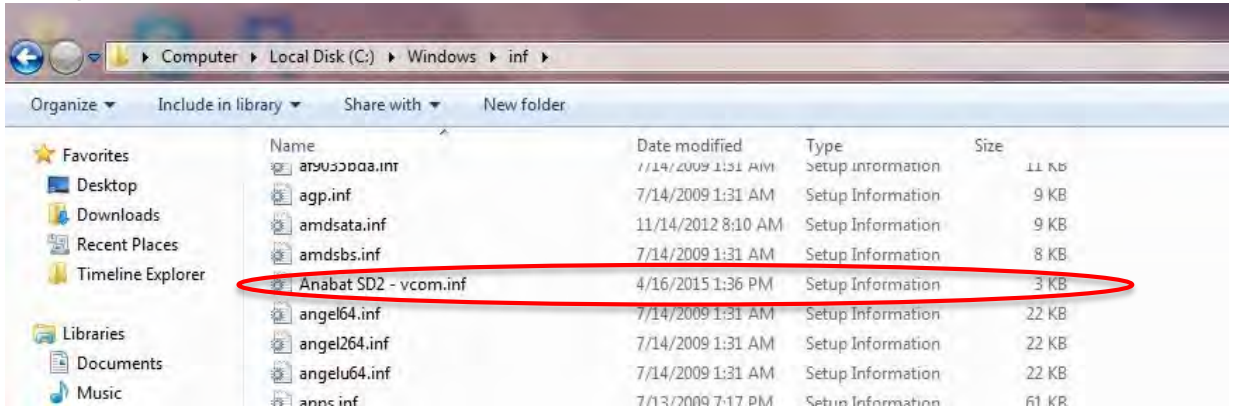


Installing Serial Driver

- AnaBat SD2 USB Serial Driver is needed to connect the detector to a computer (PC only) via a USB cable.
- Unzip the file

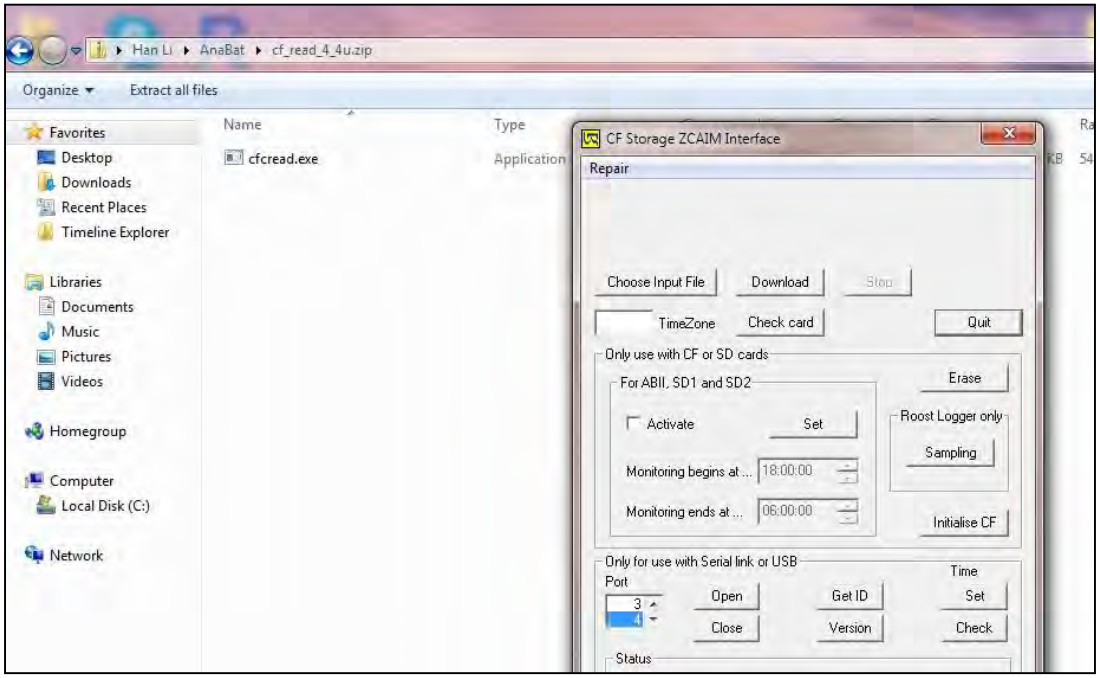


- Copy and paste the driver file (Anabat SD2-vcom.inf) to your local drive (C: or D:) /Windows/inf

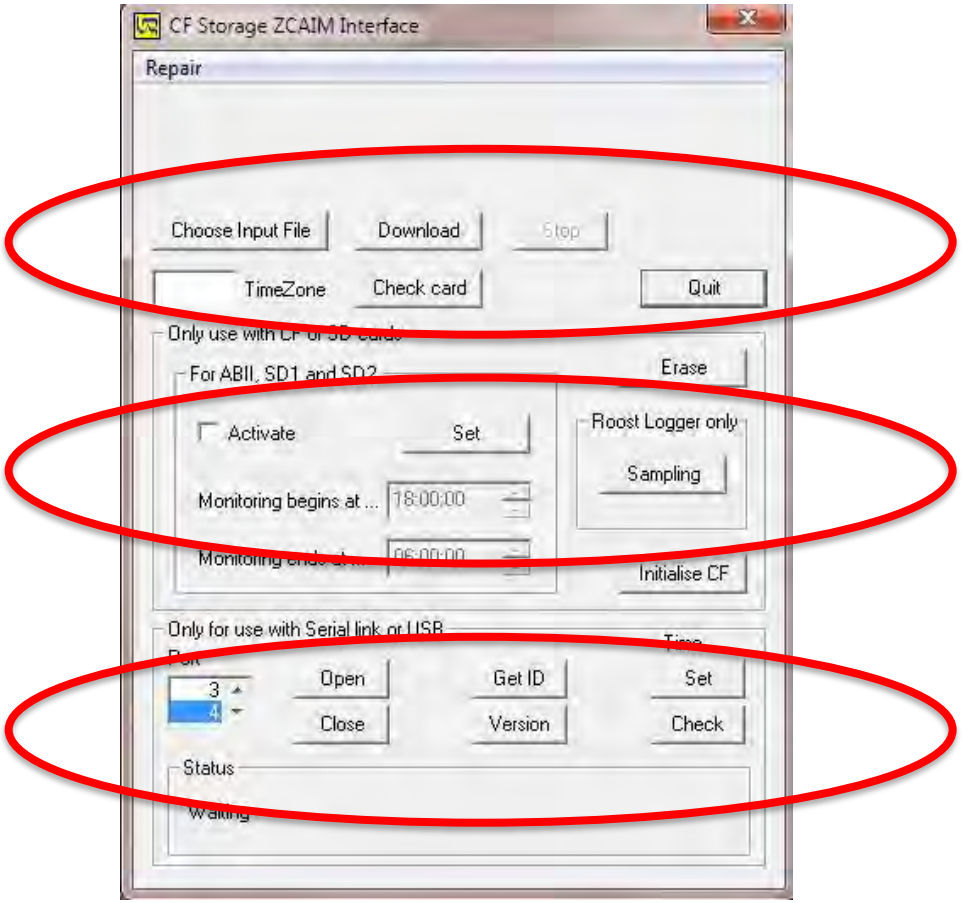


Installing CF Read 4.4

- CF Read 4.4 is used to:
  - Initialize CF cards, download data, and synchronize internal clock
- Unzip the file and open cfread.exe



Using CF Read 4.4



Download data

Initialize CF cards

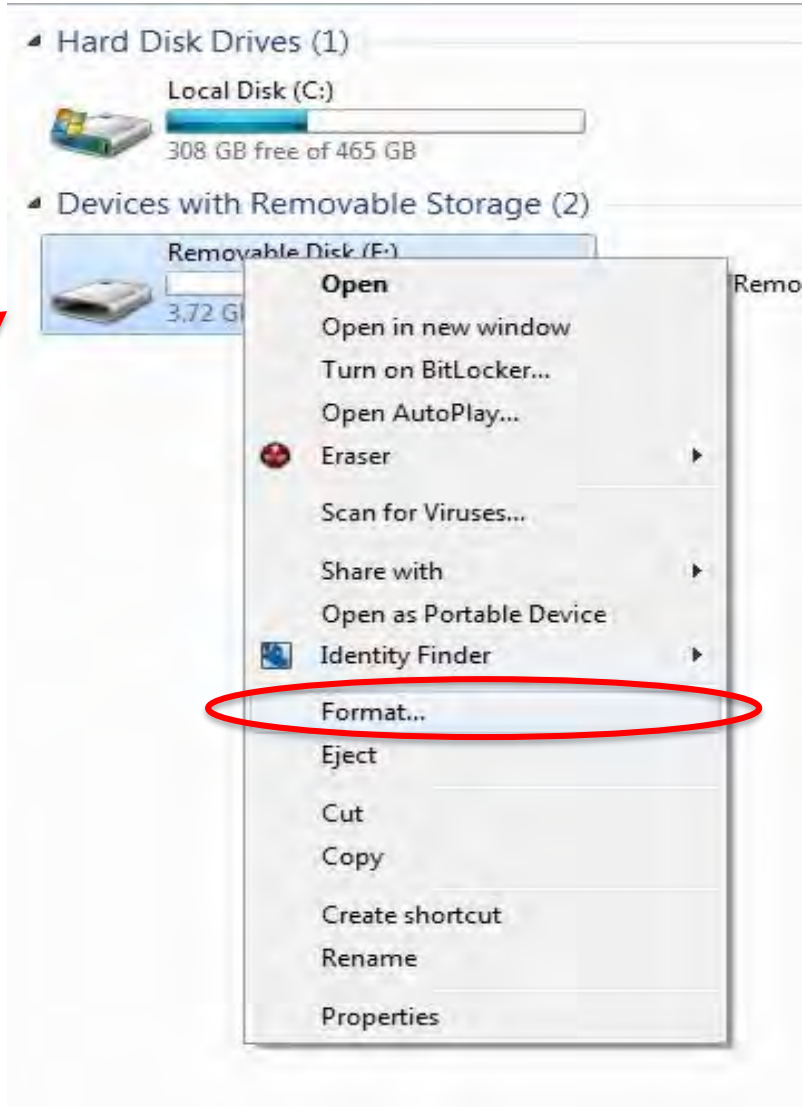
Sync internal clock

Setting Up Compact Flash Cards

If a CF card has not been used in AnaBat or has been used for other projects, it needs to be re-set for current project.

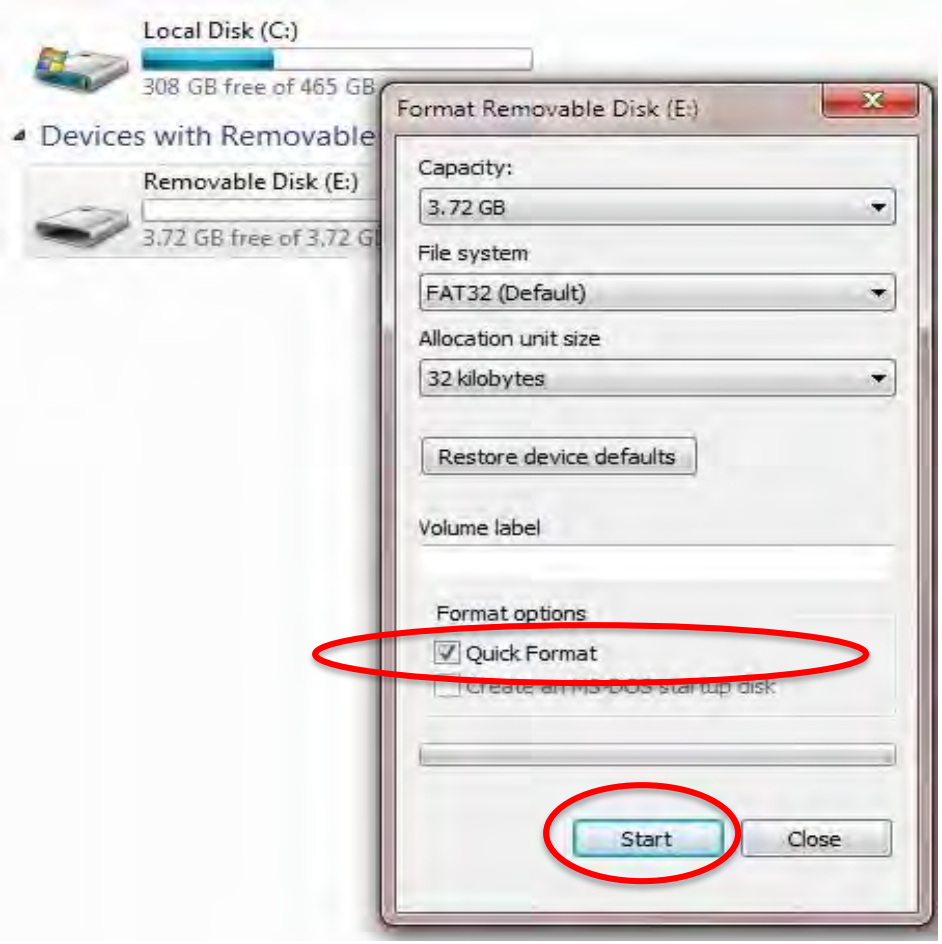
How to Format CF Cards

- Insert CF card into card reader
- Open My Computer
- Right click on Removable Disk
- Select Format



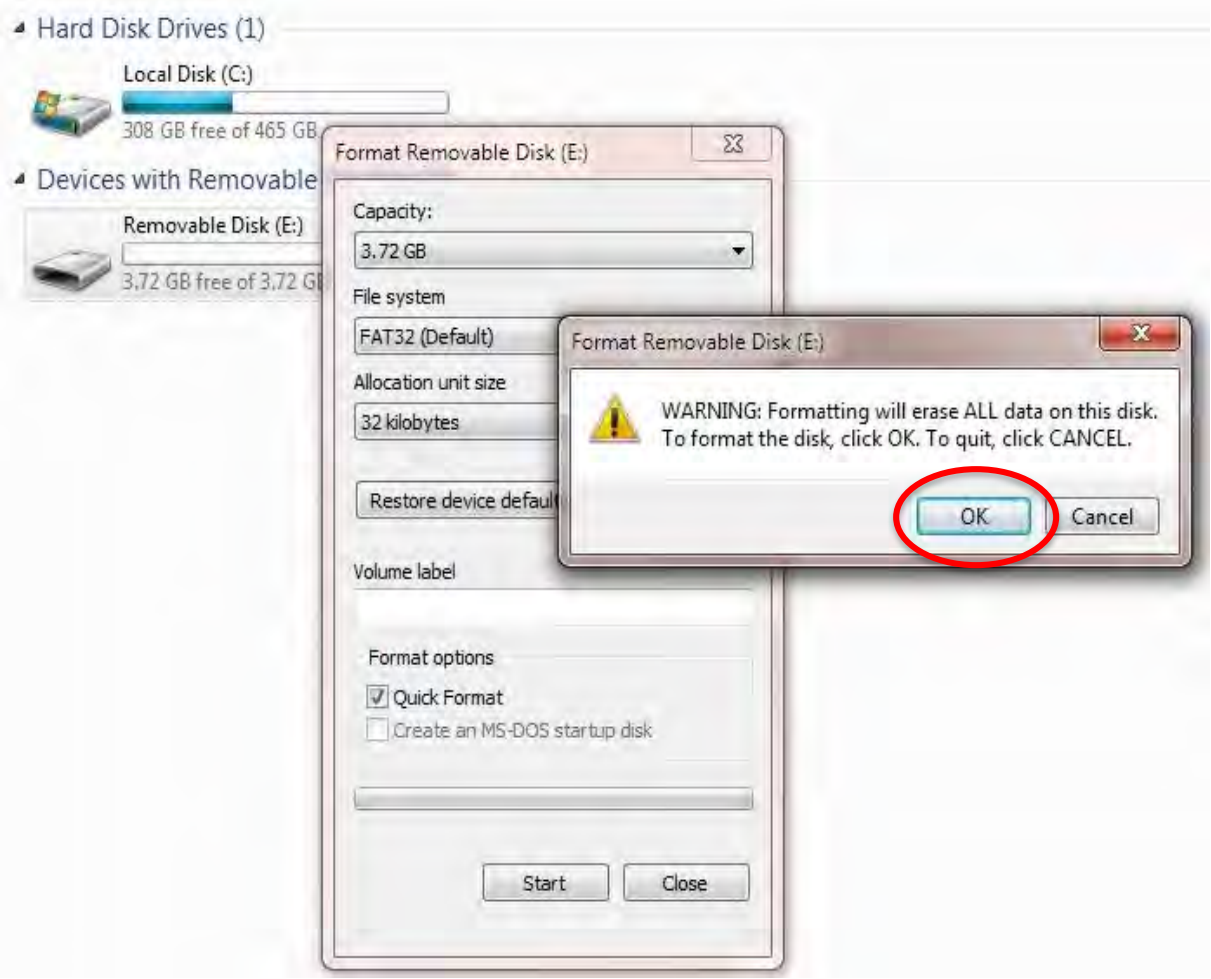
How to Format CF Cards

- New window opens
- Quick Format box should be checked
- Click Start



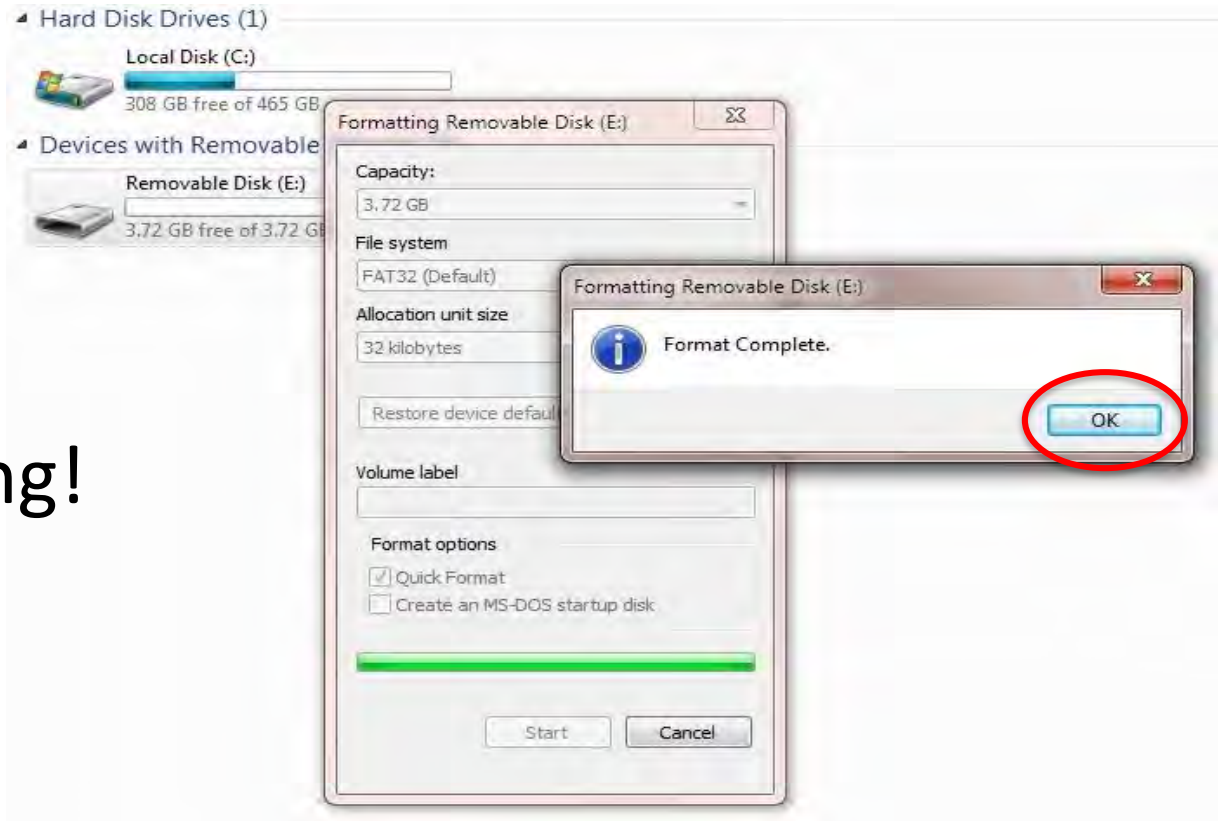
How to Format CF Cards

- Warning message will appear in a new window
- Click OK



How to Format CF Cards

- Format complete message will appear in new window
- Click OK
- Done formatting!

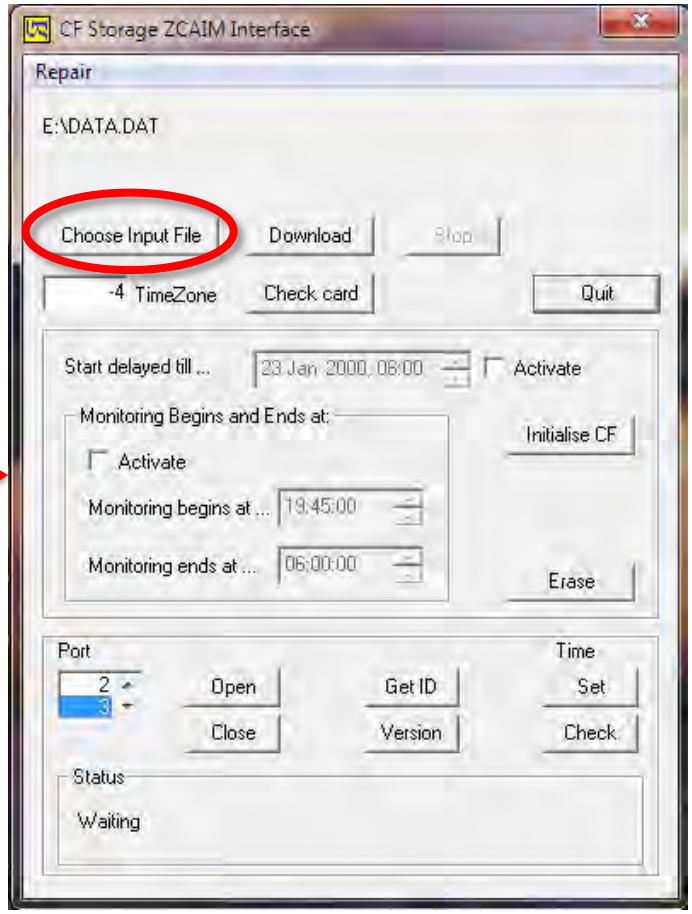


How to Initialize CF Cards

- Open cf_read.exe

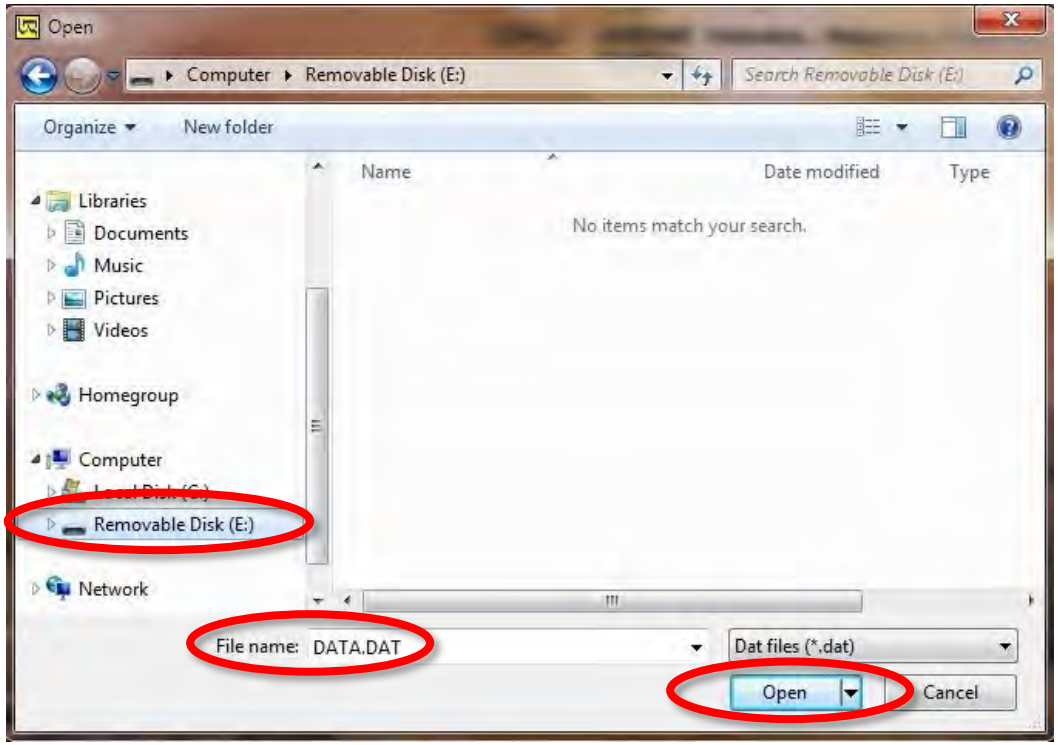
- New window opens

- Click Choose Input File



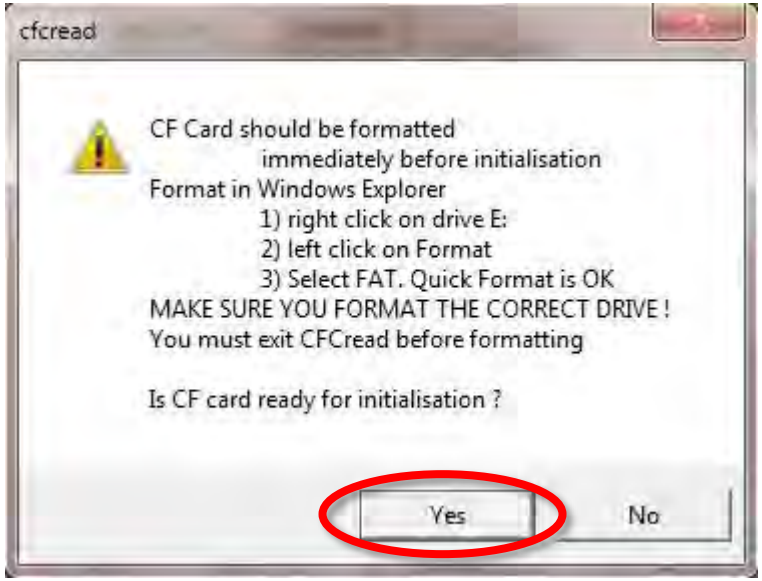
How to Initialize CF Cards

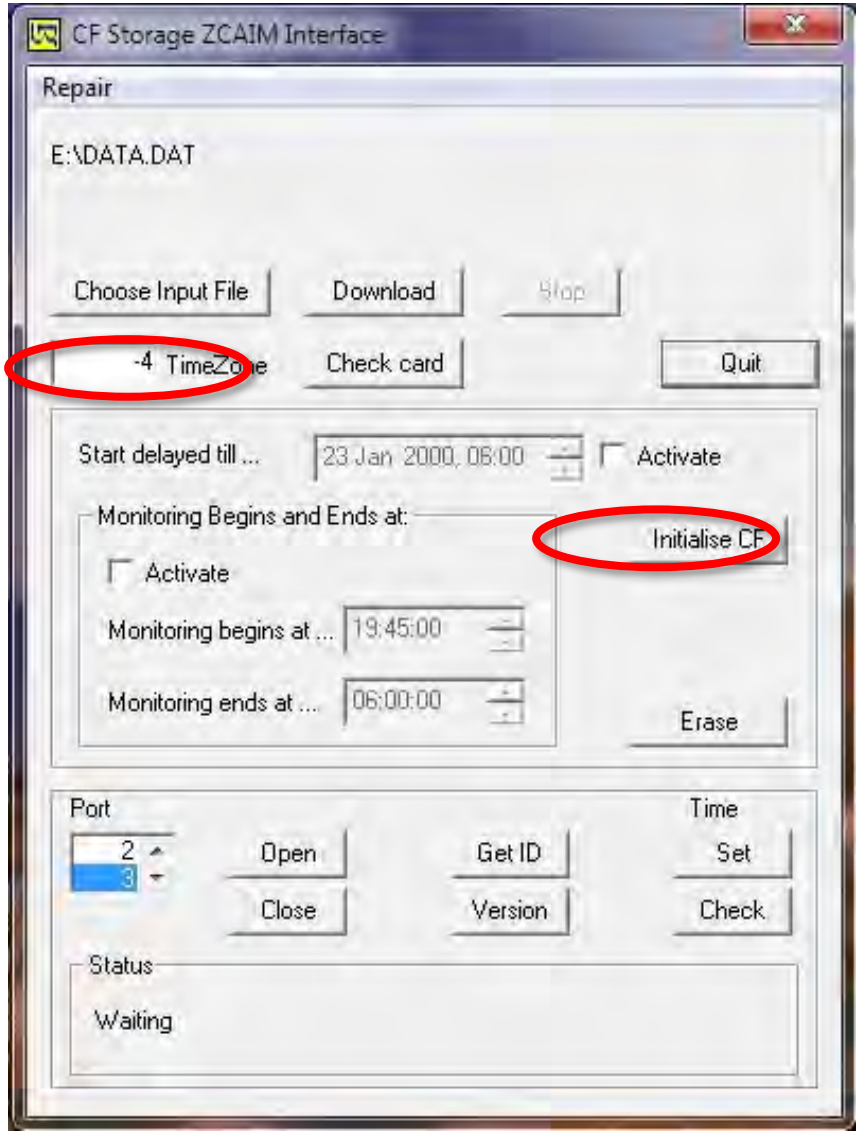
- New window opens
- Select the CF card in My Computer
- DATA.DAT file is automatically created
- Click Open



How to Initialize CF Cards

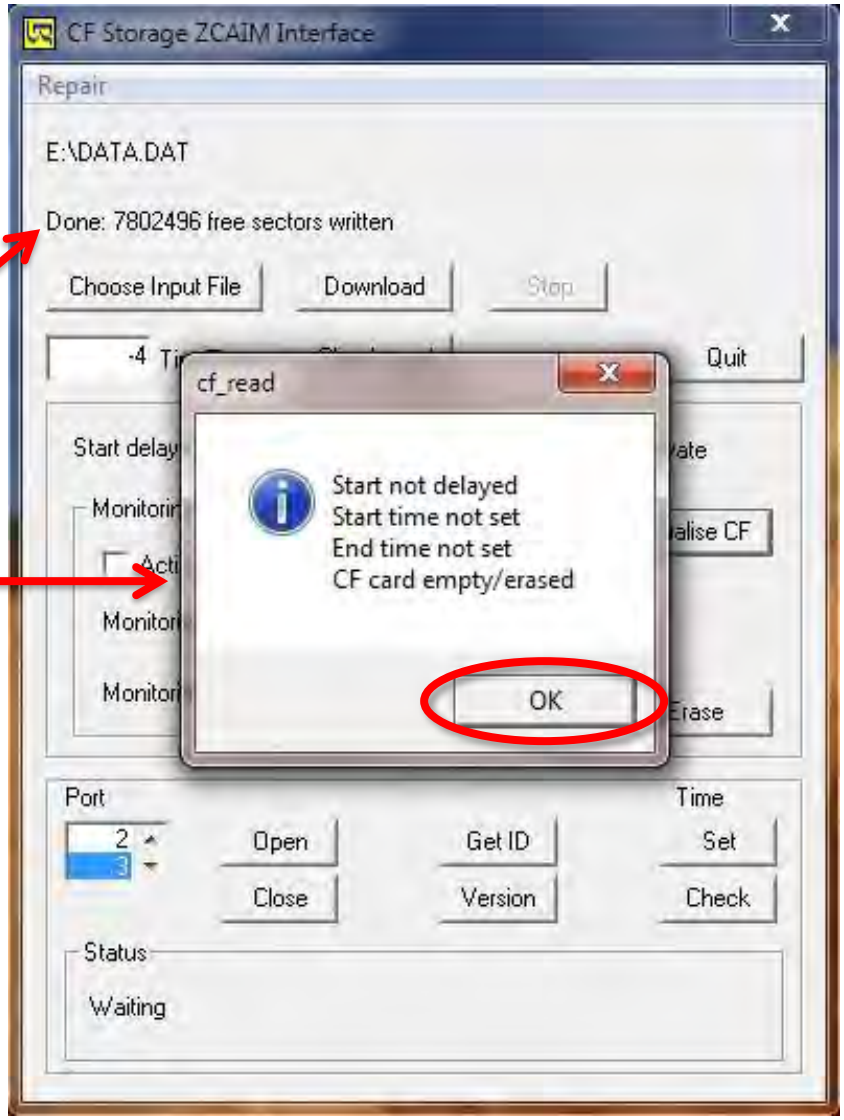
- Set TimeZone to -4 (NC)
- Click Initialise CF
- New window opens
- Click Yes





How to Initialize CF Cards

- Initialization process takes 5-10 minutes
  - This number increases
- Wait patiently
- New window opens when it is complete
- Click OK
- Done Initializing!



Setting Up AnaBat Internal Clock

If a detector has not been used for a few month, the internal clock needs to checked

How to Sync the Internal Clock

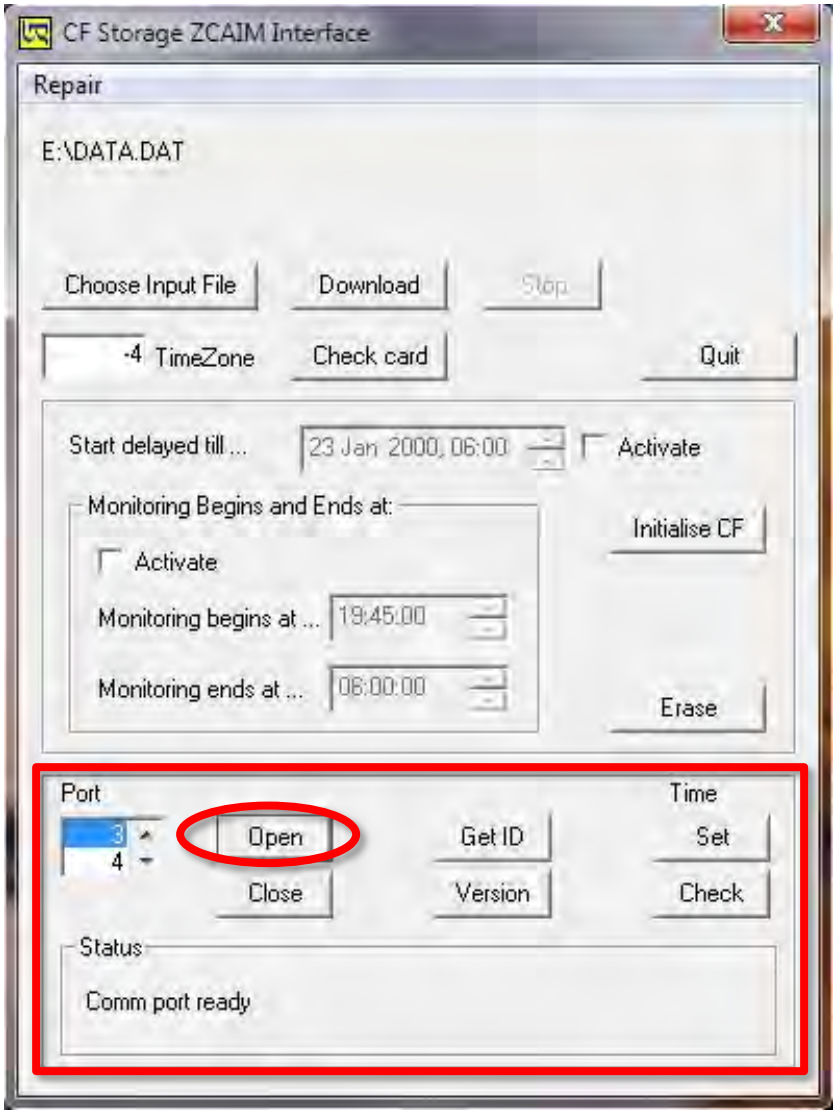
- Before you can do this:
  - 1) The serial driver must be installed
  - 2) CF card must be removed from the AnaBat
  - 3) Clock on your computer must be correct
    - The internal clock clock in the AnaBat will be synched to the computer's clock
- You will need:
  - USB cable
  - AnaBat
  - Computer

How to Sync the Internal Clock

- Connect the USB cable to the detector
- Turn on the detector BEFORE connecting it to the computer
- Connect the detector to the computer through USB port
- Open CF_read.exe

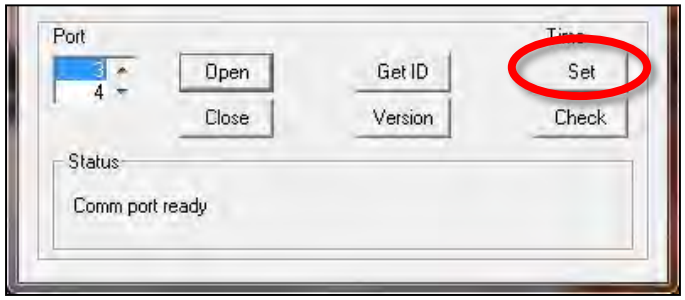
How to Sync the Internal Clock

- Select Port 3
  *sometimes 4
- Click Open
- It should display Comm port ready in Status
- If error message displays in Status repeat process with Port 4 selected

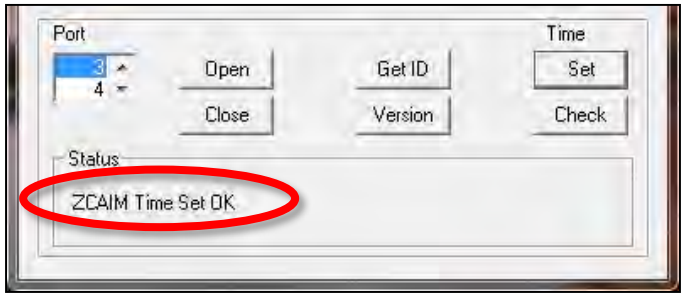


How to Sync the Internal Clock

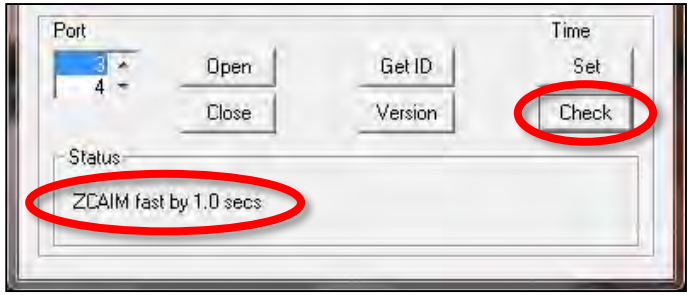
- Click Set



- In Status ZCAIM Time Set OK will show up



- Click Check



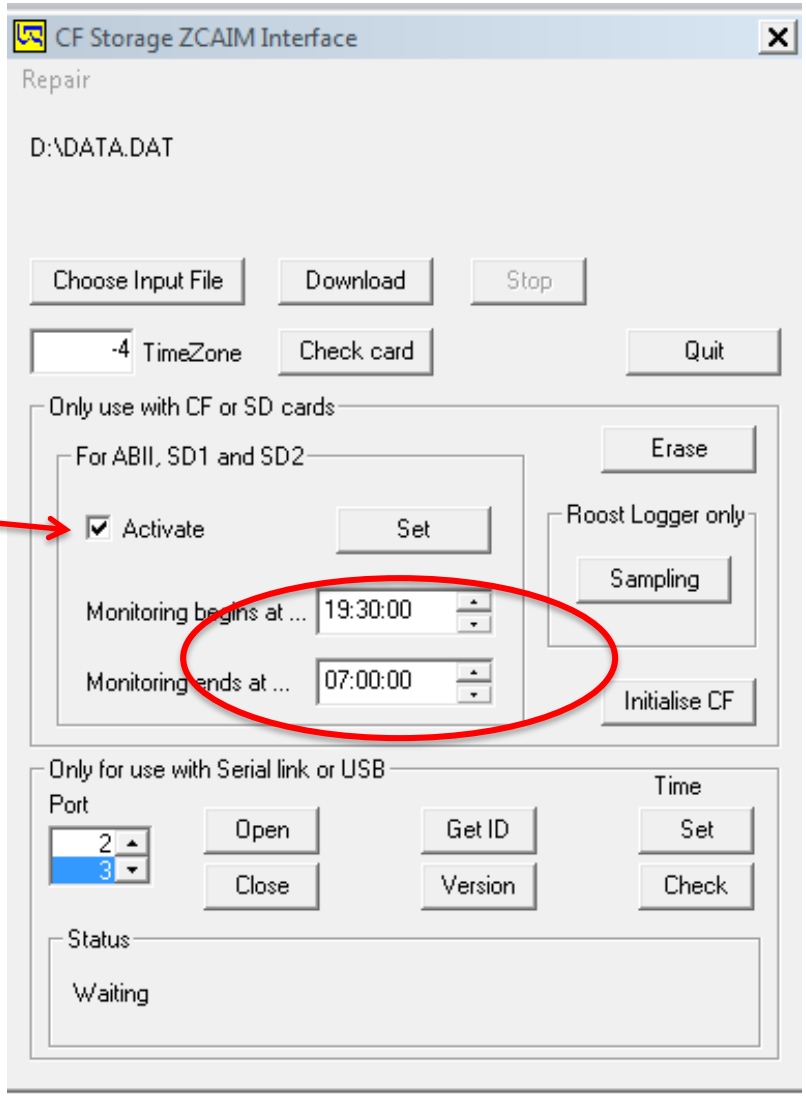
* ZCAIM fast by 1.0 secs is good

Setting Up Detector Recording Time

For a stationary survey project, the detector can be set to automatically turn on/off based on pre-set time. For summer monitoring in NC, 19:30-7:00 should be sufficient to cover sunset to sunrise.

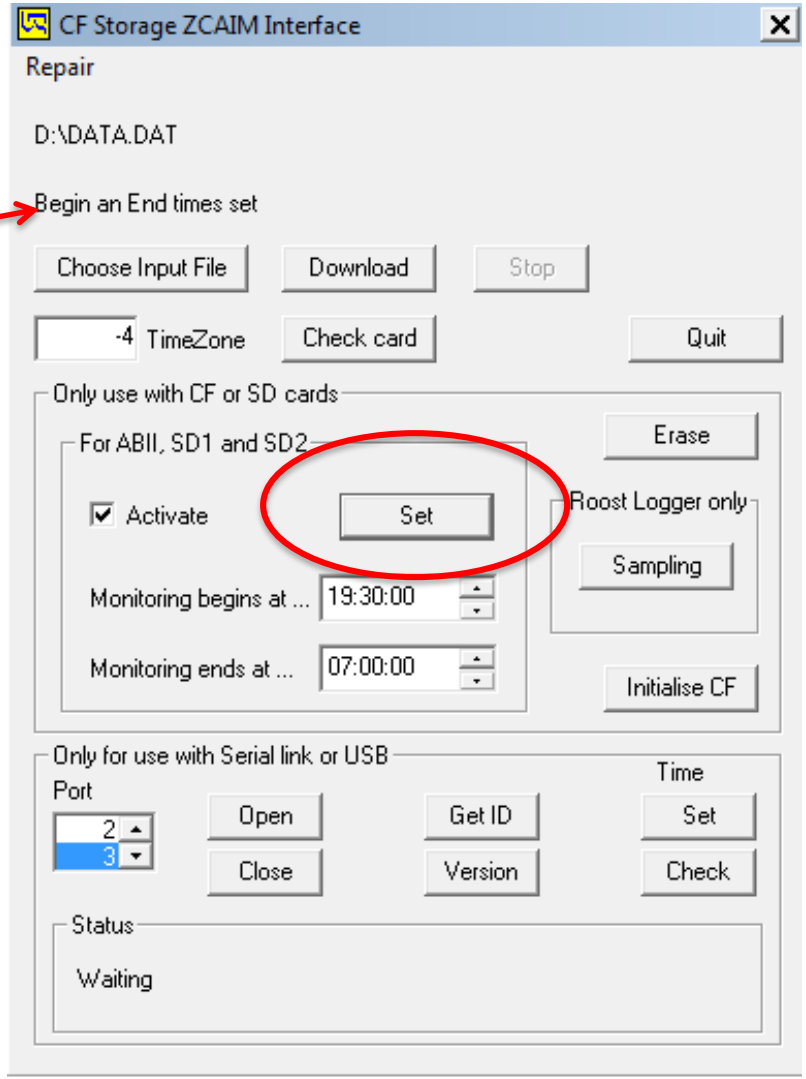
Set up monitoring time

- Insert CF card into card reader
- Open CF_Read.exe
- Check activate
- Enter time: 19:30:00 – 07:00:00 for summer monitoring in NC



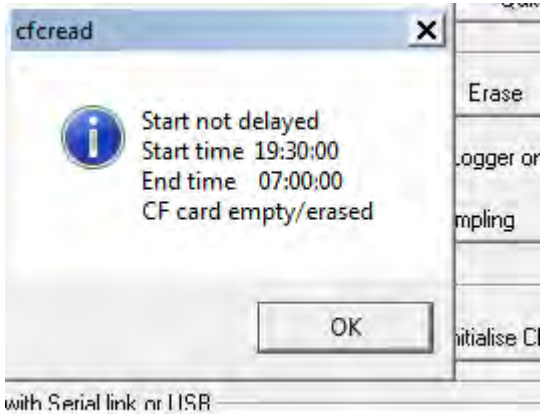
Set up monitoring time

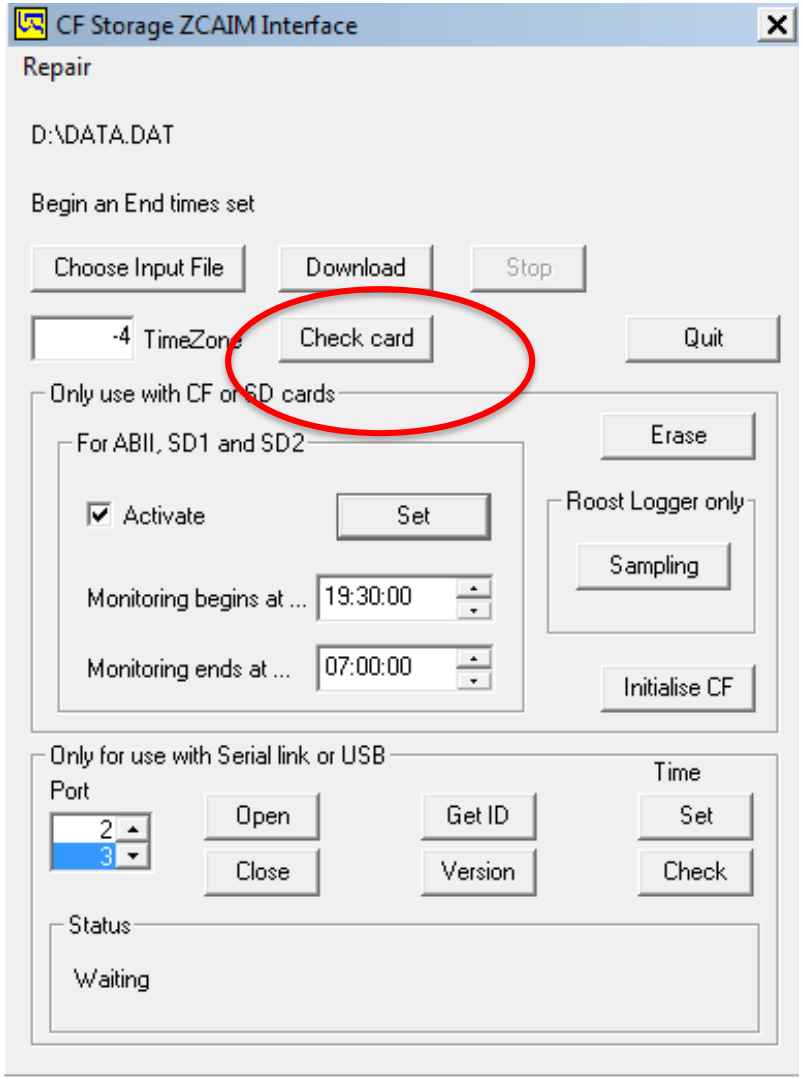
- Click set
- Software will show time set



Set up monitoring time

- You can check the time setting by clicking check card
- A small window will pop up and show the setting





Turn on AnaBat detector

Install CF Card

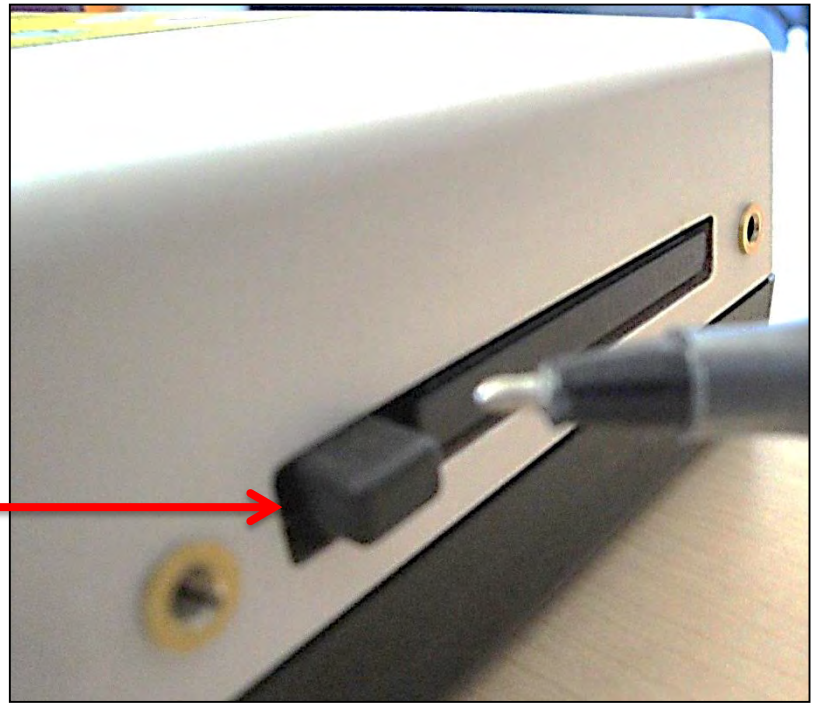
1) Unscrew Rear Panel

2) Remove Rear Panel

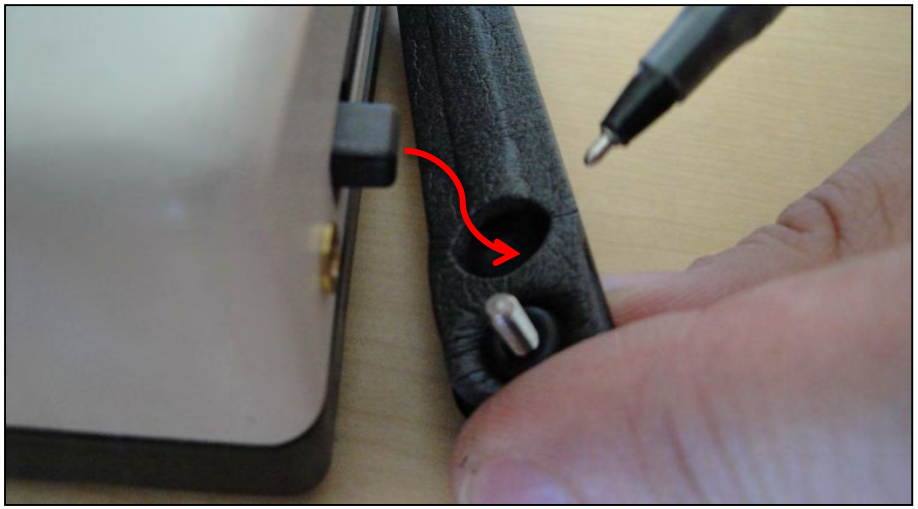
Install CF Card

3) Insert the CF Card <u>picture side up</u>

When inserted properly
black button will pop out

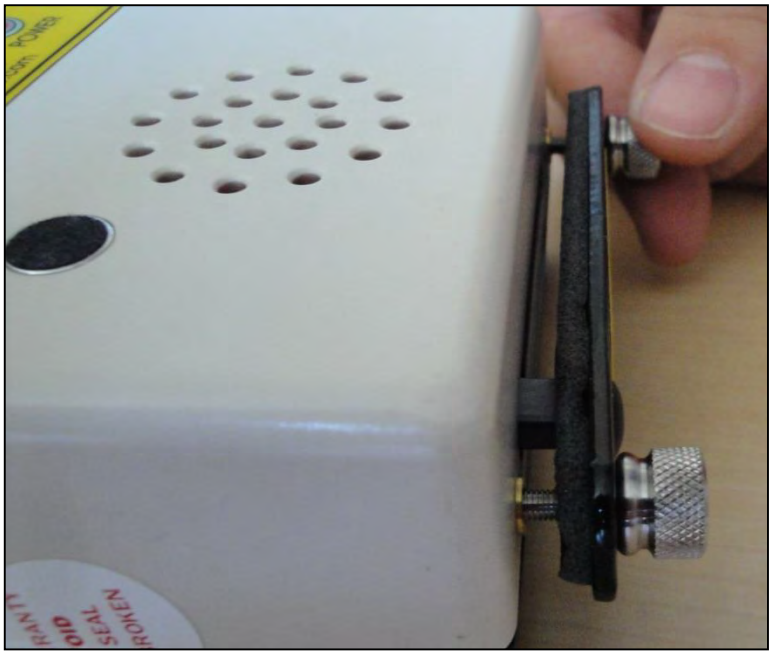


Install CF Card

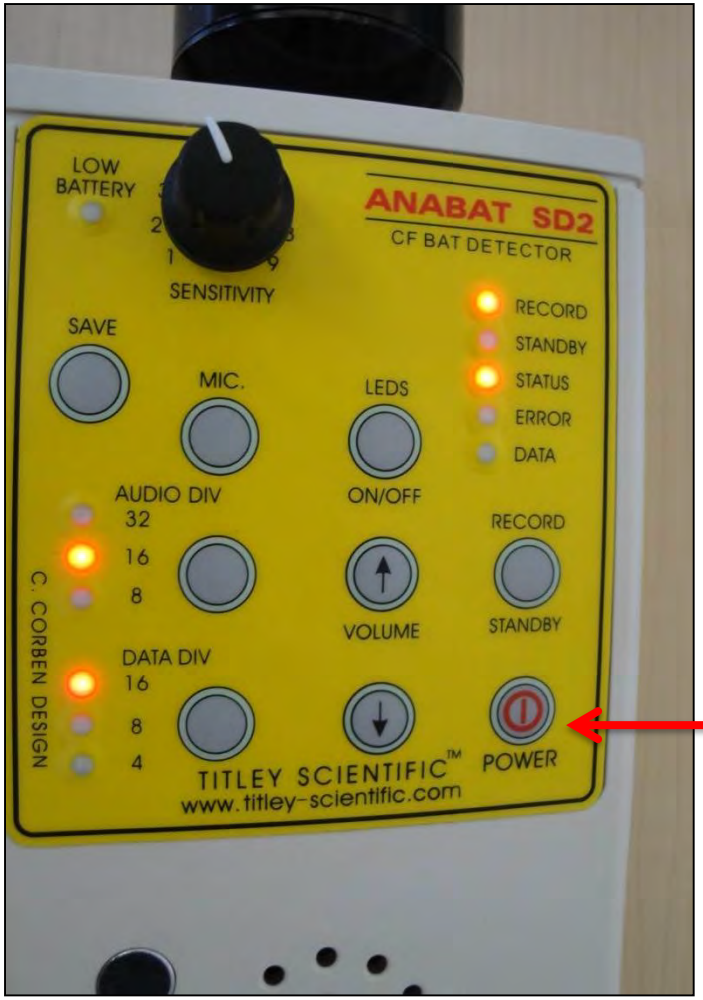


4) Replace rear panel

*button will fit in the indentation on the panel



Turn on AnaBat



Press POWER button

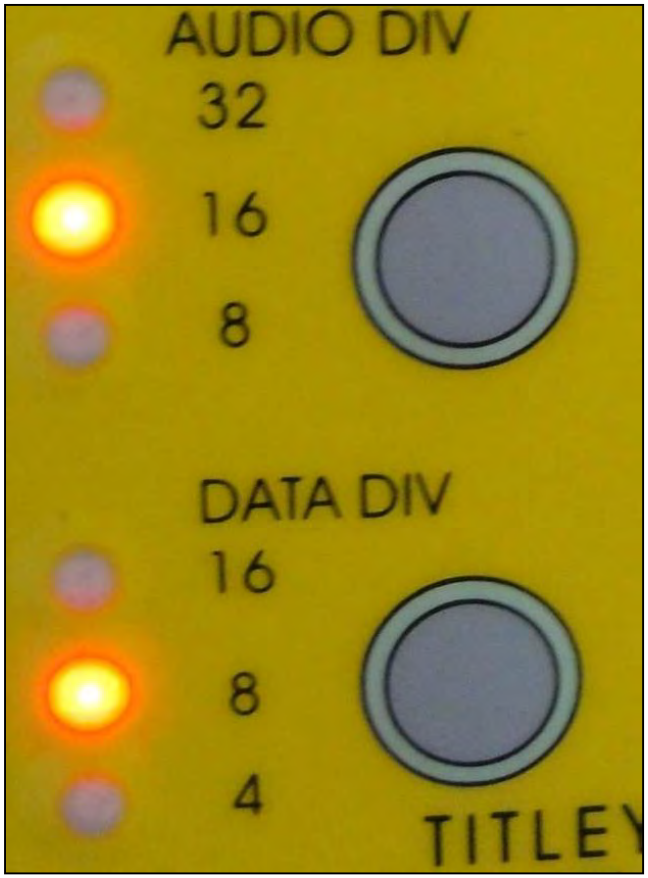
Wait 5 -10 seconds and if no lights come on press LEDS ON/OFF button

Indicator Lights

If the detector is turned on during pre-set recording hours (19:30:-7:00) RECORD light should be on; otherwise STANDBY light should be on for about 10-15 seconds. Then all lights will turn off and detector goes to sleep till the pre-set start time.

* If ERROR light is on, reformat and initialize the CF card (see previous section)

AUDIO DIV/DATA DIV

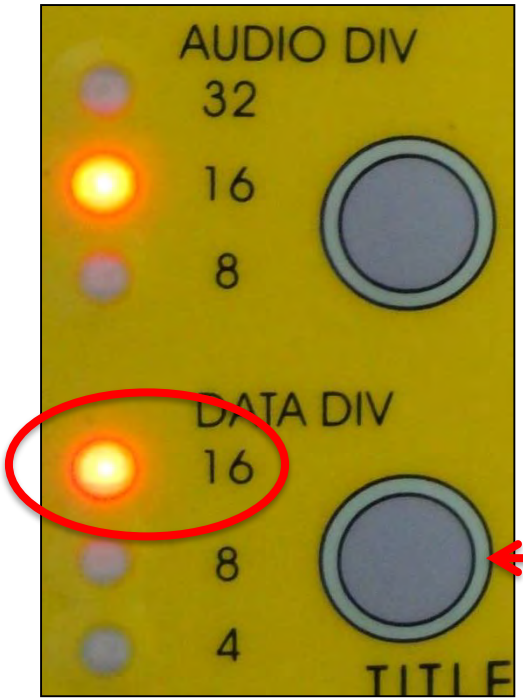


AUDIO DIV light should indicate 16

DATA DIV light should indicate 8

*If the light indicates other values see next slide

AUDIO DIV/DATA DIV

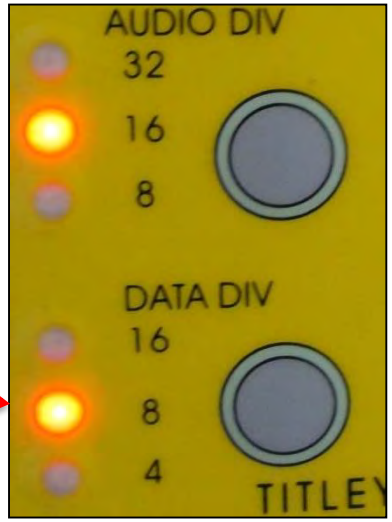


How to change AUDIO DIV or DATA DIV

Example:
DATA DIV is 16 but should be 8

Press button one time

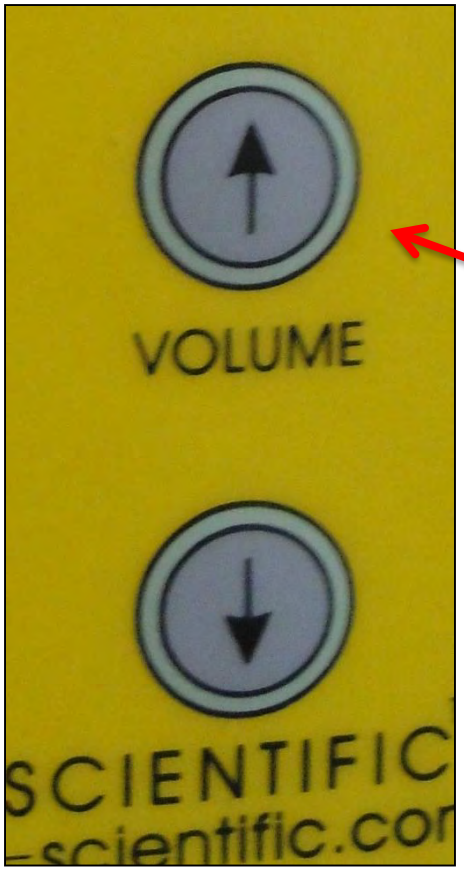
Indicator light should change to 8



SENSITIVITY Knob

SENSETIVITY should be set between 4 – 5

Volume



To confirm AnaBat is detecting noise snap fingers or shake keys in front of the microphone, you should hear some noise.

*If you do not hear anything adjust volume by pressing VOLUME up button.

* It is recommended to turn down volume for stationary monitoring to conserve battery

Site choice
and
Detector placement

Site choice

- Sites surveyed should represent the most typical habitat within a grid cell
- Please see NC site specific guide for more details

Detector placement

- The microphone needs to be at least 1.5m above the ground
- No leaves or branches near the microphone
- Do not point the microphone directly to vegetation clutters that are less than 5m away

Backing Up CF Card
and
Downloading Data from CF Card

Back-Up Data

- This step should be done after each 4 nights of stationary monitoring

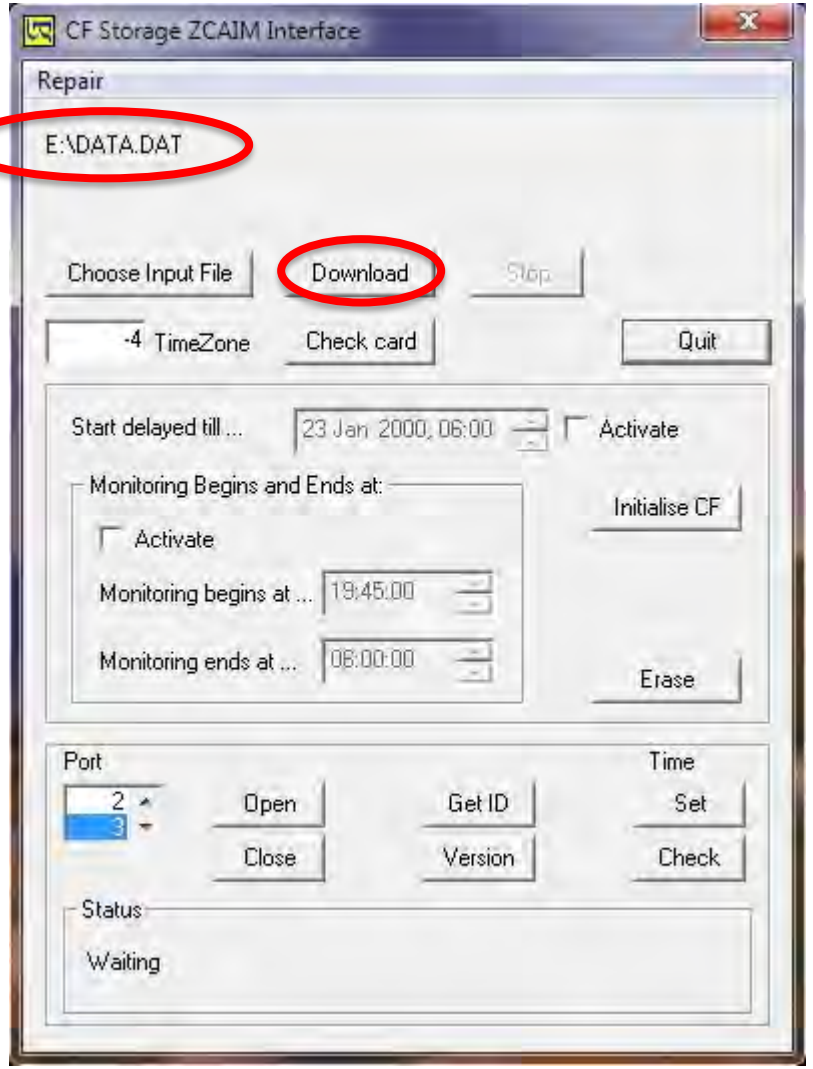
Back-Up Data

- Go to My Computer
- On your computer's hard drive create new folder
  - Name the folder accordingly
- Click on the CF card
- COPY ALL files
- Paste files into the folder you just created

Download Data

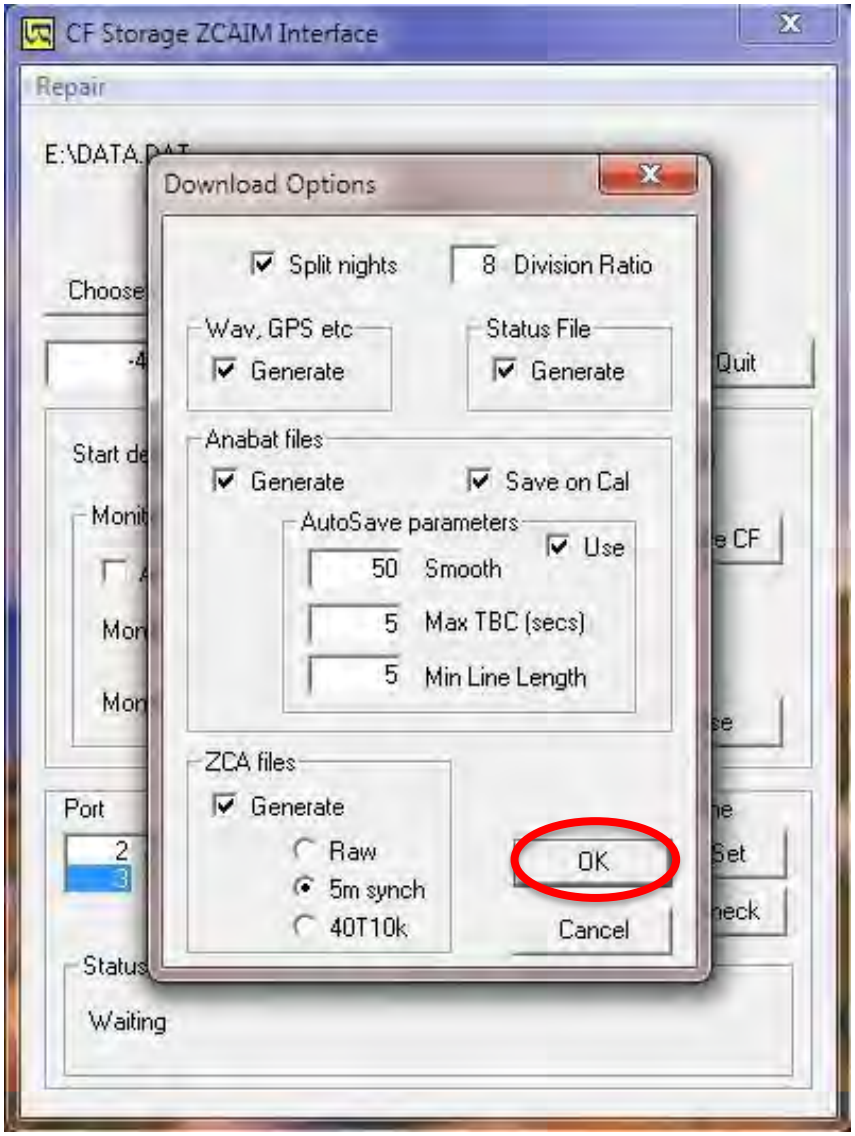
- Recordings stored in the CF card need to be downloaded
- Connect CF card to computer with card reader
- Open CF_read.exe 

- Input File should be DATA.DAT
- Click Download



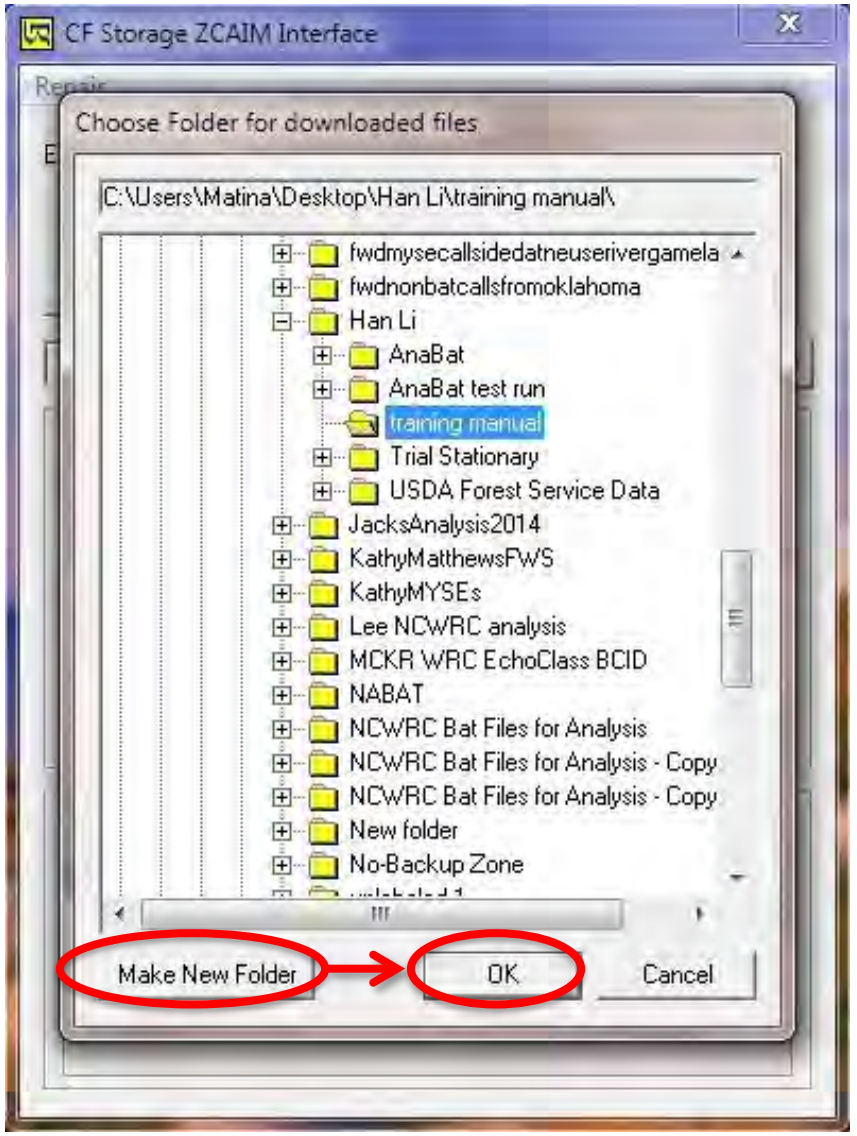
Download Data

- Default download options are used
- Click OK



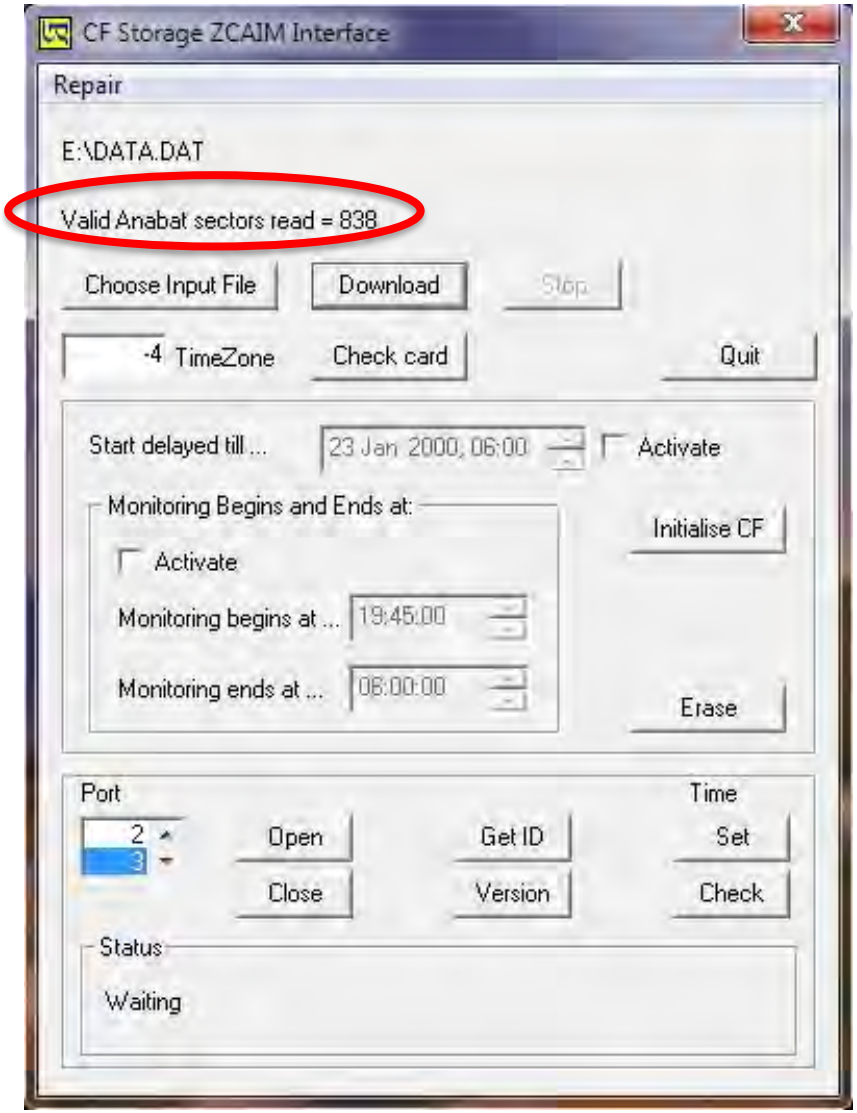
Download Data

- New window opens
- Choose where to store file
- Click Make New Folder
- Name new folder
- Click OK



Download Data

- Download is complete when Valid Anabat sectors read = ###



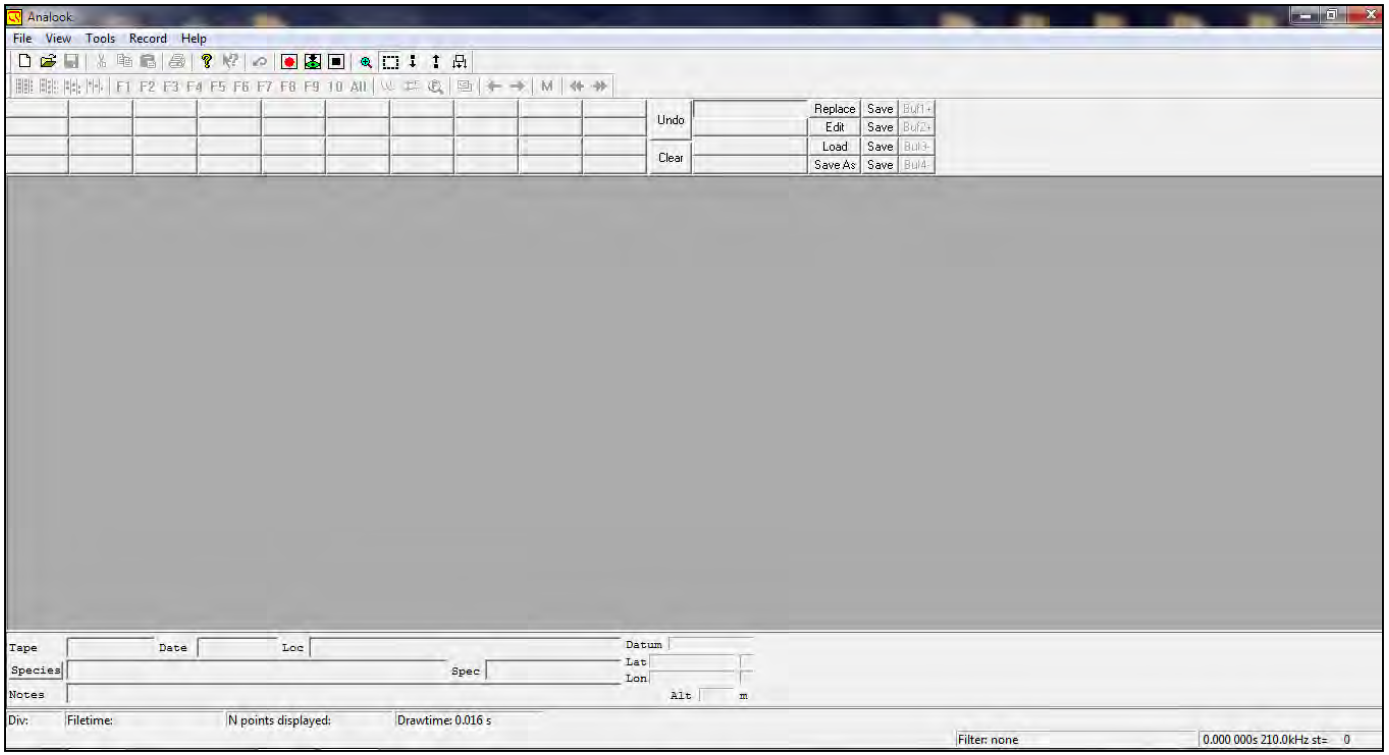
Looking at Recordings

Installing AnaLookW 4.1t

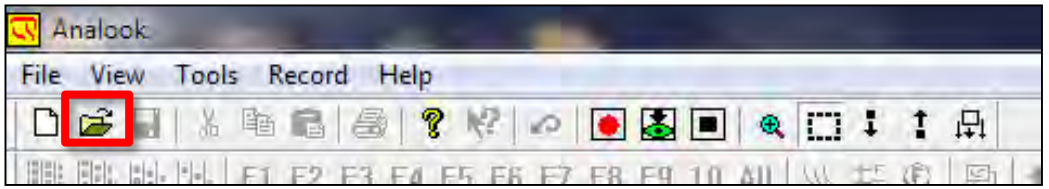
- AnaLook is used to look at recordings
- NEVER look at the original file that is on CF card (for data safety)
- Unzip the folder and open it

Using AnaLookW 4.1t

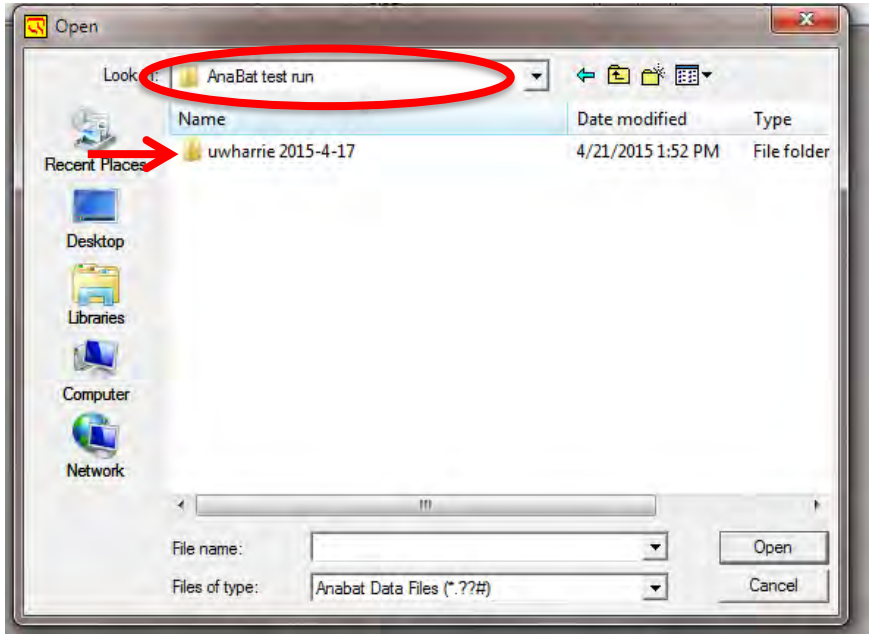
- How to look at your recordings, if you want to
- Open analookw.exe 



Using AnaLookW 4.1t

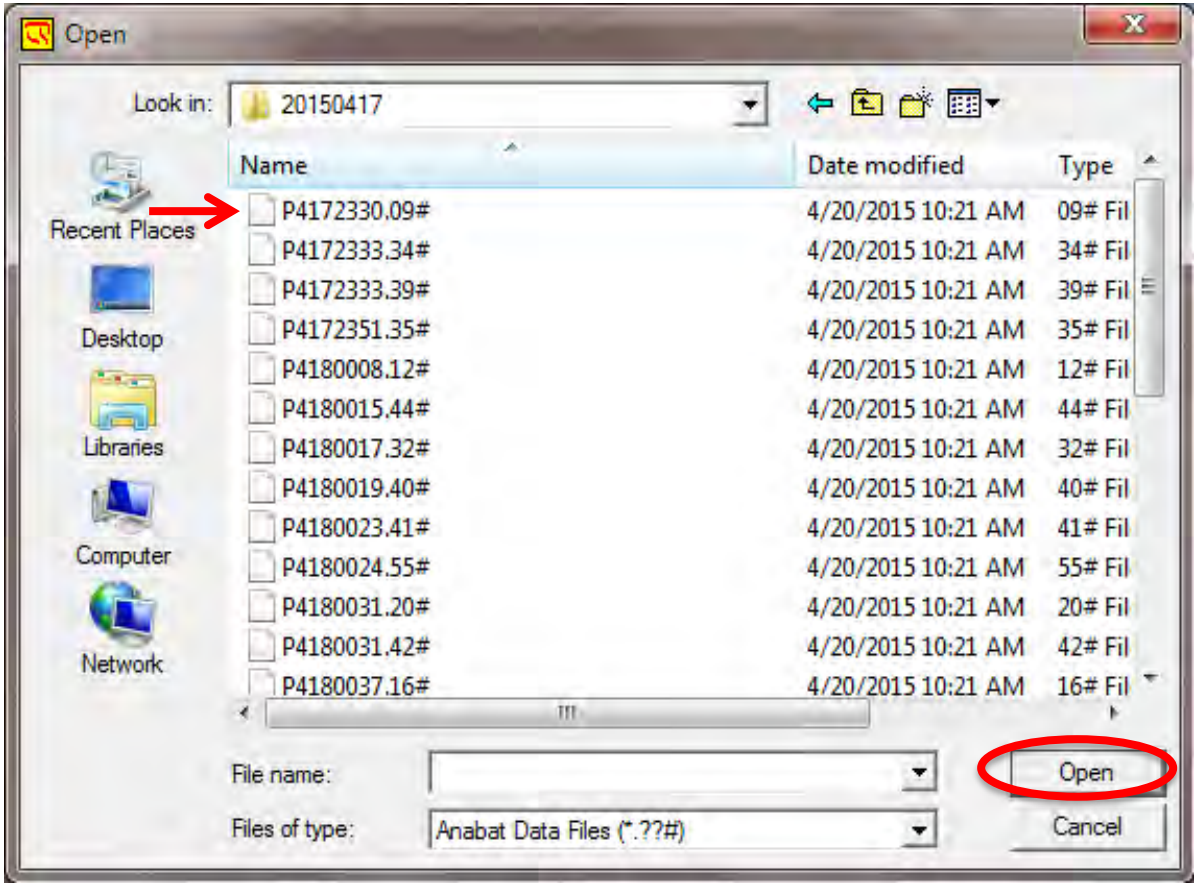


- Click open icon
- New window opens
- Click drop down menu Look in and navigate to where you saved the backed-up data
- Double click on folder



Using AnaLookW 4.1t

- Select a file
- Click Open



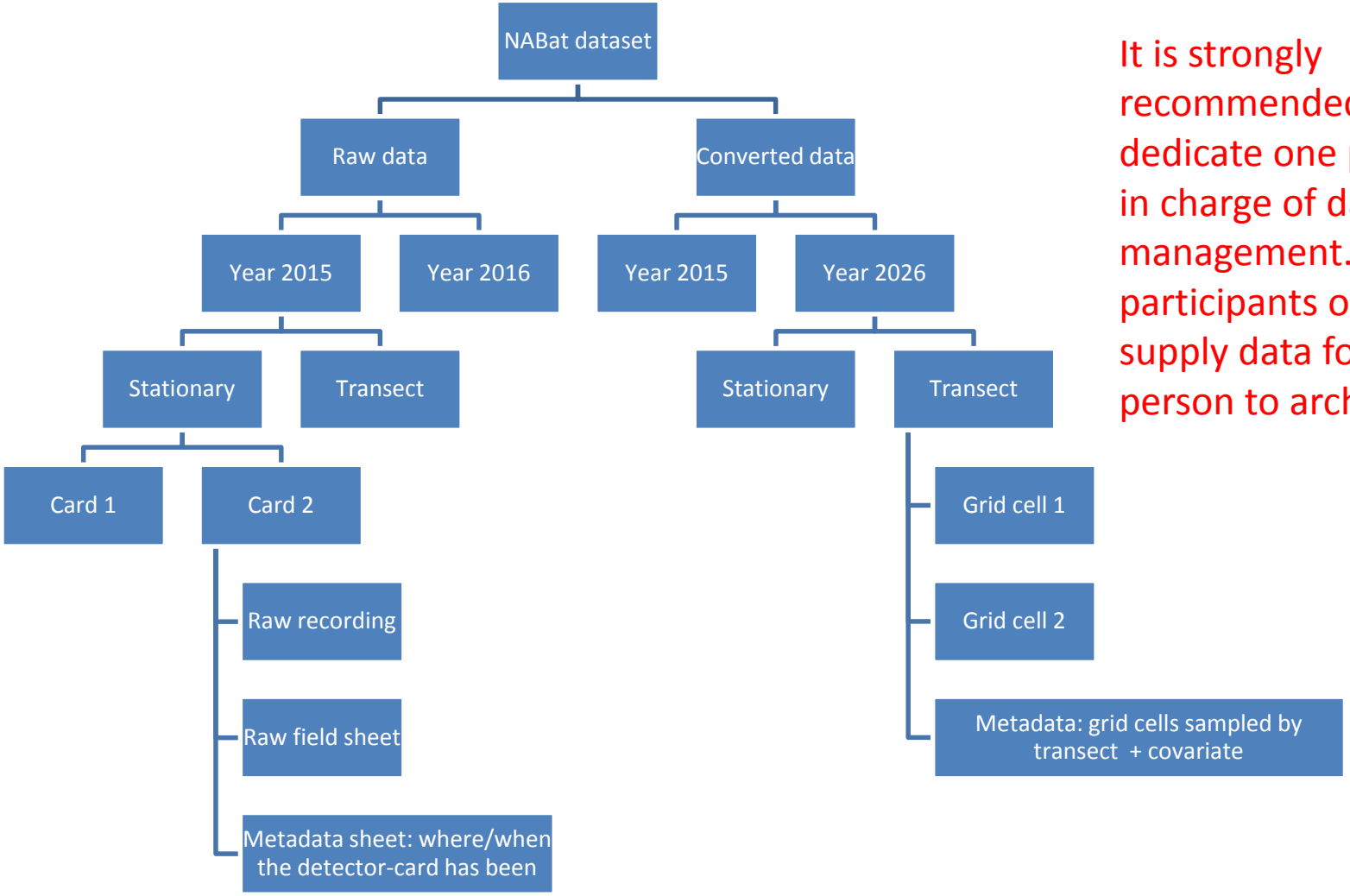
Data Management and Acoustic Analysis Protocols

North American Bat Monitoring Program
North Carolina Division

Han Li, PhD
University of North Carolina Greensboro

This protocol is written for the assumption of using AnaBat SD 2 from Titley Scientific. For information regarding to other brands or makes of bat detectors, please contact Dr. Han Li via h_li6@uncg.edu

Overall data storage structure

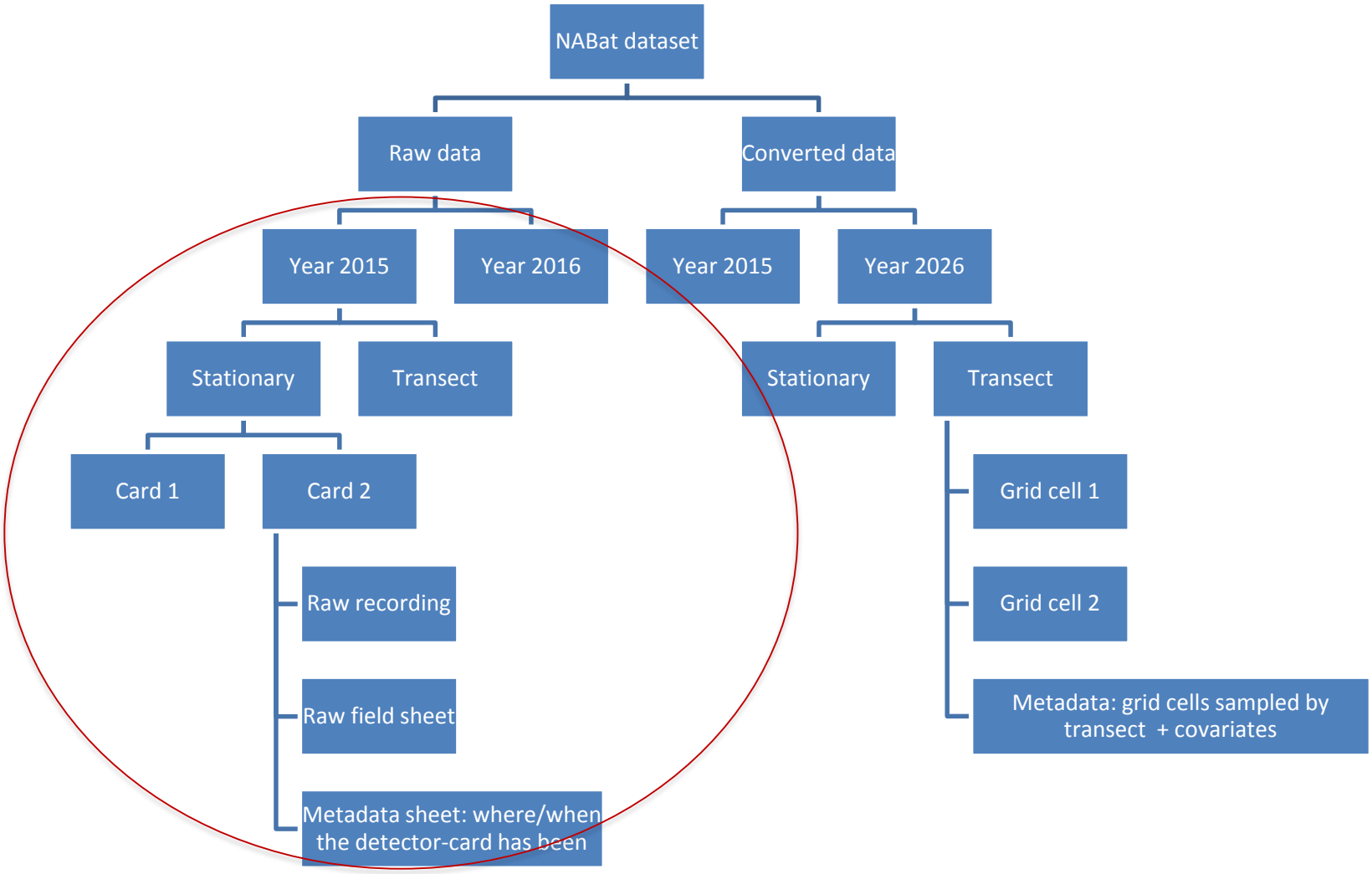


It is strongly recommended to dedicate one person in charge of data management. Other participants only supply data for the person to archive.

Step 1 Original data back up

- Back up all raw field data
  - Raw recording back up
    - AnaBat or other raw recordings need to be backed up before being converted into separated sound files.
    - See details in the NABat AnaBat training manual Page 41 – 42
  - Raw field notes back up
    - If possible scan the field datasheets or make copies

Overall data storage structure

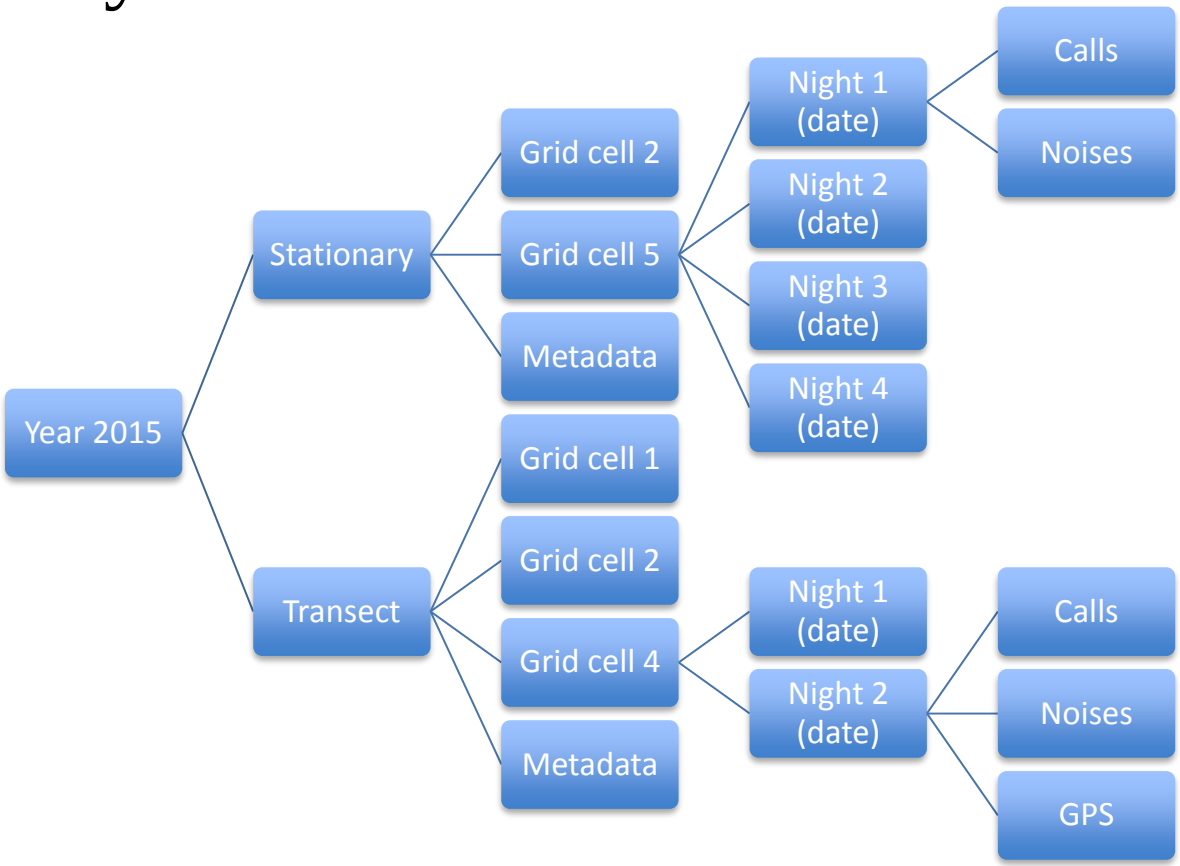


Notes on raw data storage

- Prevent conversion issues (time stamp, file type…)
- Raw data can be stored separately
- Do NOT erase the card during the season
  - No need to, card large enough for the whole season
- Only archive the very last version raw data backup after a season
- Meta data document where the detector-card pair has been over the season

Step 2 Downloading (converting) recordings

- Before downloading the recording, set up the hierarchy of destination folders



Step 2 (continue)

- Converting/downloading data in AnaBat
  - Using CFRead.exe (free, available at http://www.titley-scientific.com/us/index.php/software_firmware) to convert a DATA.DAT file sound files
  - Make sure it is the most updated version!
  - See details in the NABat AnaBat training manual Page 43 – 46
  - Do NOT convert as ZC files!

Pre-step 3 choose species ID goal

- Different goals of specie identification decide how to proceed with acoustic analysis
- Common goals/outcomes
  - Presence/no recording
  - Presence/no recording with statistical inference
  - Relative activity level/abundance

Pre-step 3 (continue)

- Presence/no recording
  - Can be summarized by night or by site (night combined)
  - No statistical inference (cannot the probability)
  - When summarized by night, good for occupancy model, distribution mapping, or other further statistic analysis
  - Basic steps
    - Automated identification with multiple programs (step 4, skip step 3 in this protocol)
    - Search for program id concordance (step 5)
    - Manually verify a few files per species (step 6)

Pre-step 3 (continue)

- Presence/no recording with statistical inference
  - Usually MLE (maximum likelihood estimate) is reported
  - How to properly report reliable automated identification results of one particular program
  - Use only one automated identification program
  - Basic steps
    - Screen for good calls only (step 3)
    - Auto-id with one program and collect MLE (step 4)
    - Manually verify selected species (usually myotis or other species of concern) (step 6)

Pre-step 3 (continue)

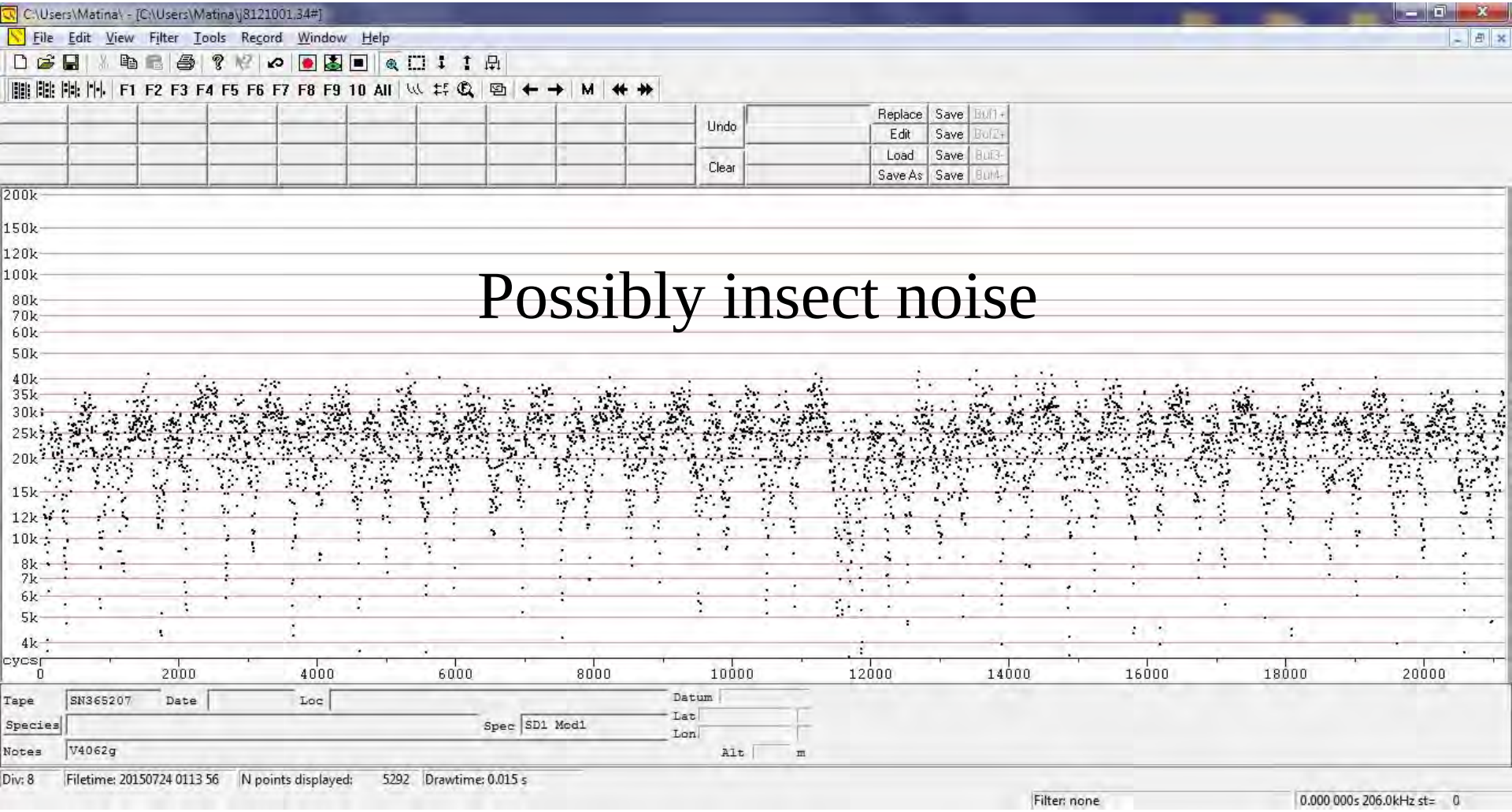
- Relative activity level/abundance
  - All files need to be identified
  - Completely manual id
    - Suitable for small dataset (thousands of calls)
    - Construct a reliable reference library
      - Step 4 and step 5 with multiple programs
      - Use publications or other known call files
  - Set up a threshold in auto id results at file level
    - BCID – discriminant probability
    - Echoclass/Kaleidoscope – agreement percentage

Step 3 Screen for analyzable calls

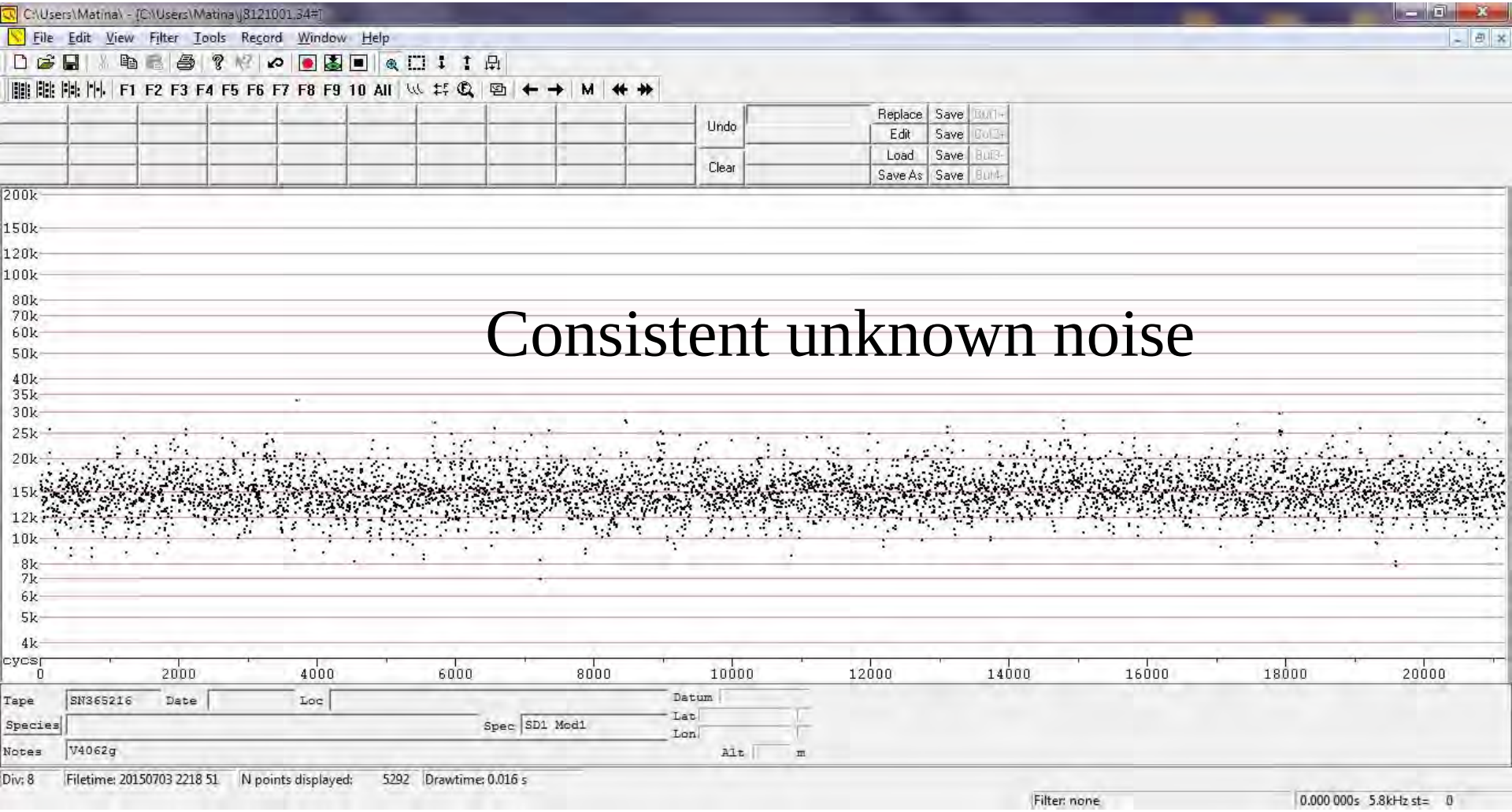
- Certain recordings might not include any bat echolocation pulses or might only include low quality pulses. These “bad” recordings will cause issues when using an automated identification program. Thus they need to be manually separated. This can be done via Analook (free, available at http://www.titley-scientific.com/us/index.php/software_firmware)
- See details in the NABat AnaBat training manual Page 1 – 9 on how to obtain Analook

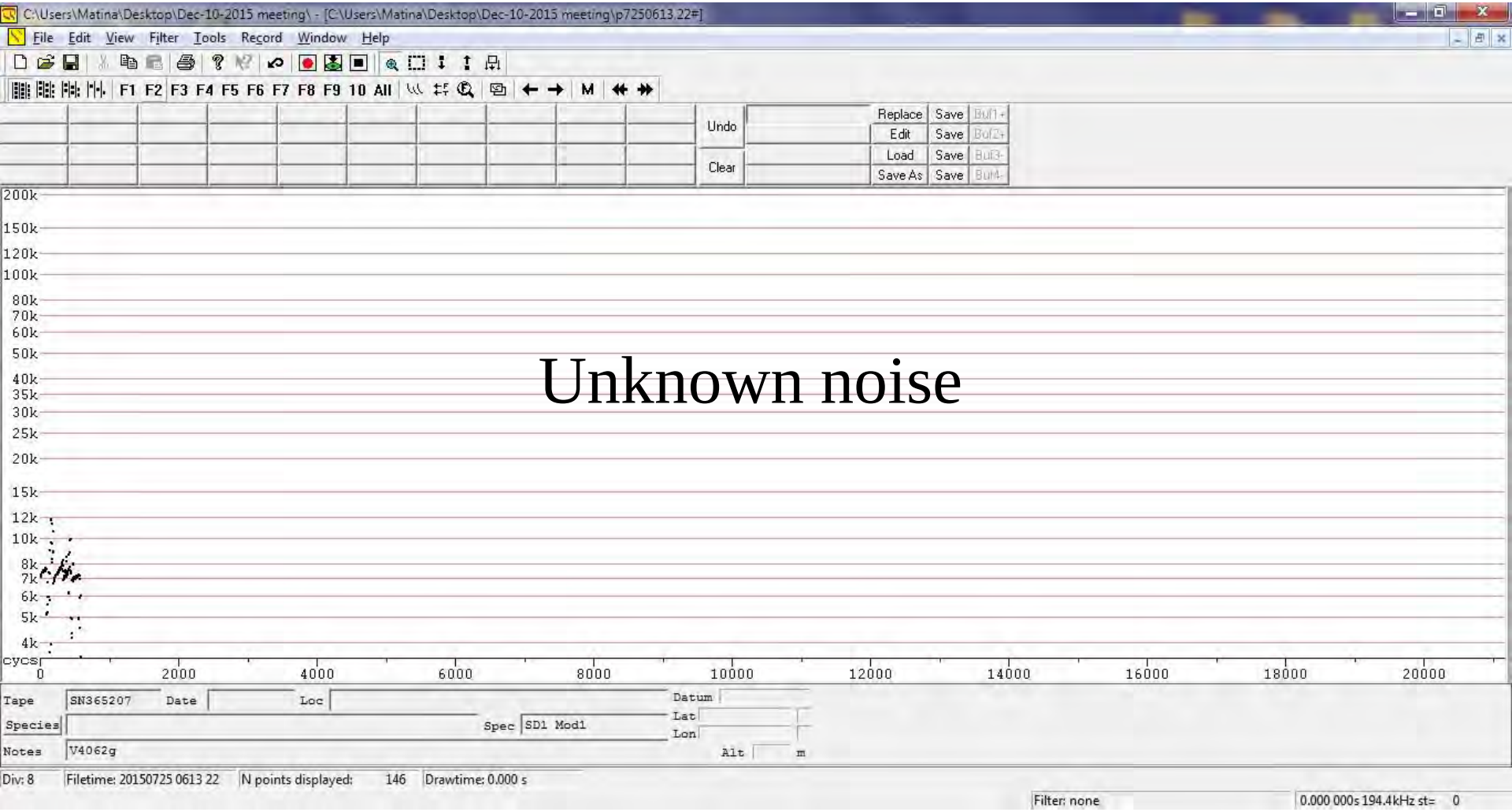
Step 3 (continue)

- Using AnaLook to manually separate sound files that include these, the following pages give examples of each type of undesirable files:
  - Noises with no bat call
  - Low quality calls
    - Low number of pulses (usually less than 3 pulses)
    - Fragmentary calls
    - Noise mixed with calls
    - Feeding calls/social calls
    - Multiple bats

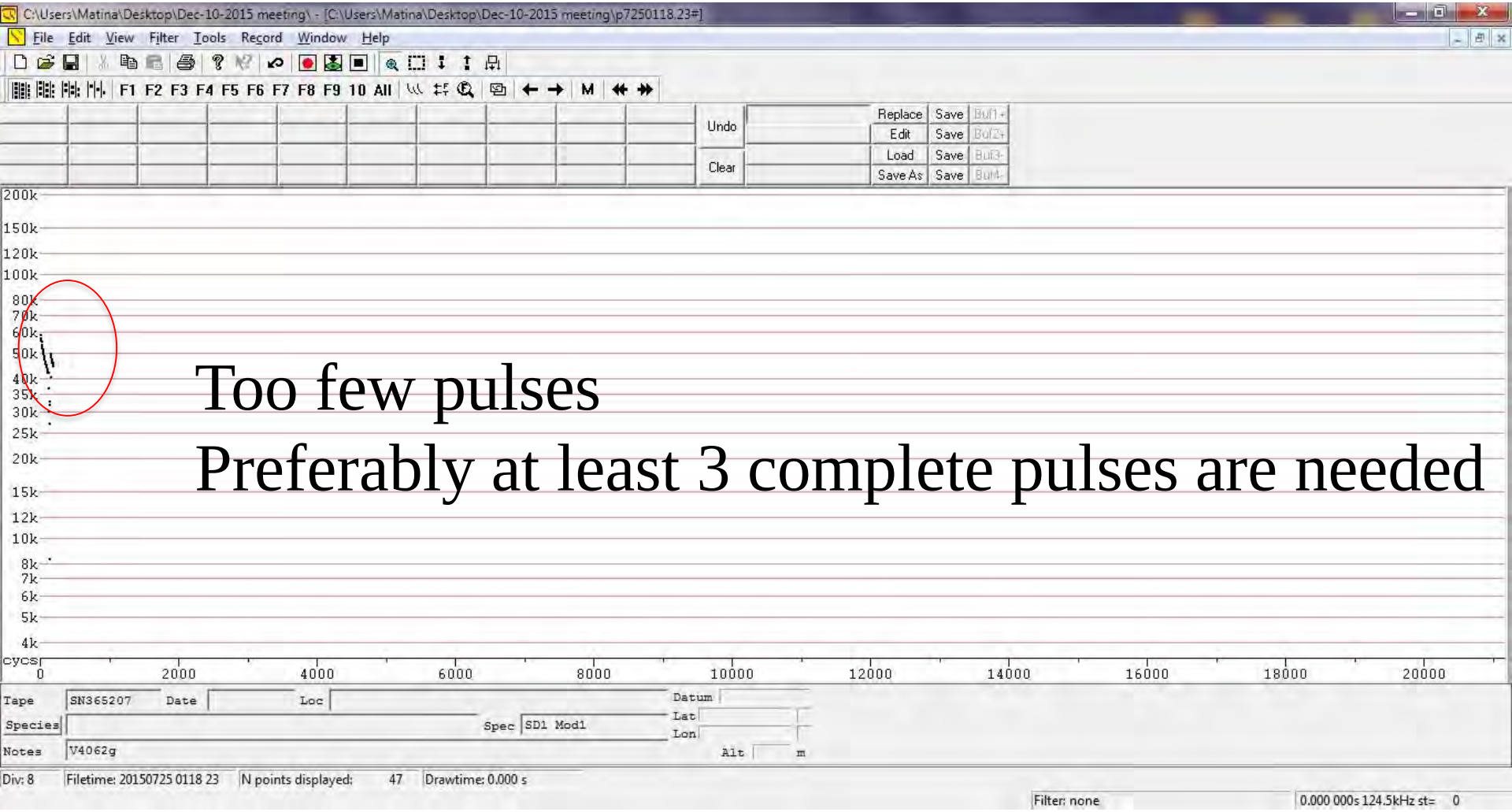


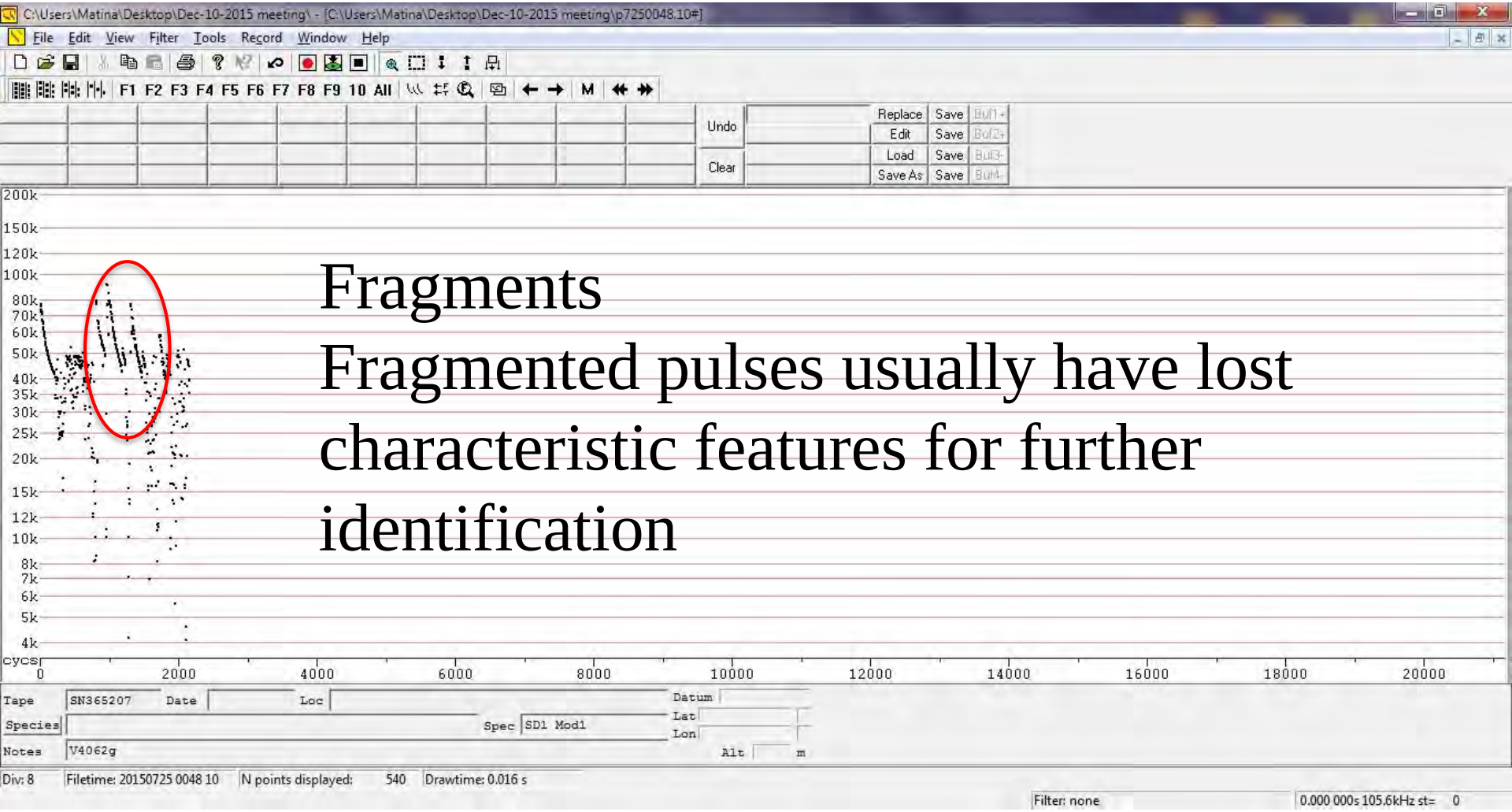
Possibly insect noise


C:\Users\Matina\ - [C:\Users\Matina\j8121001.34#]
File Edit View Filter Tools Record Window Help
F1 F2 F3 F4 F5 F6 F7 F8 F9 10 All M
Undo
Clear
Replace Save
Edit Save
Load Save
Save As Save
Consistent unknown noise
200k 150k 120k 100k 80k 70k 60k 50k 40k 35k 30k 25k 20k 15k 12k 10k 8k 7k 6k 5k 4k
cycs
0 2000 4000 6000 8000 10000 12000 14000 16000 18000 20000
Tape SN365216 Date Loc Datum
Species Spec SD1 Mod1 Lat Lon
Notes V4062g Alt m
Div: 8 Filetime: 20150703 2218 51 N points displayed: 5292 Drawtime: 0.016 s
Filter: none 0.000 000s 5.8kHz st= 0



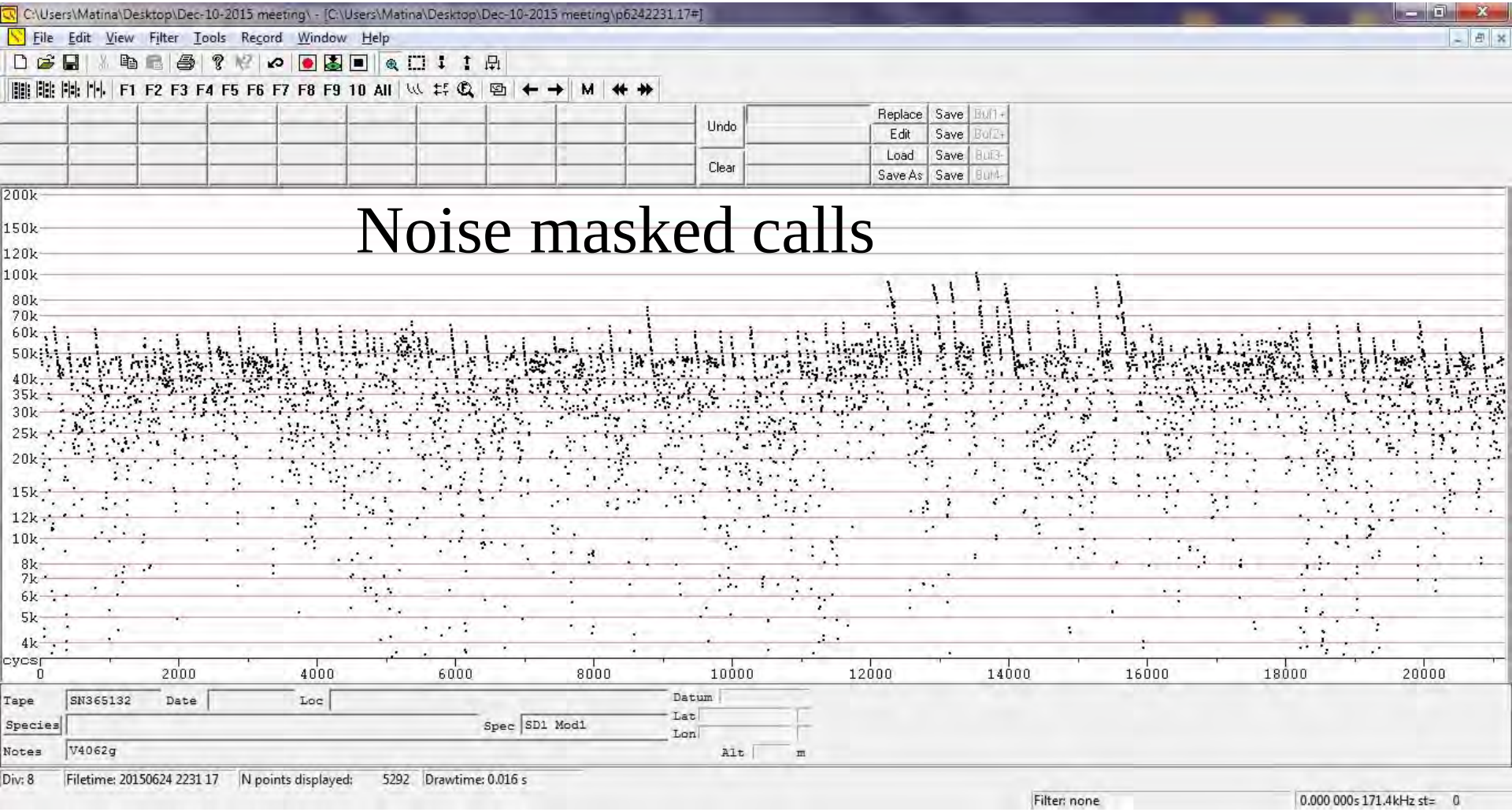
Unknown noise


C:\Users\Matina\Desktop\Dec-10-2015 meeting\ - [C:\Users\Matina\Desktop\Dec-10-2015 meeting\p7250118.23#]
File Edit View Filter Tools Record Window Help
F1 F2 F3 F4 F5 F6 F7 F8 F9 10 All M
Undo
Clear
Replace Save
Edit Save
Load Save
Save As Save
200k
150k
120k
100k
80k
70k
60k
50k
40k
35k
30k
25k
20k
15k
12k
10k
8k
7k
6k
5k
4k
cycs
0
2000
4000
6000
8000
10000
12000
14000
16000
18000
20000
Too few pulses
Preferably at least 3 complete pulses are needed
Tape SN365207 Date Loc Datum
Species Spec SD1 Mod1 Lat
Notes V4062g Lon
Alt m
Div: 8 Filetime: 20150725 0118 23 N points displayed: 47 Drawtime: 0.000 s
Filter: none
0.000 000s 124.5kHz st= 0

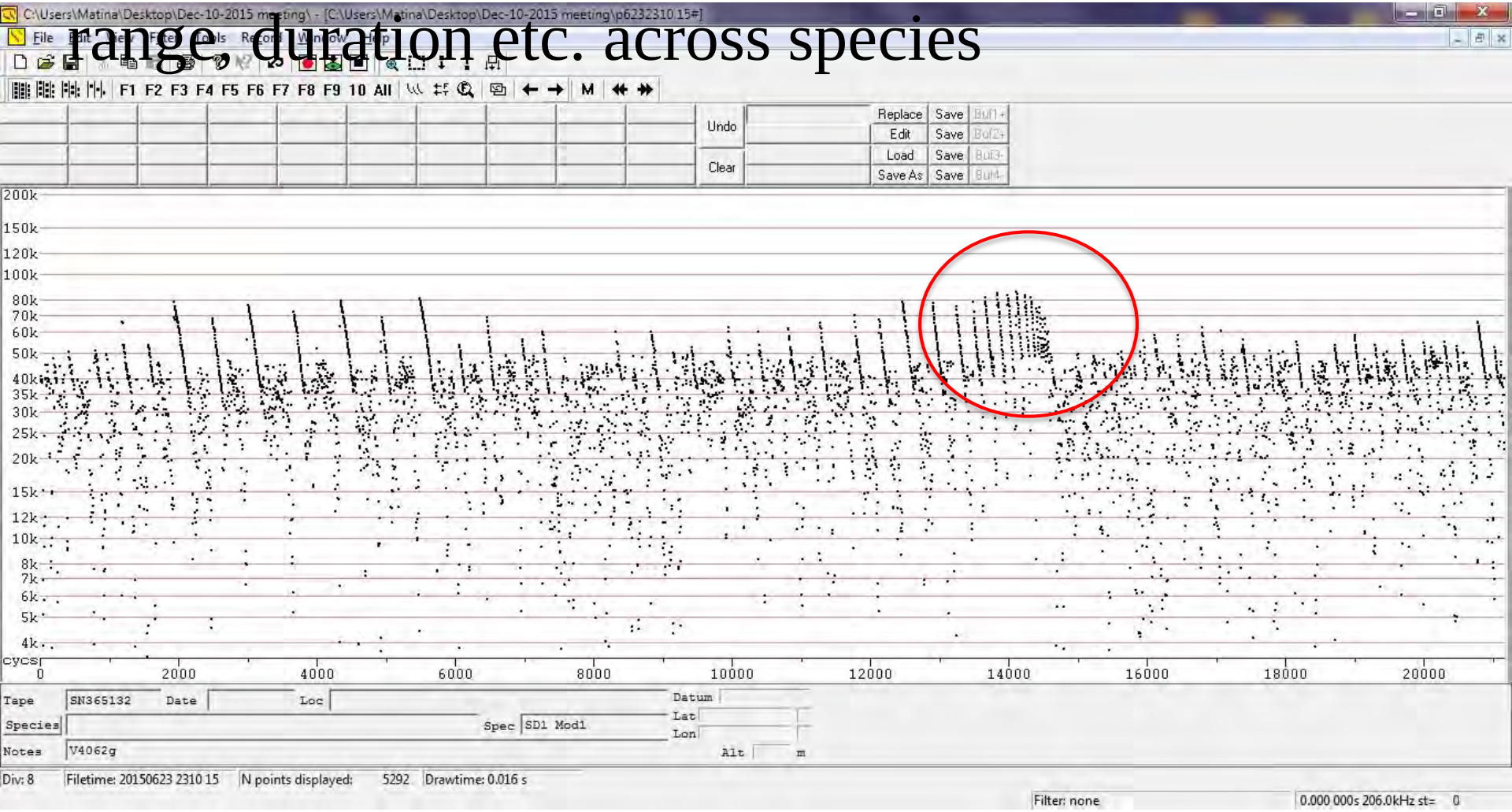


Fragments

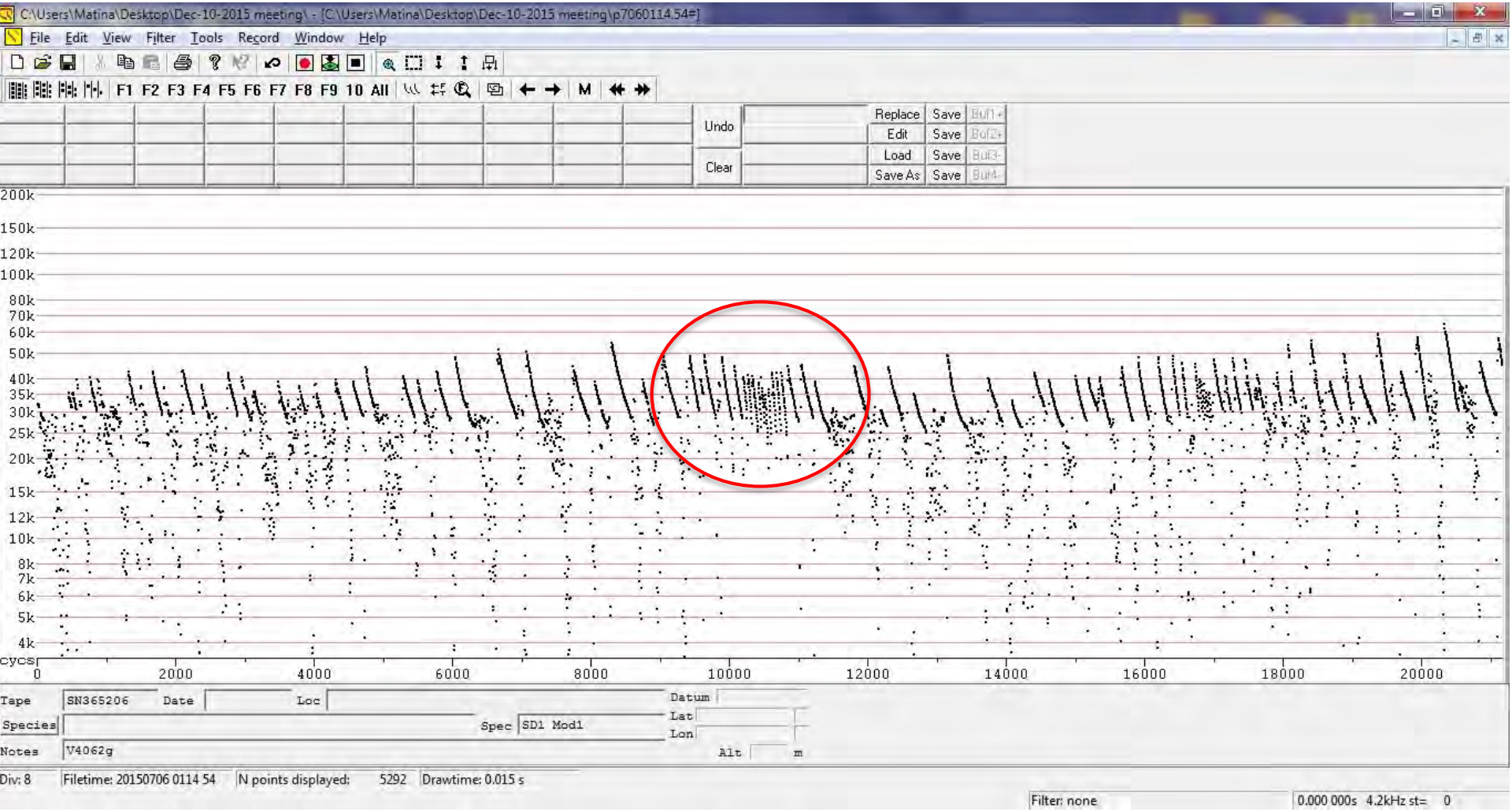
Fragmented pulses usually have lost characteristic features for further identification

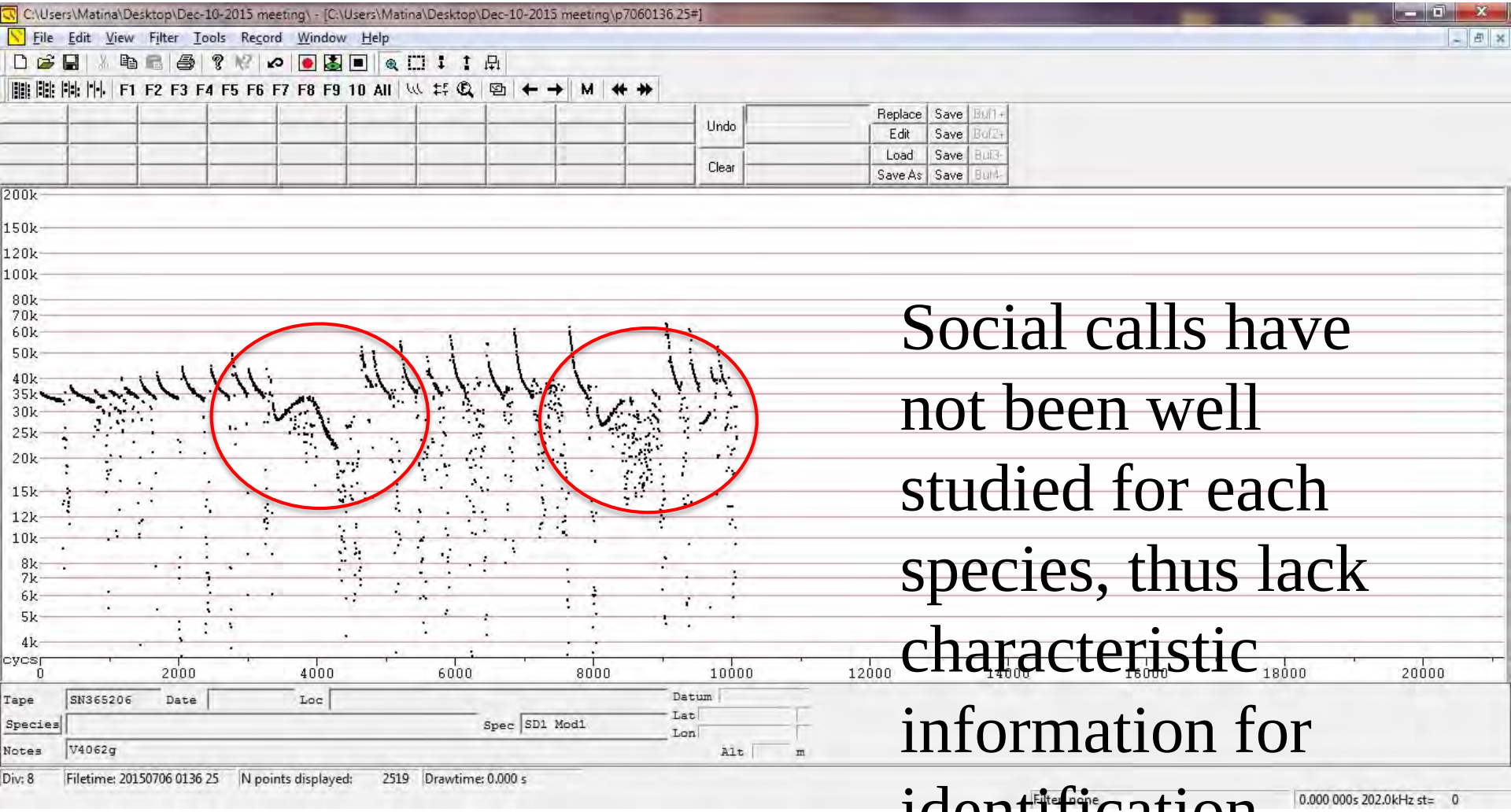

C:\Users\Matina\Desktop\Dec-10-2015 meeting\ - [C:\Users\Matina\Desktop\Dec-10-2015 meeting\p6242231.17#]
File Edit View Filter Tools Record Window Help
F1 F2 F3 F4 F5 F6 F7 F8 F9 10 All M
Undo
Clear
Replace Save
Edit Save
Load Save
Save As Save
Noise masked calls
200k 150k 120k 100k 80k 70k 60k 50k 40k 35k 30k 25k 20k 15k 12k 10k 8k 7k 6k 5k 4k
cycs
0 2000 4000 6000 8000 10000 12000 14000 16000 18000 20000
Tape SN365132 Date Loc Datum
Species Spec SD1 Mod1 Lat Lon
Notes V4062g Alt m
Div: 8 Filetime: 20150624 2231 17 N points displayed: 5292 Drawtime: 0.016 s
Filter: none
0.000 000s 171.4kHz st= 0

Feeding calls usually have similar frequency range, duration etc. across species

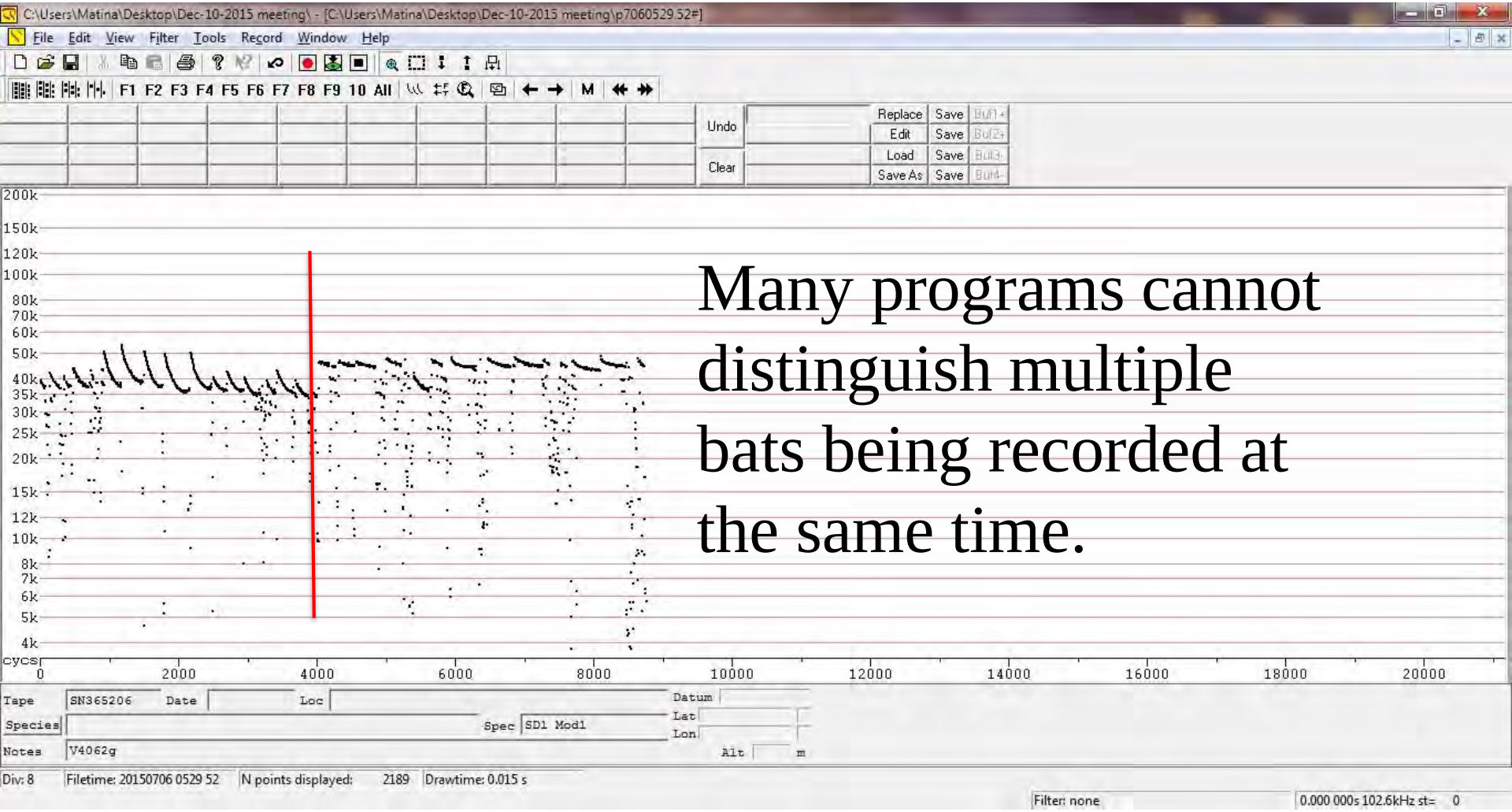


Feeding calls



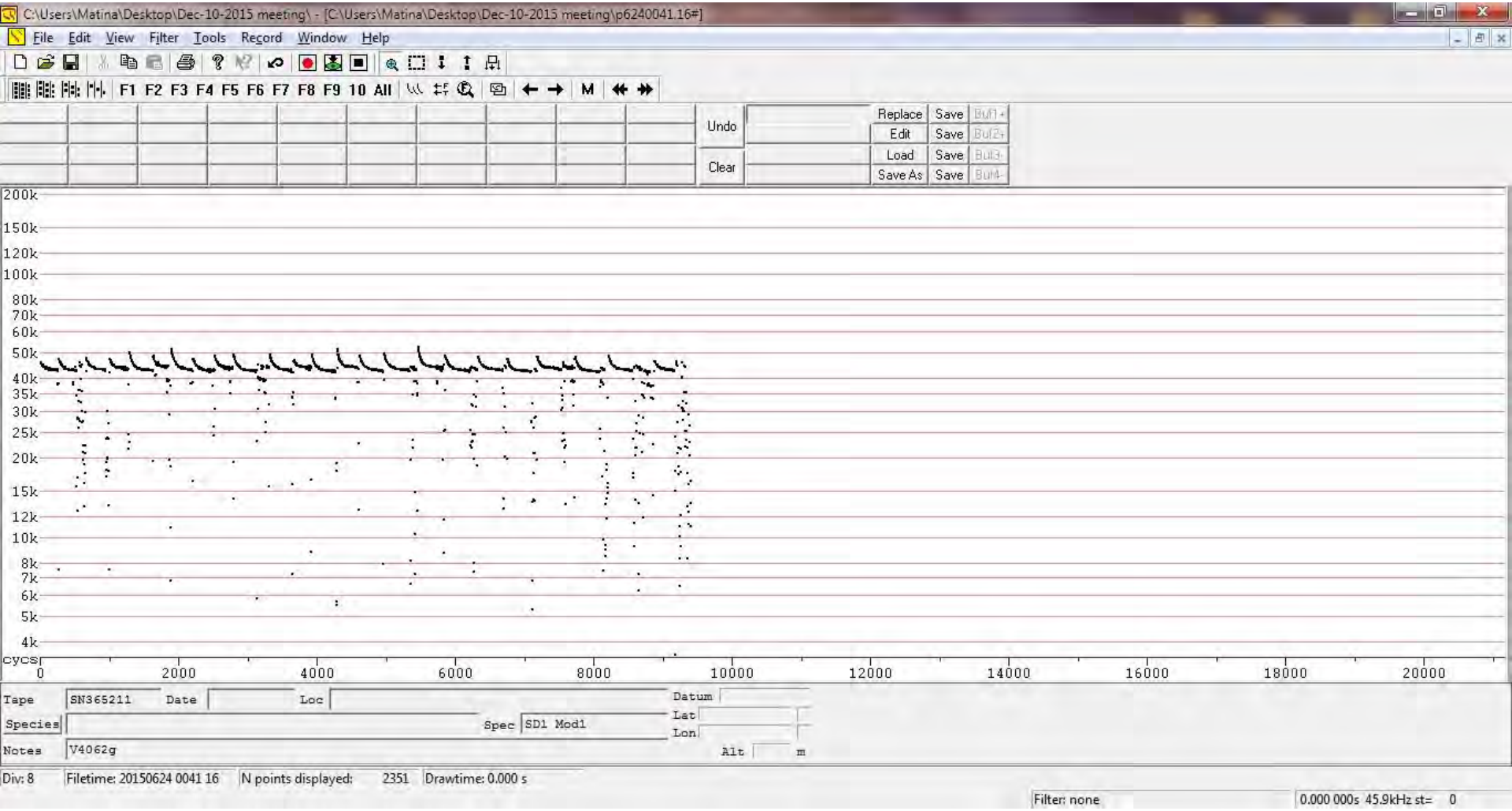


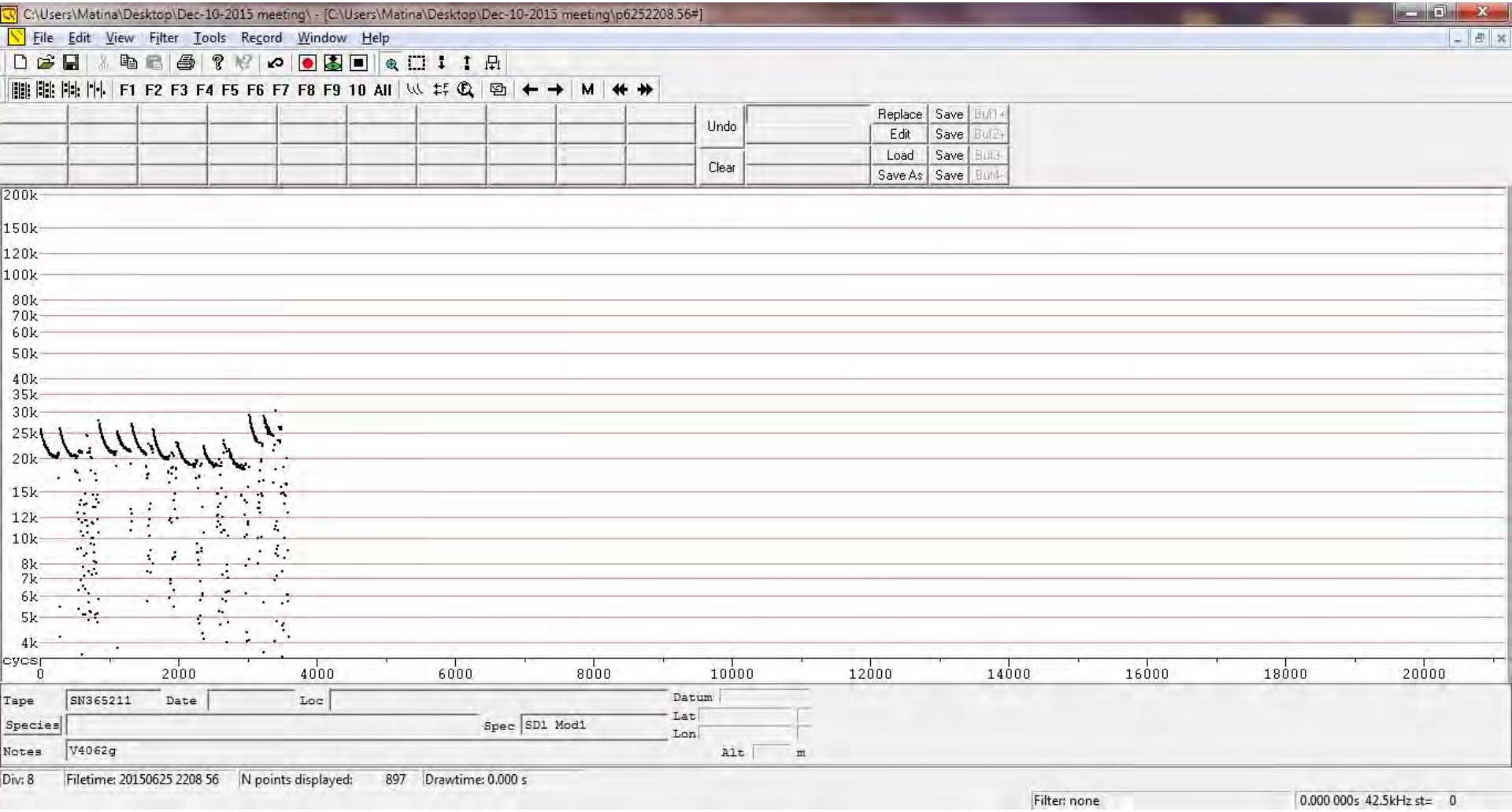
Social calls have not been well studied for each species, thus lack characteristic information for identification



Many programs cannot distinguish multiple bats being recorded at the same time.

The next two pages show good quality calls that are suitable for automated identification




C:\Users\Matina\Desktop\Dec-10-2015 meeting\ - [C:\Users\Matina\Desktop\Dec-10-2015 meeting\p6252208.56#]
File Edit View Filter Tools Record Window Help
F1 F2 F3 F4 F5 F6 F7 F8 F9 10 All M
Undo
Clear
Replace Save
Edit Save
Load Save
Save As Save
200k 150k 120k 100k 80k 70k 60k 50k 40k 35k 30k 25k 20k 15k 12k 10k 8k 7k 6k 5k 4k
cycs
0 2000 4000 6000 8000 10000 12000 14000 16000 18000 20000
Tape SN365211 Date Loc Datum
Species Spec SD1 Mod1 Lat Lon
Notes V4062g Alt m
Div: 8 Filetime: 20150625 2208 56 N points displayed: 897 Drawtime: 0.000 s
Filter: none 0.000 000s 42.5kHz st= 0

Step 3 (continue)

- It is recommended to have three folders that separate good quality calls, low quality calls, and noise files with no call.
- Low quality calls can still be manually identified via trained experts or counted towards the total bat activity within a sampling night.

Step 4 Automated identification

- There are a few automated identification programs available. In this protocol, we only demonstrate two:
  - EchoClass
  - BCID
- It is strongly recommended to consult with software manuals or contact developers directly for specific questions.

Step 4 (continue)

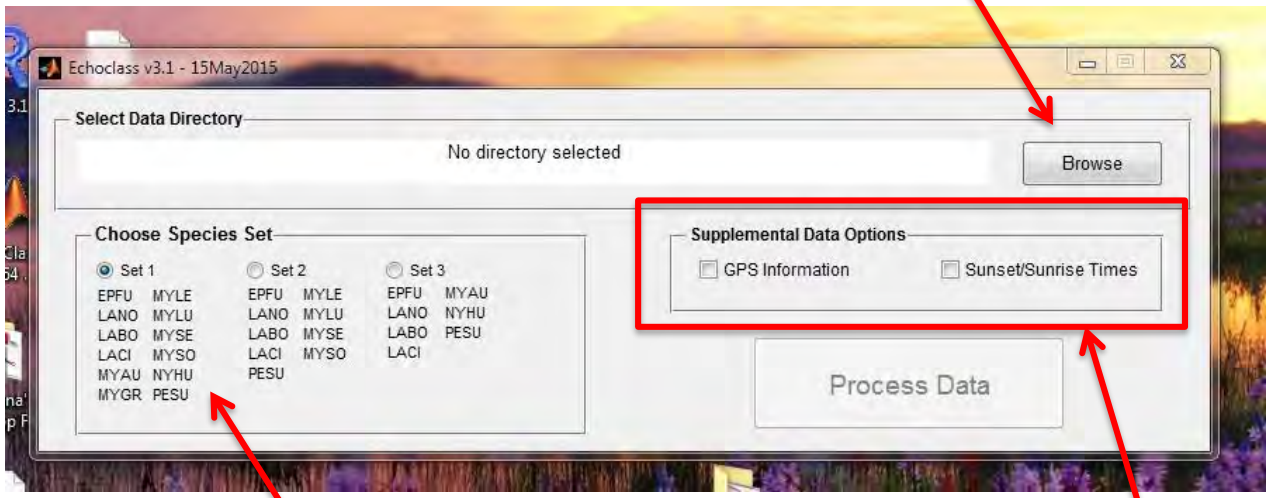
- EchoClass
  - Developed by Dr. Eric Britzke, U.S. Army Engineer Research and Development Center, Vicksburg, Mississippi
  - Free, downloadable from http://www.fws.gov/midwest/endangered/mammals/inba/surveys/inbaAcousticSoftware.html

Step 4 (continue)

- EchoClass
  - Software user instruction is available at: http://www.fws.gov/midwest/endangered/mammals/inba/surveys/pdf/EchoclassV3Instructions.pdf
  - Once properly installed, a desktop shortcut should be available
  - Will NOT recognize ZC files. Must be number files

Once open the program the interface below will show up

Click here to choose folder to analyze

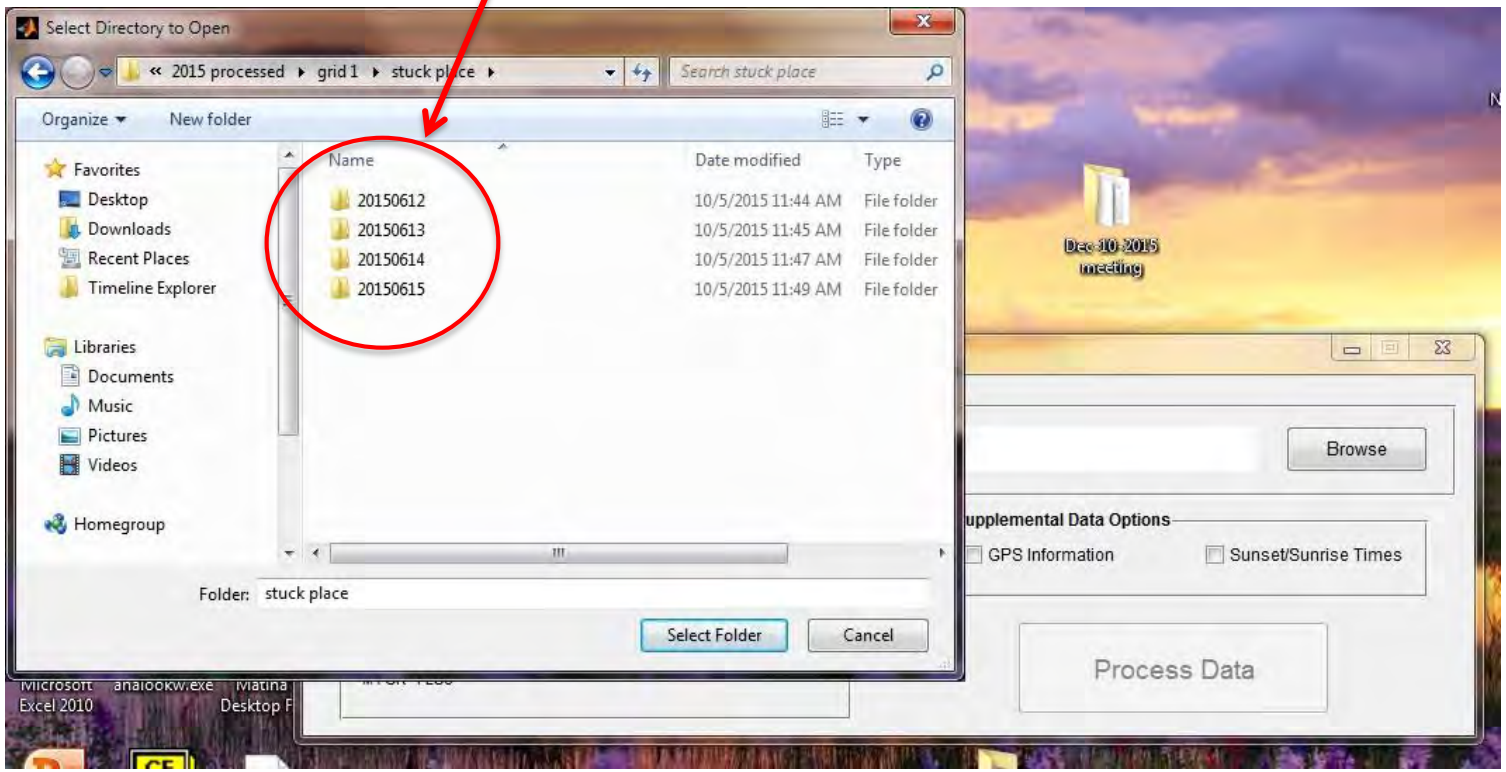


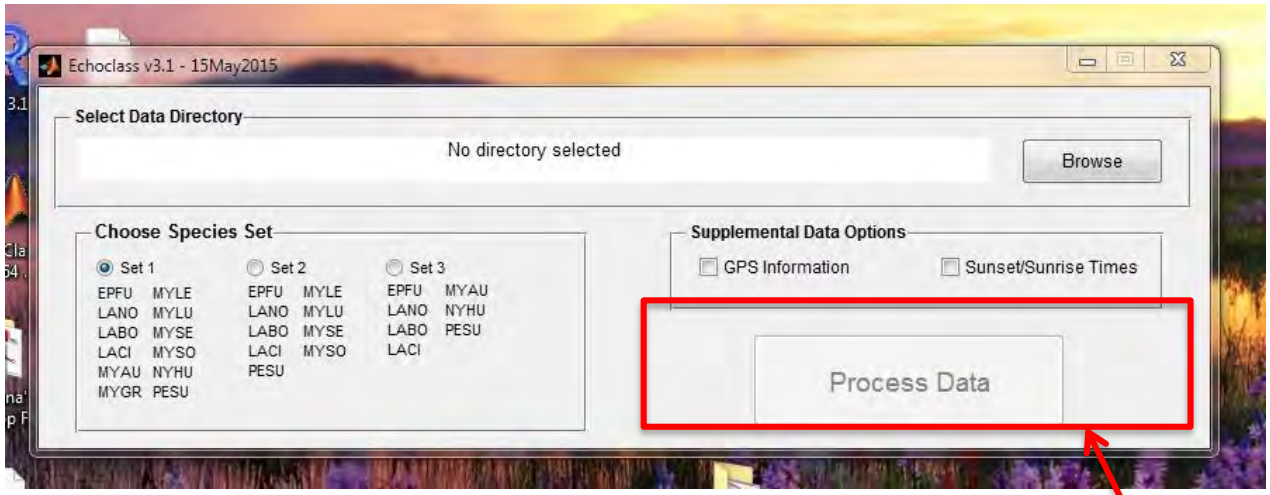
Choose candidate species

Set 1 is recommended for NC

Combine optional data

EchoClass requires a unique way to name folders
Note the name of the LAST level of folders:
It has to be the 8-digit date



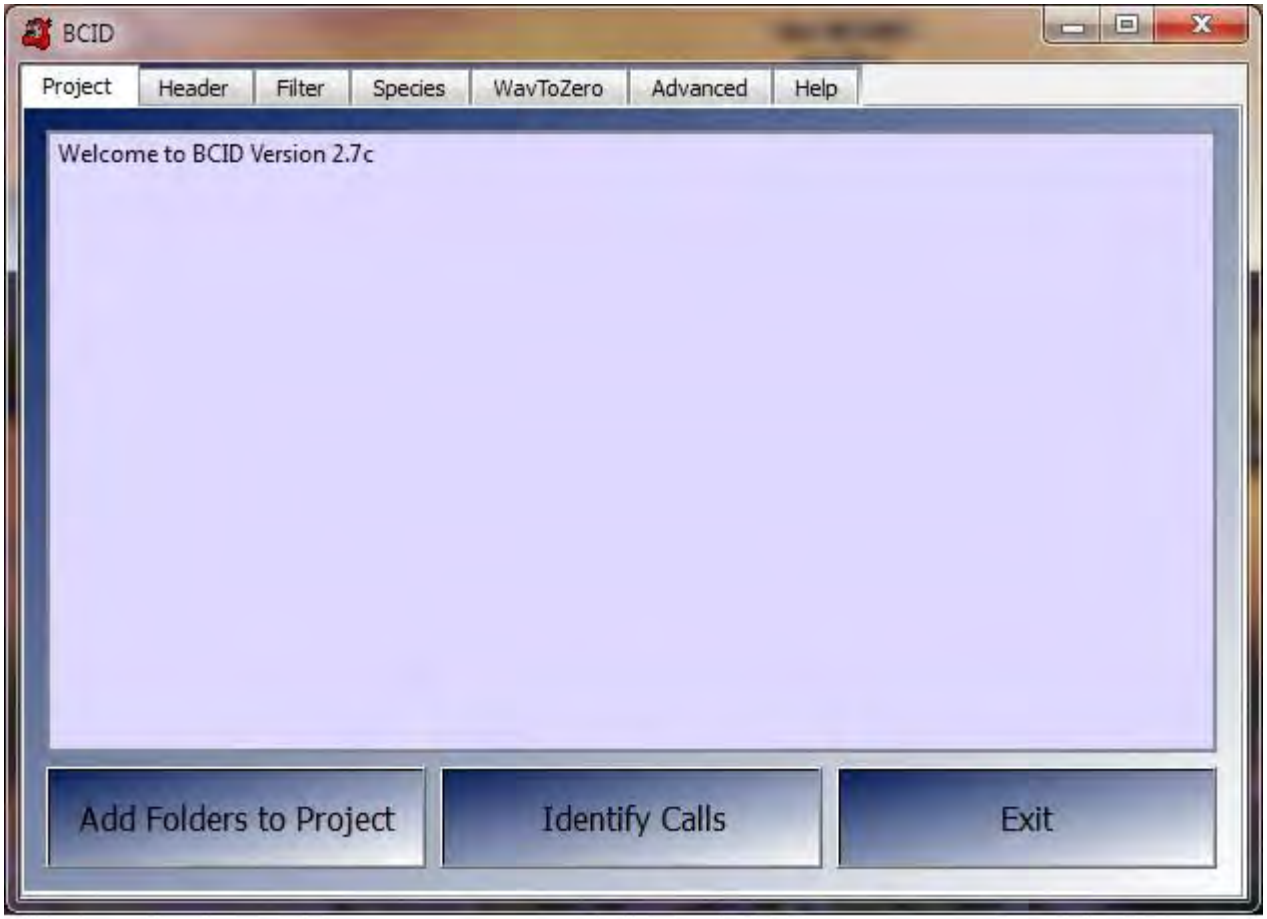


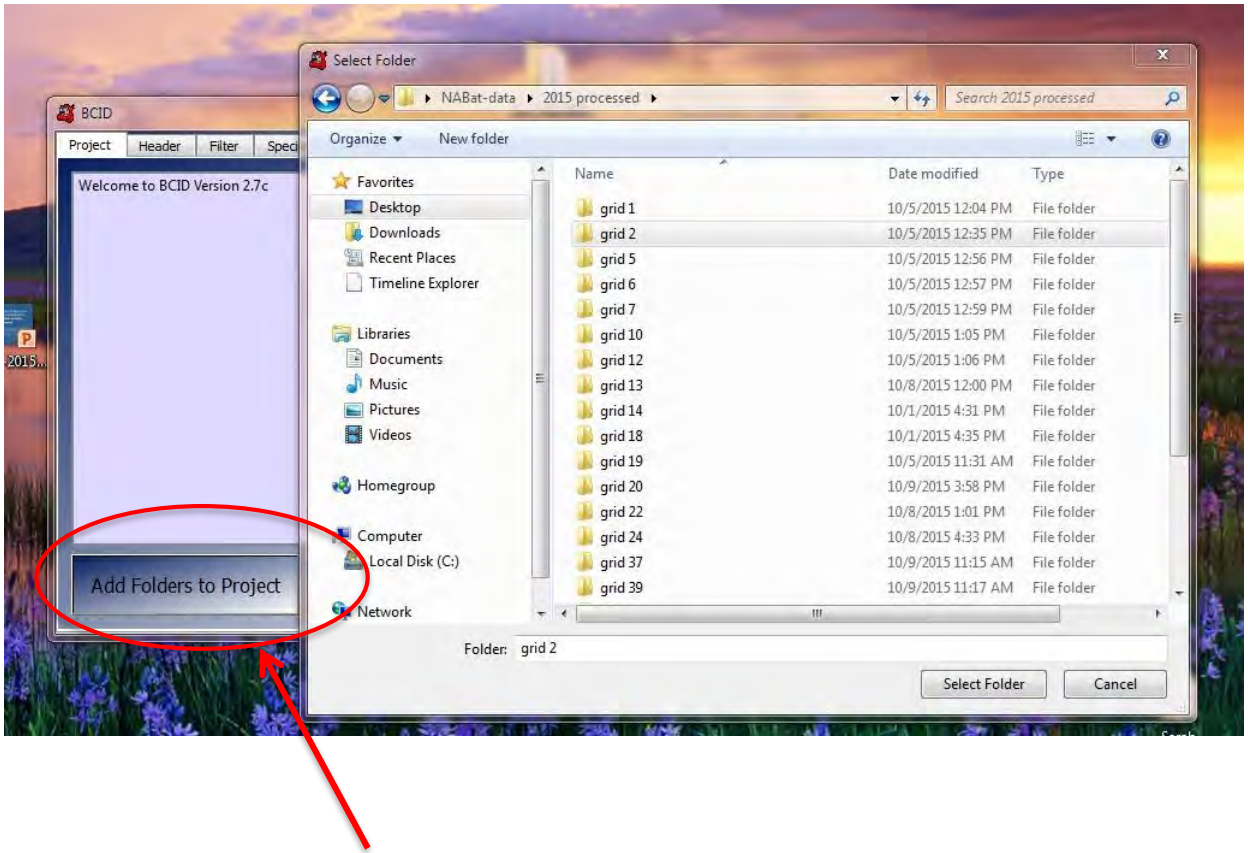
Click here to run the analysis EchoClass is relatively computing power and time consuming.

Step 4 (continue)

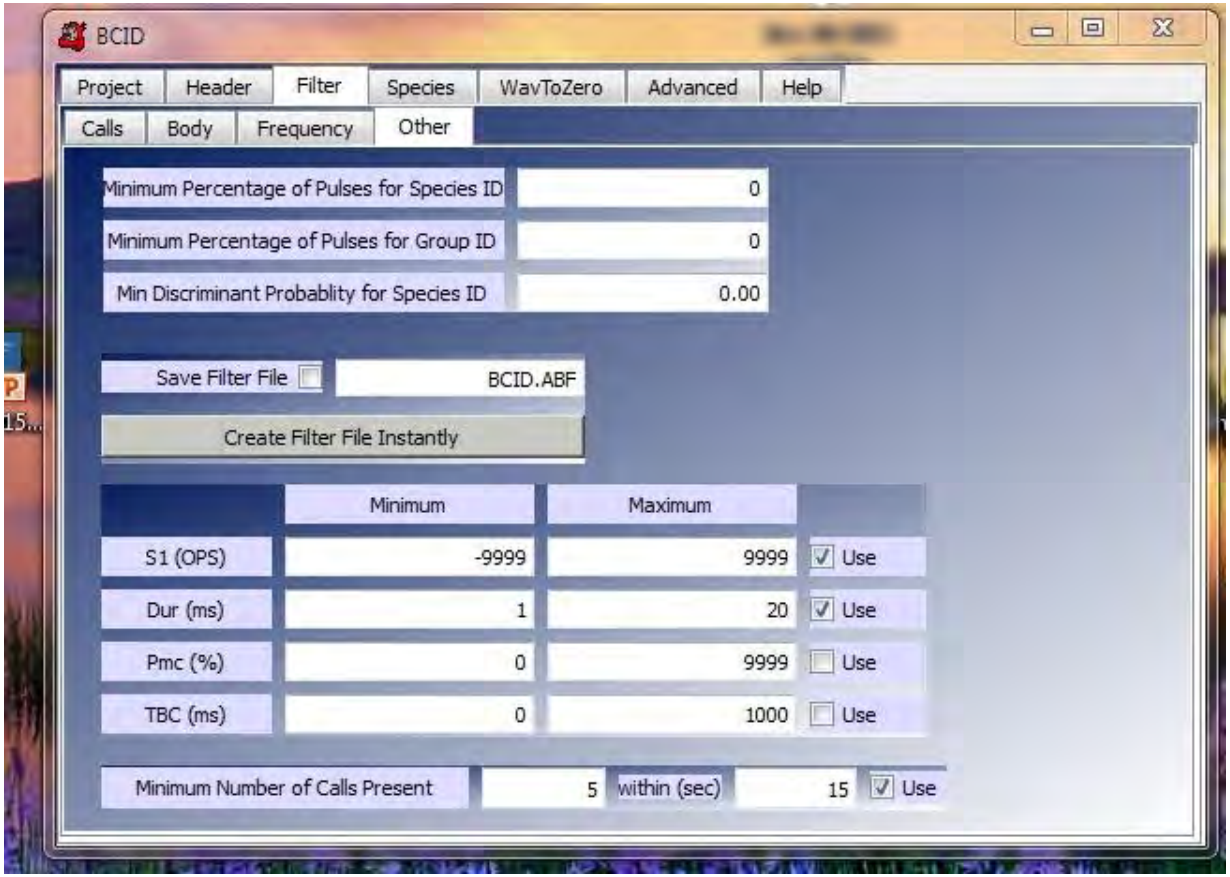
- BCID
  - Purchase required via http://www.batcallid.com/allsoftware.html
  - Once properly installed, a desktop shortcut should be available
  - A user manual is included once the program has been purchased and installed (under “Help” tab).

Program interface



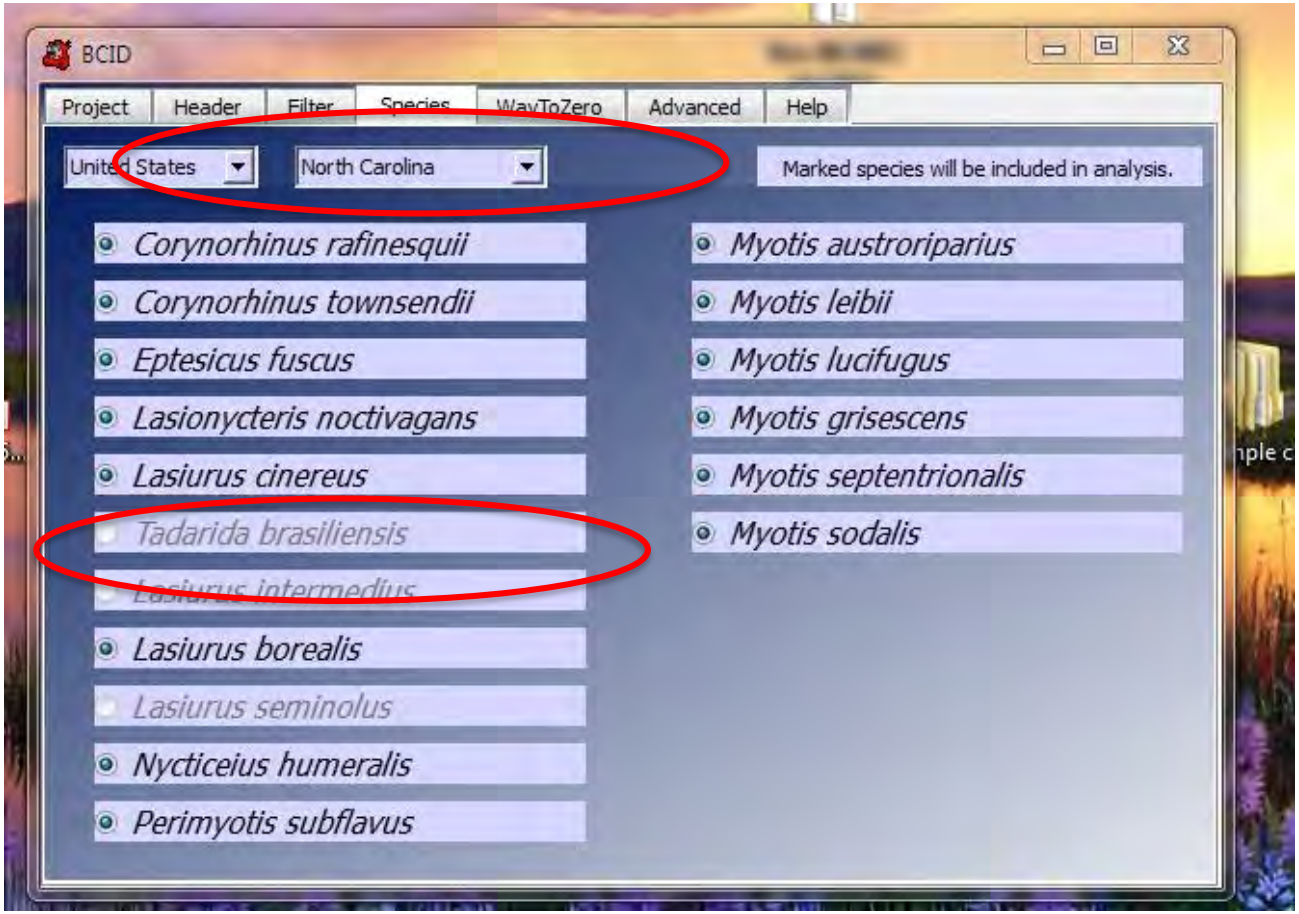


Choose the folder to analyze



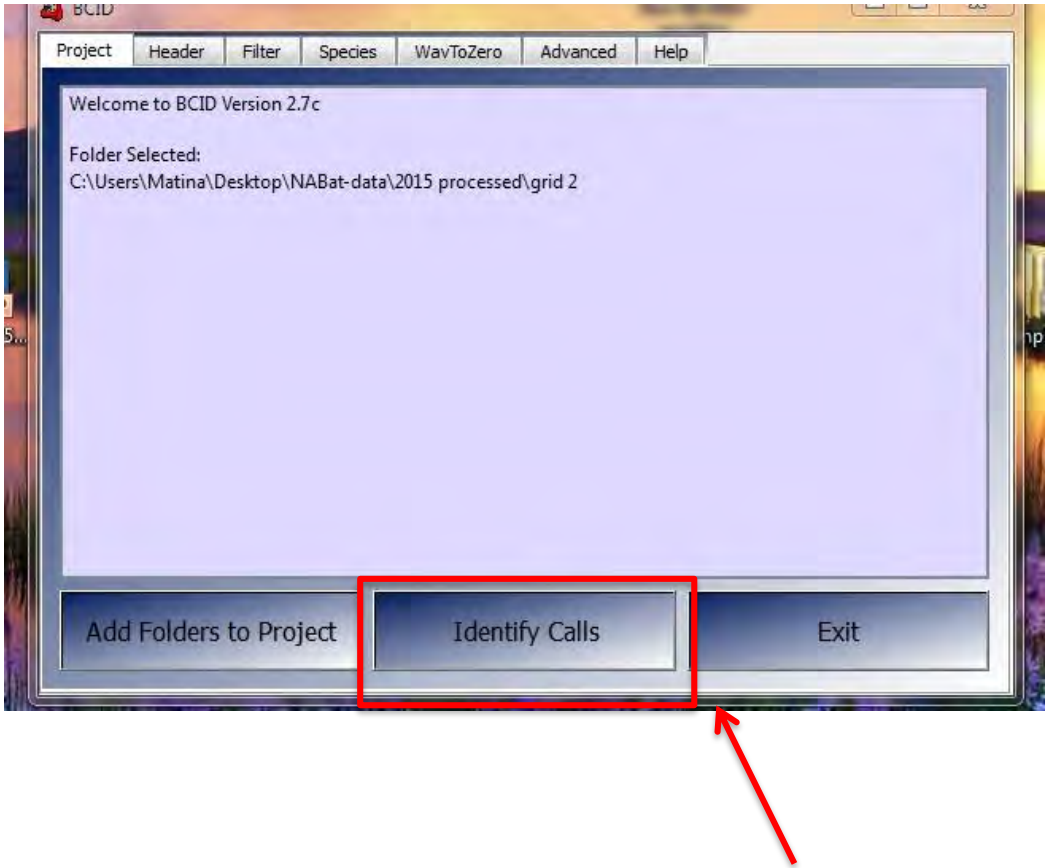
Choose the filters.

If no special reason, use the default filters



Choose candidate species

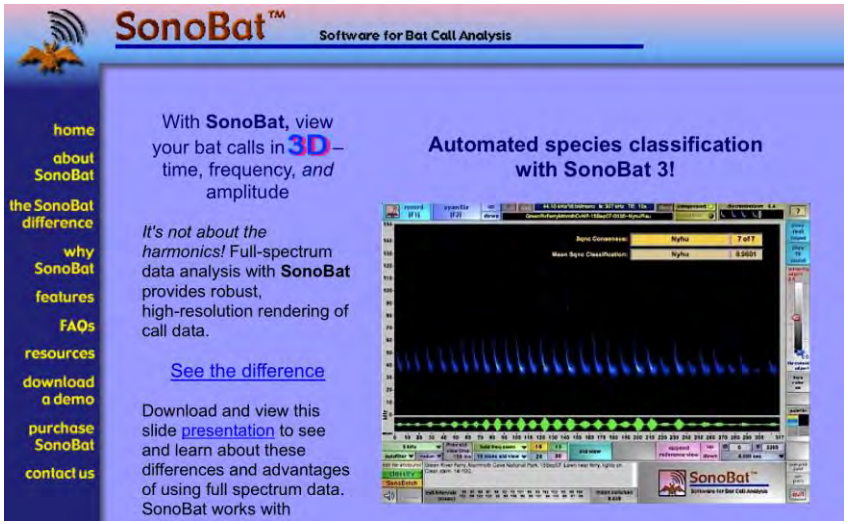
Note certain species are unavailable



Run the analysis

Step 4 (continue)

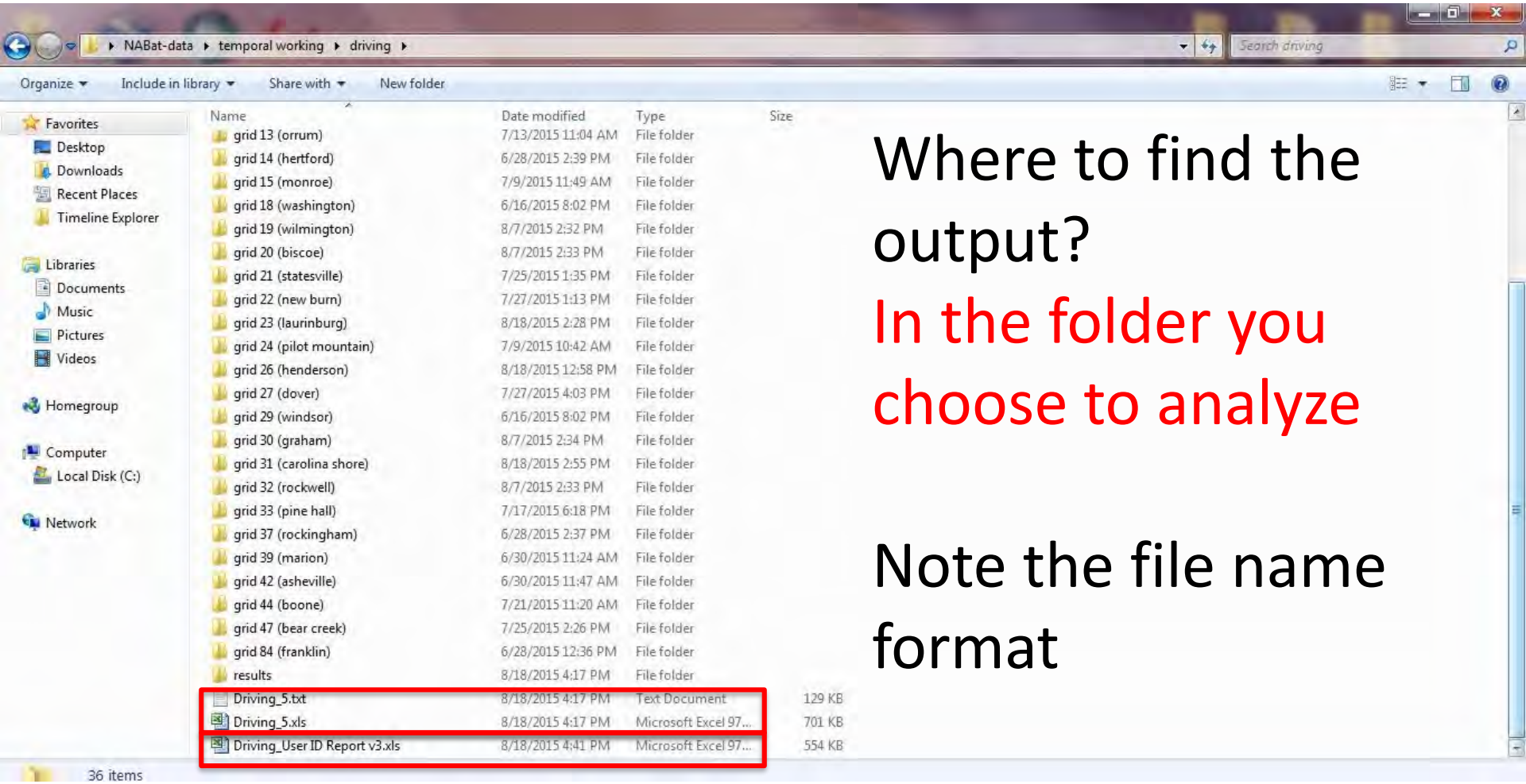
- Other possible software
  - Kaleidoscope (support AnaBat recording)
  - SonoBat (does NOT support AnaBat recording)



Step 4 (continue)

- Interpret software outputs
  - Outputs are always in the folder that has been chosen to analyze
  - BCID names outputs as “folder name_5” and provides two files (one Excel, one text).
  - EchoClass names outputs as “folder name_User ID Report v3” in one Excel file.

BCID: always “folder name_5”; two files



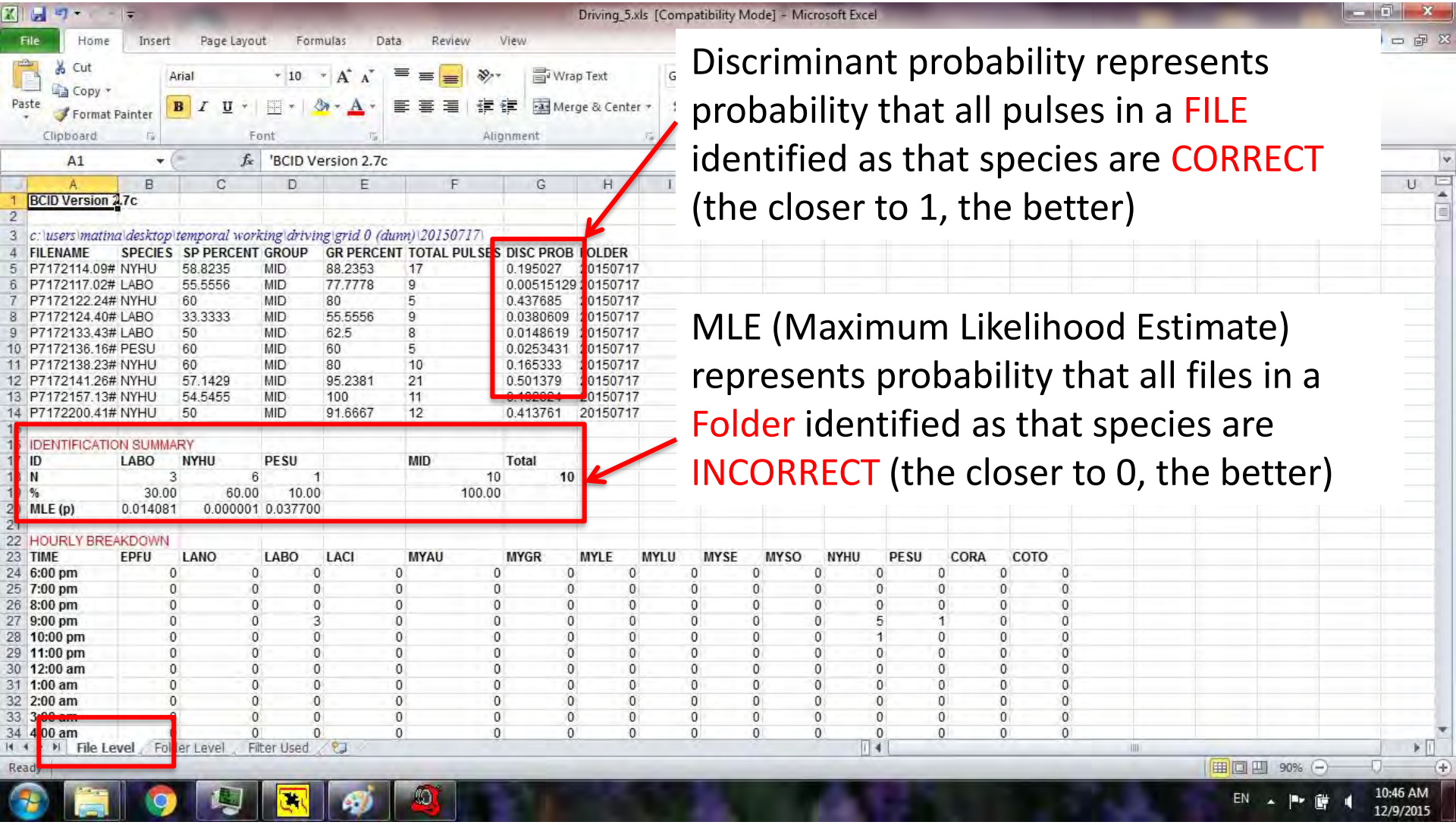
Where to find the output?

In the folder you choose to analyze

Note the file name format

EchoClass: always “folder name_User ID Report v3”; one file

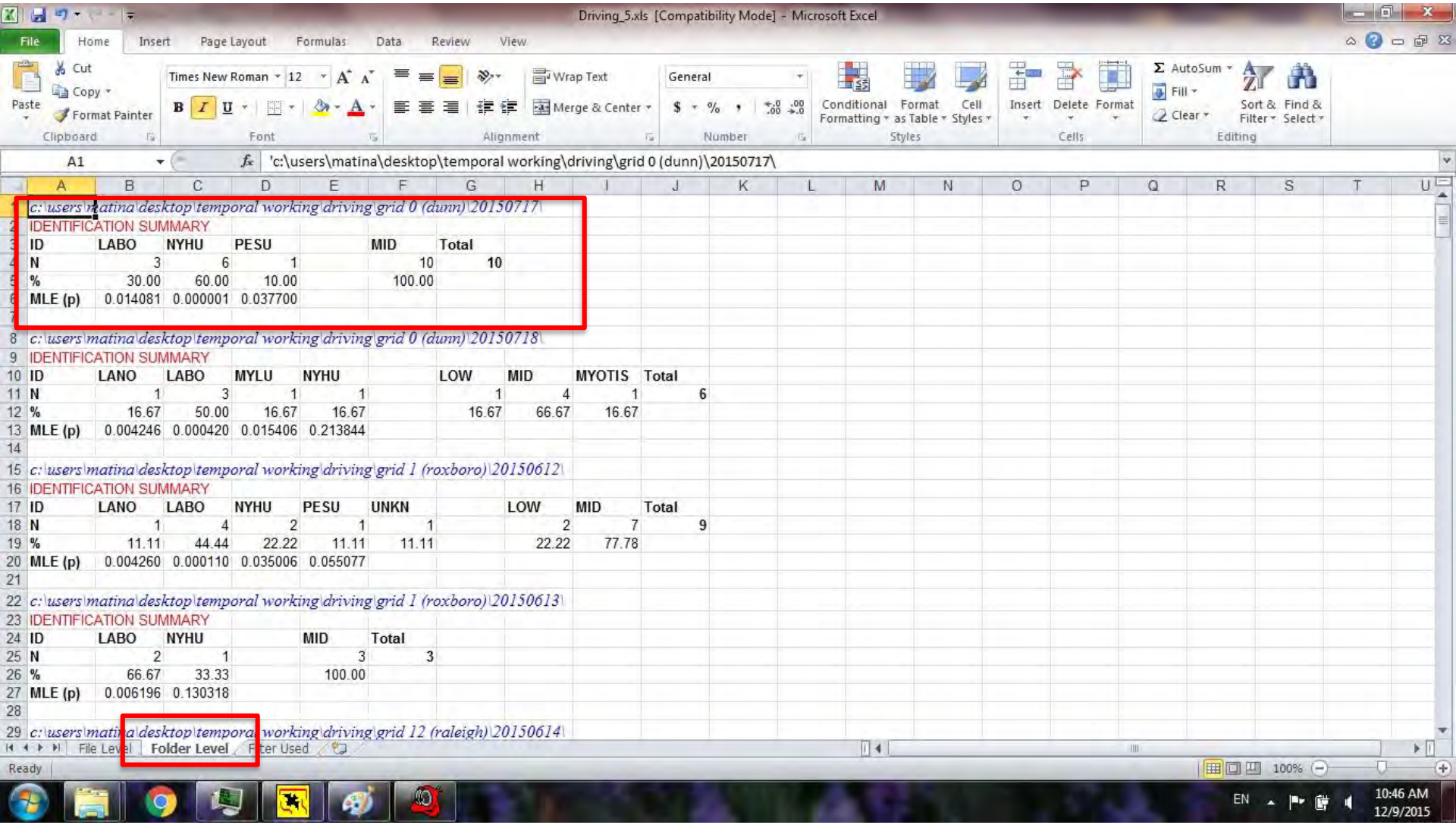
BCID output: File Level



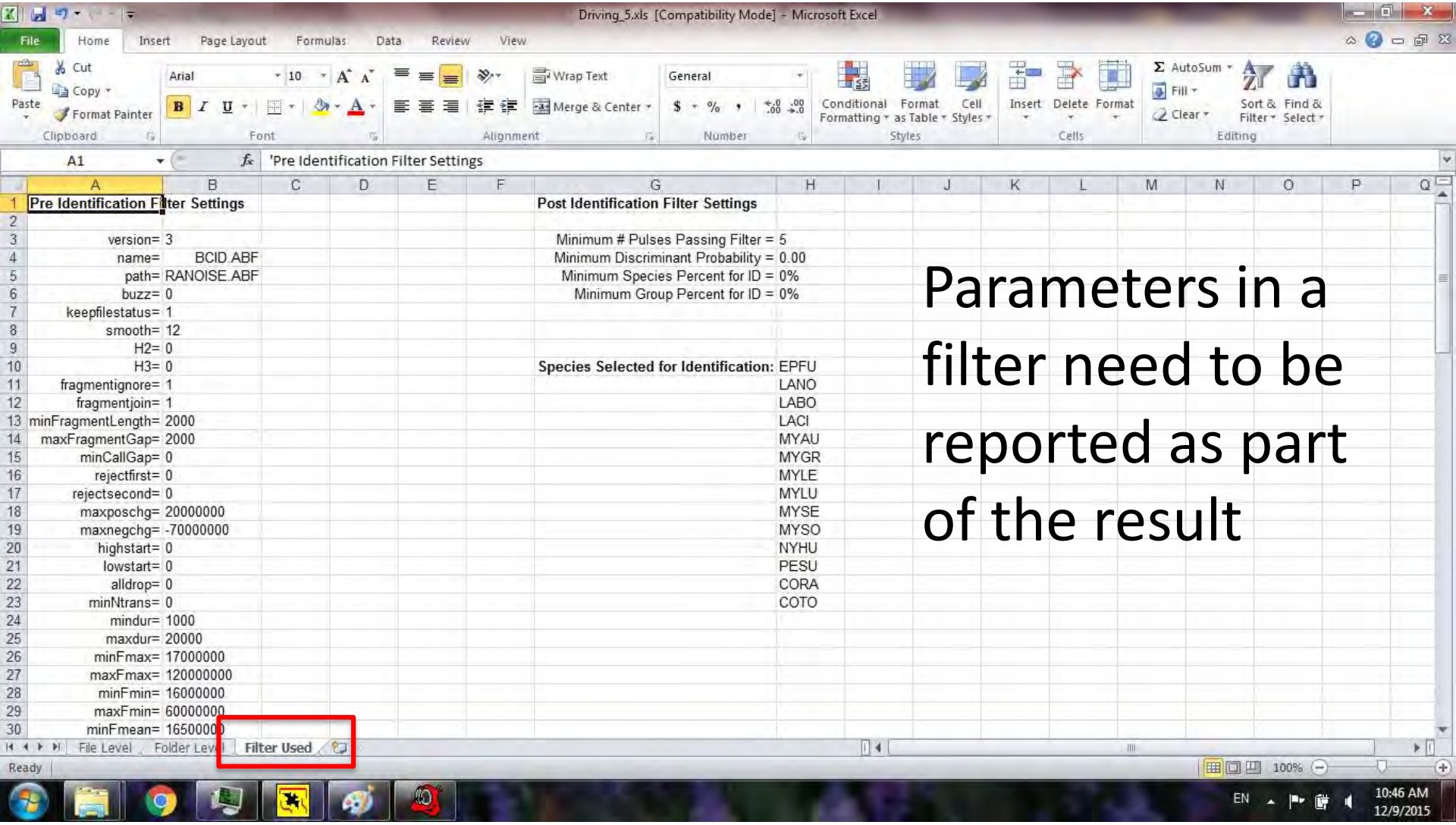
Discriminant probability represents probability that all pulses in a FILE identified as that species are CORRECT (the closer to 1, the better)

MLE (Maximum Likelihood Estimate) represents probability that all files in a Folder identified as that species are INCORRECT (the closer to 0, the better)

BCID output: Folder Level

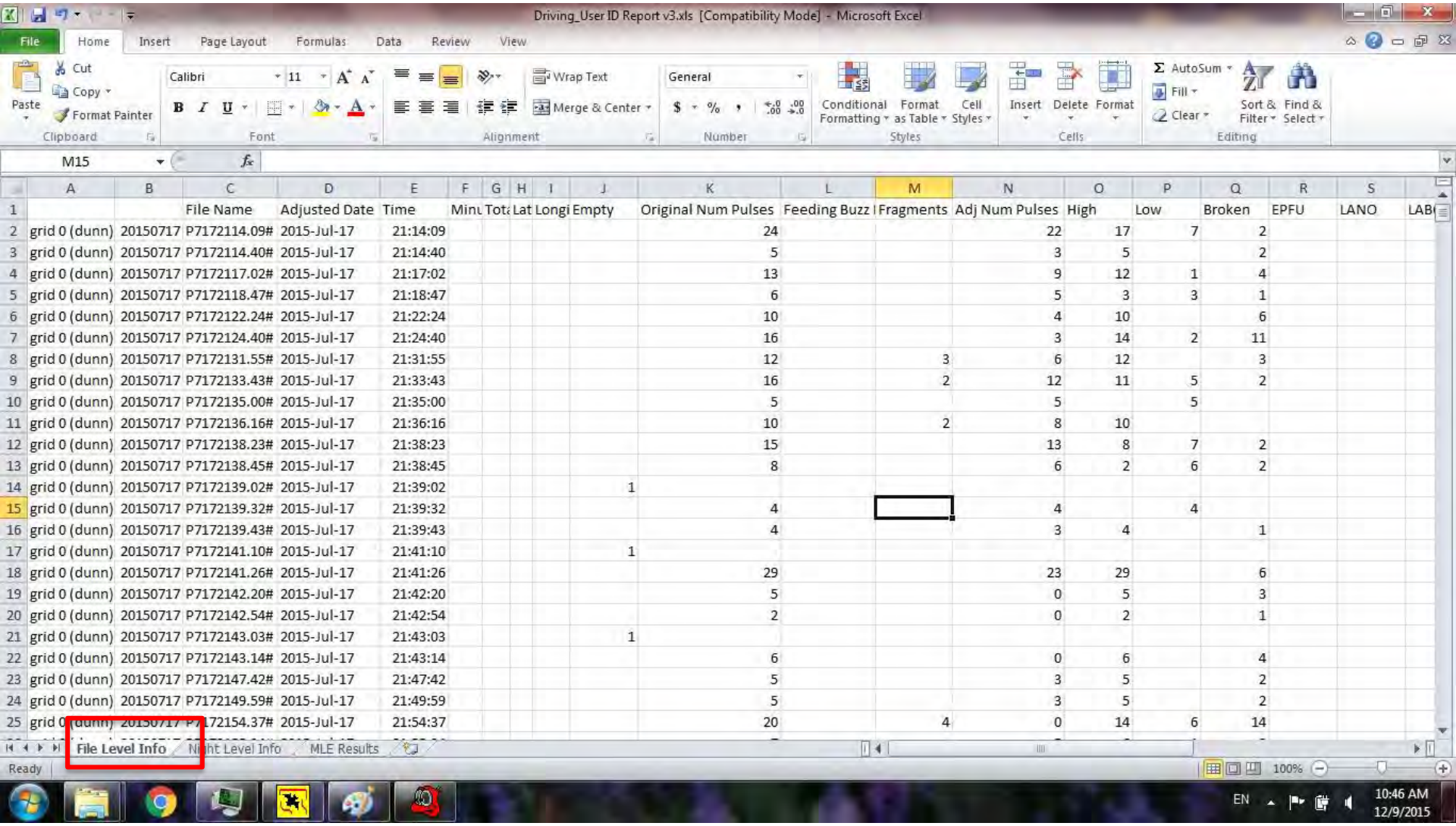


BCID output: Filter Used



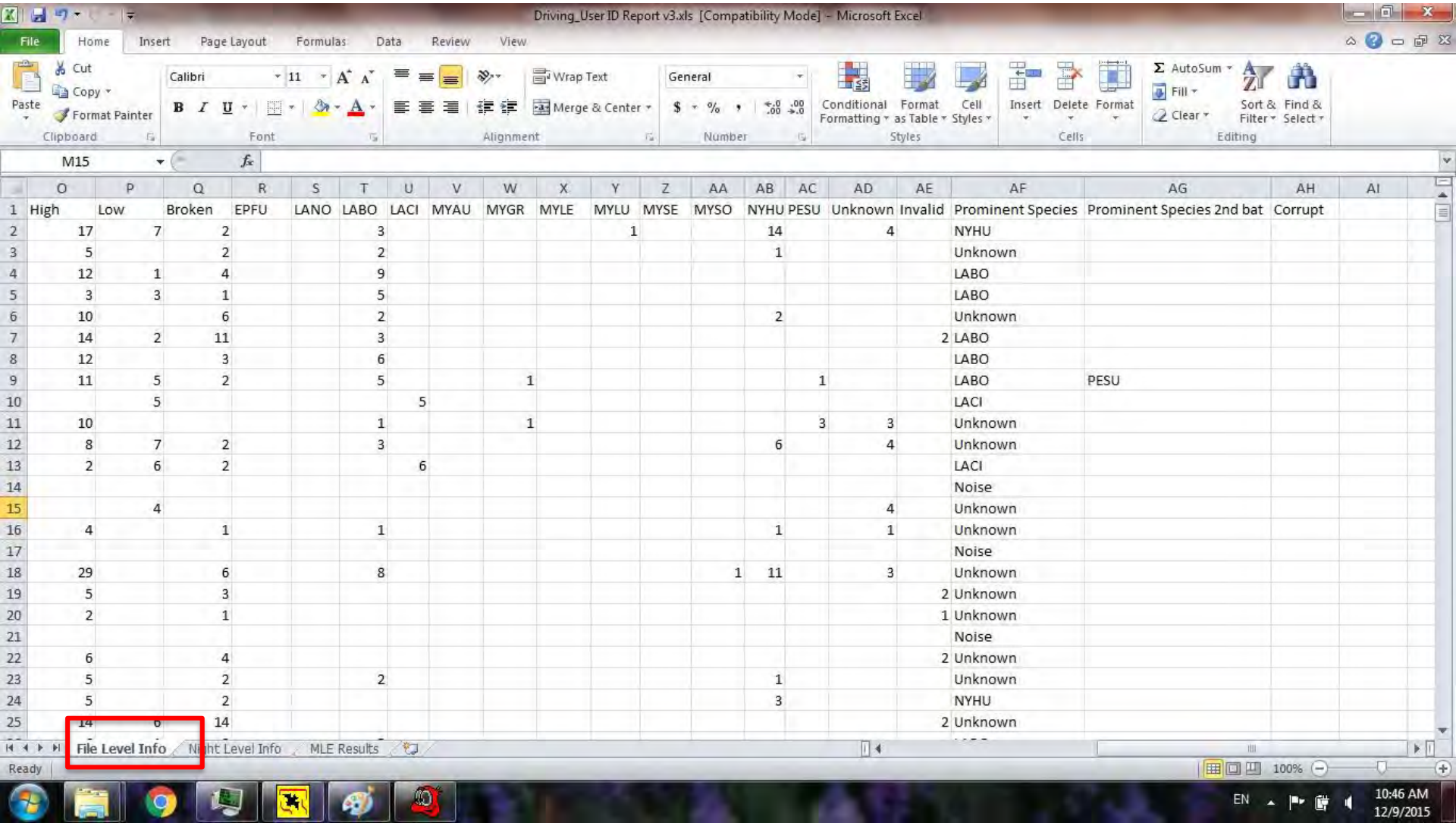
Parameters in a filter need to be reported as part of the result

EchoClass output: File Level



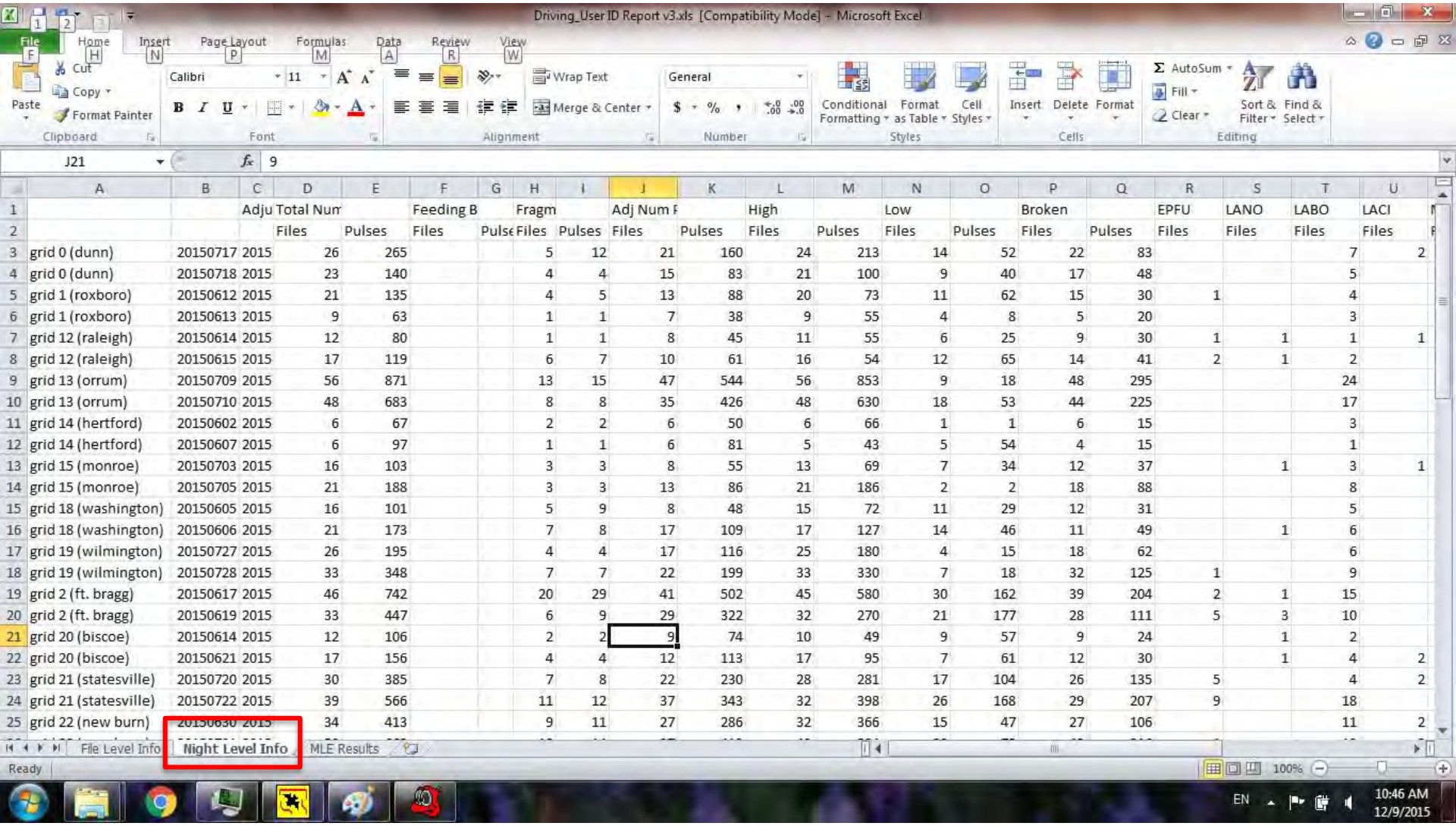
	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S
1			File Name	Adjusted Date	Time	Minu	Tota	Lat	Longi	Empty	Original Num Pulses	Feeding Buzz	Fragments	Adj Num Pulses	High	Low	Broken	EPFU	LANO
2	grid 0 (dunn)	20150717	P7172114.09#	2015-Jul-17	21:14:09						24			22	17	7	2		
3	grid 0 (dunn)	20150717	P7172114.40#	2015-Jul-17	21:14:40						5			3	5		2		
4	grid 0 (dunn)	20150717	P7172117.02#	2015-Jul-17	21:17:02						13			9	12	1	4		
5	grid 0 (dunn)	20150717	P7172118.47#	2015-Jul-17	21:18:47						6			5	3	3	1		
6	grid 0 (dunn)	20150717	P7172122.24#	2015-Jul-17	21:22:24						10			4	10		6		
7	grid 0 (dunn)	20150717	P7172124.40#	2015-Jul-17	21:24:40						16			3	14	2	11		
8	grid 0 (dunn)	20150717	P7172131.55#	2015-Jul-17	21:31:55						12		3	6	12		3		
9	grid 0 (dunn)	20150717	P7172133.43#	2015-Jul-17	21:33:43						16		2	12	11	5	2		
10	grid 0 (dunn)	20150717	P7172135.00#	2015-Jul-17	21:35:00						5			5		5			
11	grid 0 (dunn)	20150717	P7172136.16#	2015-Jul-17	21:36:16						10		2	8	10				
12	grid 0 (dunn)	20150717	P7172138.23#	2015-Jul-17	21:38:23						15			13	8	7	2		
13	grid 0 (dunn)	20150717	P7172138.45#	2015-Jul-17	21:38:45						8			6	2	6	2		
14	grid 0 (dunn)	20150717	P7172139.02#	2015-Jul-17	21:39:02					1									
15	grid 0 (dunn)	20150717	P7172139.32#	2015-Jul-17	21:39:32						4			4		4			
16	grid 0 (dunn)	20150717	P7172139.43#	2015-Jul-17	21:39:43						4			3	4		1		
17	grid 0 (dunn)	20150717	P7172141.10#	2015-Jul-17	21:41:10					1									
18	grid 0 (dunn)	20150717	P7172141.26#	2015-Jul-17	21:41:26						29			23	29		6		
19	grid 0 (dunn)	20150717	P7172142.20#	2015-Jul-17	21:42:20						5			0	5		3		
20	grid 0 (dunn)	20150717	P7172142.54#	2015-Jul-17	21:42:54						2			0	2		1		
21	grid 0 (dunn)	20150717	P7172143.03#	2015-Jul-17	21:43:03					1									
22	grid 0 (dunn)	20150717	P7172143.14#	2015-Jul-17	21:43:14						6			0	6		4		
23	grid 0 (dunn)	20150717	P7172147.42#	2015-Jul-17	21:47:42						5			3	5		2		
24	grid 0 (dunn)	20150717	P7172149.59#	2015-Jul-17	21:49:59						5			3	5		2		
25	grid 0 (dunn)	20150717	P7172154.37#	2015-Jul-17	21:54:37						20		4	0	14	6	14		

EchoClass output: File Level (continue)



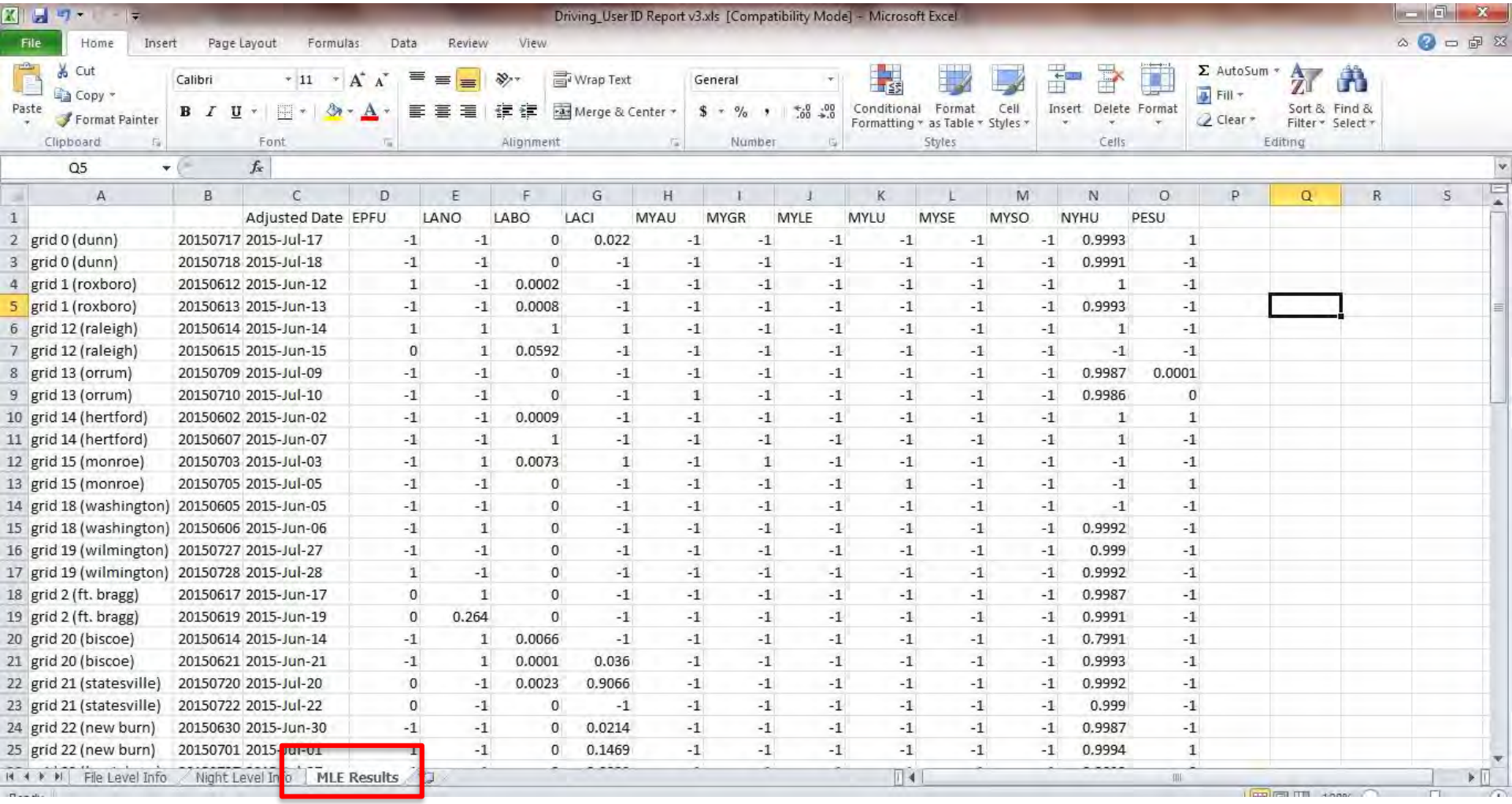
	O	P	Q	R	S	T	U	V	W	X	Y	Z	AA	AB	AC	AD	AE	AF	AG	AH	AI
1	High	Low	Broken	EPFU	LANO	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYHU	PESU	Unknown	Invalid	Prominent Species	Prominent Species 2nd bat	Corrupt	
2	17	7	2			3					1			14		4		NYHU			
3	5		2			2								1				Unknown			
4	12	1	4			9												LABO			
5	3	3	1			5												LABO			
6	10		6			2								2				Unknown			
7	14	2	11			3											2	LABO			
8	12		3			6												LABO			
9	11	5	2			5			1						1			LABO	PESU		
10		5					5											LACI			
11	10					1			1						3	3		Unknown			
12	8	7	2			3								6		4		Unknown			
13	2	6	2				6											LACI			
14																		Noise			
15		4														4		Unknown			
16	4		1			1								1		1		Unknown			
17																		Noise			
18	29		6			8							1	11		3		Unknown			
19	5		3														2	Unknown			
20	2		1														1	Unknown			
21																		Noise			
22	6		4														2	Unknown			
23	5		2			2								1				Unknown			
24	5		2											3				NYHU			
25	14	6	14														2	Unknown			

EchoClass output: Night Level



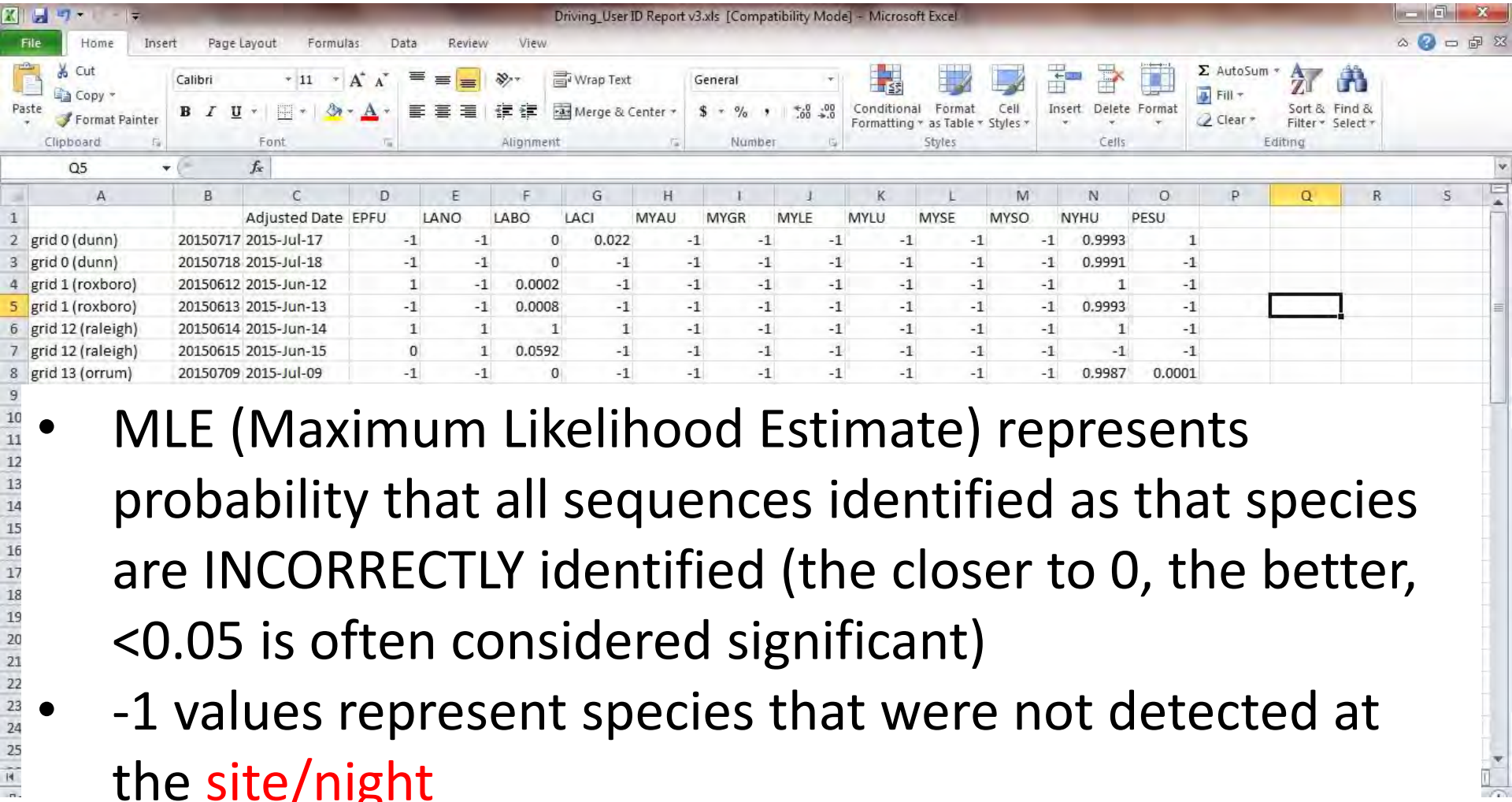
	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U
1			Adju	Total Num		Feeding B		Fragm		Adj Num F		High		Low		Broken		EPFU	LANO	LABO	LACI
2				Files	Pulses	Files	Pulse	Files	Pulses	Files	Pulses	Files	Pulses	Files	Pulses	Files	Pulses	Files	Files	Files	Files
3	grid 0 (dunn)	20150717	2015	26	265			5	12	21	160	24	213	14	52	22	83			7	2
4	grid 0 (dunn)	20150718	2015	23	140			4	4	15	83	21	100	9	40	17	48			5	
5	grid 1 (roxboro)	20150612	2015	21	135			4	5	13	88	20	73	11	62	15	30	1		4	
6	grid 1 (roxboro)	20150613	2015	9	63			1	1	7	38	9	55	4	8	5	20			3	
7	grid 12 (raleigh)	20150614	2015	12	80			1	1	8	45	11	55	6	25	9	30	1	1	1	1
8	grid 12 (raleigh)	20150615	2015	17	119			6	7	10	61	16	54	12	65	14	41	2	1	2	
9	grid 13 (orrum)	20150709	2015	56	871			13	15	47	544	56	853	9	18	48	295			24	
10	grid 13 (orrum)	20150710	2015	48	683			8	8	35	426	48	630	18	53	44	225			17	
11	grid 14 (hertford)	20150602	2015	6	67			2	2	6	50	6	66	1	1	6	15			3	
12	grid 14 (hertford)	20150607	2015	6	97			1	1	6	81	5	43	5	54	4	15			1	
13	grid 15 (monroe)	20150703	2015	16	103			3	3	8	55	13	69	7	34	12	37		1	3	1
14	grid 15 (monroe)	20150705	2015	21	188			3	3	13	86	21	186	2	2	18	88			8	
15	grid 18 (washington)	20150605	2015	16	101			5	9	8	48	15	72	11	29	12	31			5	
16	grid 18 (washington)	20150606	2015	21	173			7	8	17	109	17	127	14	46	11	49		1	6	
17	grid 19 (wilmington)	20150727	2015	26	195			4	4	17	116	25	180	4	15	18	62			6	
18	grid 19 (wilmington)	20150728	2015	33	348			7	7	22	199	33	330	7	18	32	125	1		9	
19	grid 2 (ft. bragg)	20150617	2015	46	742			20	29	41	502	45	580	30	162	39	204	2	1	15	
20	grid 2 (ft. bragg)	20150619	2015	33	447			6	9	29	322	32	270	21	177	28	111	5	3	10	
21	grid 20 (biscoe)	20150614	2015	12	106			2	2	9	74	10	49	9	57	9	24		1	2	
22	grid 20 (biscoe)	20150621	2015	17	156			4	4	12	113	17	95	7	61	12	30		1	4	2
23	grid 21 (statesville)	20150720	2015	30	385			7	8	22	230	28	281	17	104	26	135	5		4	2
24	grid 21 (statesville)	20150722	2015	39	566			11	12	37	343	32	398	26	168	29	207	9		18	
25	grid 22 (new burn)	20150630	2015	34	413			9	11	27	286	32	366	15	47	27	106			11	2

EchoClass output: MLE Results



	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S
1			Adjusted Date	EPFU	LANO	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYHU	PESU				
2	grid 0 (dunn)	20150717	2015-Jul-17	-1	-1	0	0.022	-1	-1	-1	-1	-1	-1	0.9993	1				
3	grid 0 (dunn)	20150718	2015-Jul-18	-1	-1	0	-1	-1	-1	-1	-1	-1	-1	0.9991	-1				
4	grid 1 (roxboro)	20150612	2015-Jun-12	1	-1	0.0002	-1	-1	-1	-1	-1	-1	-1	1	-1				
5	grid 1 (roxboro)	20150613	2015-Jun-13	-1	-1	0.0008	-1	-1	-1	-1	-1	-1	-1	0.9993	-1				
6	grid 12 (raleigh)	20150614	2015-Jun-14	1	1	1	1	-1	-1	-1	-1	-1	-1	1	-1				
7	grid 12 (raleigh)	20150615	2015-Jun-15	0	1	0.0592	-1	-1	-1	-1	-1	-1	-1	-1	-1				
8	grid 13 (orrum)	20150709	2015-Jul-09	-1	-1	0	-1	-1	-1	-1	-1	-1	-1	0.9987	0.0001				
9	grid 13 (orrum)	20150710	2015-Jul-10	-1	-1	0	-1	1	-1	-1	-1	-1	-1	0.9986	0				
10	grid 14 (hertford)	20150602	2015-Jun-02	-1	-1	0.0009	-1	-1	-1	-1	-1	-1	-1	1	1				
11	grid 14 (hertford)	20150607	2015-Jun-07	-1	-1	1	-1	-1	-1	-1	-1	-1	-1	1	-1				
12	grid 15 (monroe)	20150703	2015-Jul-03	-1	1	0.0073	1	-1	1	-1	-1	-1	-1	-1	-1				
13	grid 15 (monroe)	20150705	2015-Jul-05	-1	-1	0	-1	-1	-1	-1	1	-1	-1	-1	1				
14	grid 18 (washington)	20150605	2015-Jun-05	-1	-1	0	-1	-1	-1	-1	-1	-1	-1	-1	-1				
15	grid 18 (washington)	20150606	2015-Jun-06	-1	1	0	-1	-1	-1	-1	-1	-1	-1	0.9992	-1				
16	grid 19 (wilmington)	20150727	2015-Jul-27	-1	-1	0	-1	-1	-1	-1	-1	-1	-1	0.999	-1				
17	grid 19 (wilmington)	20150728	2015-Jul-28	1	-1	0	-1	-1	-1	-1	-1	-1	-1	0.9992	-1				
18	grid 2 (ft. bragg)	20150617	2015-Jun-17	0	1	0	-1	-1	-1	-1	-1	-1	-1	0.9987	-1				
19	grid 2 (ft. bragg)	20150619	2015-Jun-19	0	0.264	0	-1	-1	-1	-1	-1	-1	-1	0.9991	-1				
20	grid 20 (biscoe)	20150614	2015-Jun-14	-1	1	0.0066	-1	-1	-1	-1	-1	-1	-1	0.7991	-1				
21	grid 20 (biscoe)	20150621	2015-Jun-21	-1	1	0.0001	0.036	-1	-1	-1	-1	-1	-1	0.9993	-1				
22	grid 21 (statesville)	20150720	2015-Jul-20	0	-1	0.0023	0.9066	-1	-1	-1	-1	-1	-1	0.9992	-1				
23	grid 21 (statesville)	20150722	2015-Jul-22	0	-1	0	-1	-1	-1	-1	-1	-1	-1	0.999	-1				
24	grid 22 (new burn)	20150630	2015-Jun-30	-1	-1	0	0.0214	-1	-1	-1	-1	-1	-1	0.9987	-1				
25	grid 22 (new burn)	20150701	2015-Jul-01	1	-1	0	0.1469	-1	-1	-1	-1	-1	-1	0.9994	1				

EchoClass output: MLE Results (continue)



	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S
1			Adjusted Date	EPFU	LANO	LABO	LACI	MYAU	MYGR	MYLE	MYLU	MYSE	MYSO	NYHU	PESU				
2	grid 0 (dunn)	20150717	2015-Jul-17	-1	-1	0	0.022	-1	-1	-1	-1	-1	-1	0.9993	1				
3	grid 0 (dunn)	20150718	2015-Jul-18	-1	-1	0	-1	-1	-1	-1	-1	-1	-1	0.9991	-1				
4	grid 1 (roxboro)	20150612	2015-Jun-12	1	-1	0.0002	-1	-1	-1	-1	-1	-1	-1	1	-1				
5	grid 1 (roxboro)	20150613	2015-Jun-13	-1	-1	0.0008	-1	-1	-1	-1	-1	-1	-1	0.9993	-1				
6	grid 12 (raleigh)	20150614	2015-Jun-14	1	1	1	1	-1	-1	-1	-1	-1	-1	1	-1				
7	grid 12 (raleigh)	20150615	2015-Jun-15	0	1	0.0592	-1	-1	-1	-1	-1	-1	-1	-1	-1				
8	grid 13 (orrum)	20150709	2015-Jul-09	-1	-1	0	-1	-1	-1	-1	-1	-1	-1	0.9987	0.0001				

- MLE (Maximum Likelihood Estimate) represents probability that all sequences identified as that species are INCORRECTLY identified (the closer to 0, the better, <0.05 is often considered significant)
- -1 values represent species that were not detected at the site/night
- 1 values represent species that only had 1 sequence detected at the site/night

Step 5

- Software outputs concordance
  - Use Excel vlookup() and if() function
  - Echoclass and Kaleidoscope results are directly comparable
  - BCID results need vlookup() function
  - The goal is to find files that were identified as the same species by different programes

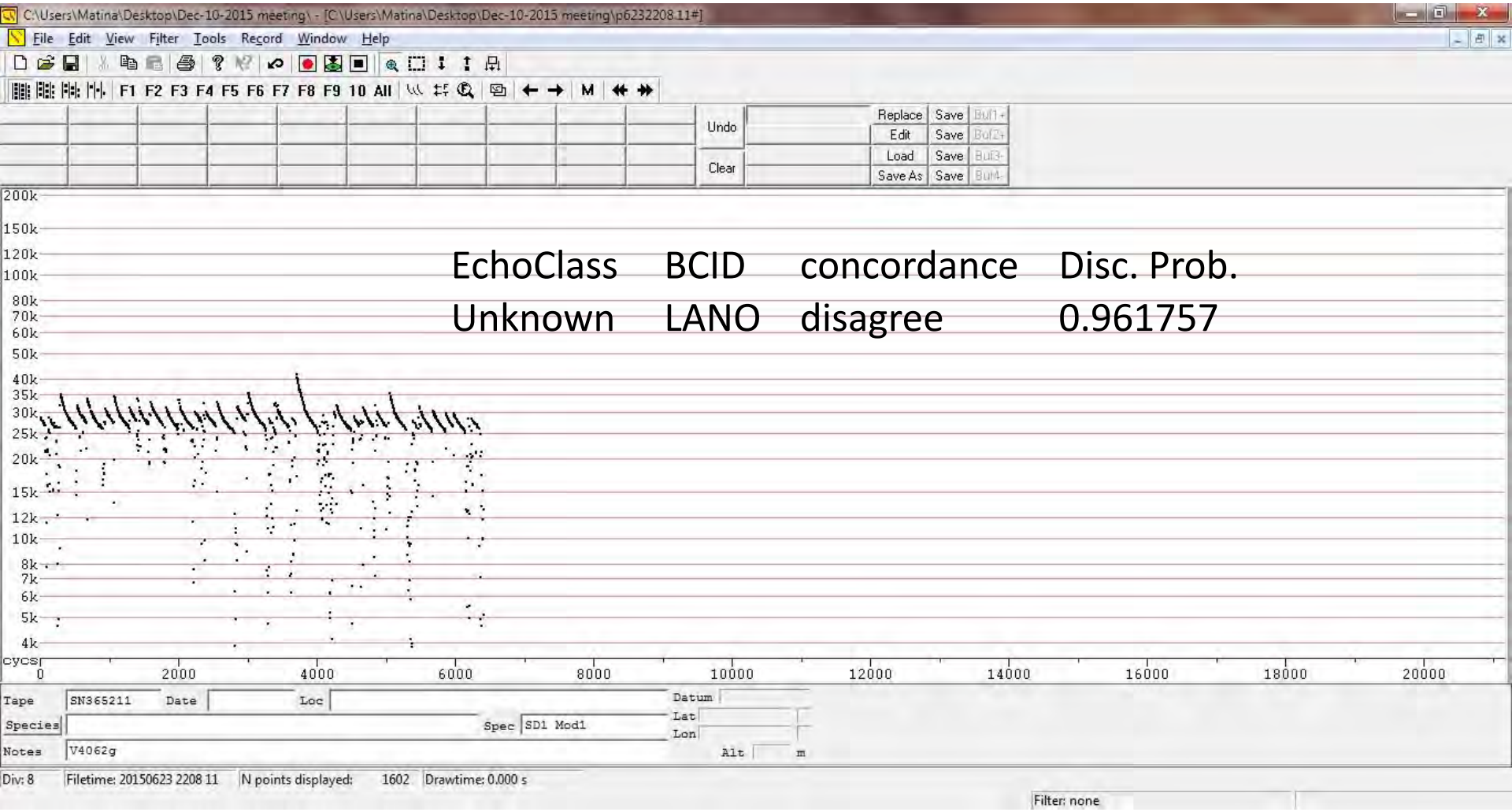
Software concordance

	A	B	C	D	E	F	G	H	I	J	K	L
1	id	Prominent Species	Prominent Species 2nd bat	bcid result	concordance							
2	GL cross road-P6012130.18	Unknown		NOID	disagree							
3	GL cross road-P6012228.14	Unknown		NOID	disagree			GL cross road	GL edge	GL ferry	all	
4	GL cross road-P6012305.39	LABO		NYHU	disagree		range	2	191	336	443	
5	GL cross road-P6012312.32	LABO		LABO	LABO		disagree	158	134	103	396	
6	GL cross road-P6012325.36	Unknown		MYLU	disagree		LABO	23	3	0	26	
7	GL cross road-P6020013.03	LABO		NOID	disagree		MYSO	2	1	0	3	
8	GL cross road-P6020013.30	Unknown		LABO	disagree		EPFU	2	3	0	5	
9	GL cross road-P6020119.31	MYSO		MYSO	MYSO		NYHU	2	2	2	6	
10	GL cross road-P6020152.21	LABO		NYHU	disagree		MYLU	2	0	0	2	
11	GL cross road-P6020210.53	Unknown		MYLU	disagree		PESU	0	1	1	2	
12	GL cross road-P6020242.13	MYSO		MYSO	MYSO		MYLE	0	1	0	1	
13	GL cross road-P6020248.36	Unknown		NOID	disagree		LACI	0	0	1	1	
14	GL cross road-P6020254.25	Unknown		NYHU	disagree		agreed	31	11	4	46	
15	GL cross road-P6020254.40	Unknown		NYHU	disagree		total	189	145	107	442	
16	GL cross road-P6020259.36	Unknown		LABO	disagree			0.16402116	0.07586207	0.03738318	0.1040724	
17	GL cross road-P6020301.38	Unknown		LABO	disagree							
18	GL cross road-P6020309.03	EPFU		EPFU	EPFU							
19	GL cross road-P6020312.25	MYLU		NYHU	disagree							
20	GL cross road-P6020312.49	Unknown		NYHU	disagree							
21	GL cross road-P6020317.15	Unknown		NYHU	disagree							
22	GL cross road-P6020318.01	LABO		NOID	disagree							
23	GL cross road-P6020325.26	Unknown		EPFU	disagree							
24	GL cross road-P6020345.36	EPFU		NOID	disagree							
25	GL cross road-P6020350.00	LABO		LABO	LABO							
26	GL cross road-P6020351.35	EPFU		NOID	disagree							
27	GL cross road-P6020355.06	EPFU		LANO	disagree							
28	GL cross road-P6020356.02	Unknown		LANO	disagree							
29	GL cross road-P6020400.46	EPFU		NOID	disagree							
30	GL cross road-P6020401.52	EPFU		NOID	disagree							
31	GL cross road-P6020401.59	LABO		NOID	disagree							
32	GL cross road-P6020411.02	LABO		CORA	disagree							
33	GL cross road-P6020413.54	Unknown		MYLU	disagree							
34	GL cross road-P6020416.20	Unknown		EPFU	disagree							
35	GL cross road-P6020419.07	EPFU		LANO	disagree							
36	GL cross road-P6020422.59	LABO		LABO	LABO							
37	GL cross road-P6020425.56	Unknown		EPFU	disagree							
38	GL cross road-P6020427.08	NYHU		NYHU	NYHU							
39	GL cross road-P6020428.00	LABO		NYHU	disagree							
40	GL cross road-P6020428.26	LABO		NYHU	disagree							
41	GL cross road-P6022158.08	Unknown		LABO	disagree							
42	GL cross road-P6022213.49	LABO		LABO	LABO							
43	GL cross road-P6022216.46	Unknown		PESU	disagree							
44	GL cross road-P6022218.55	Unknown		NYHU	disagree							
45	GL cross road-P6022219.26	LABO		LABO	LABO							
46	GL cross road-P6022219.42	Unknown		NYHU	disagree							
47	GL cross road-P6022220.24	LABO		NYHU	disagree							
48	GL cross road-P6022229.59	Unknown		NOID	disagree							

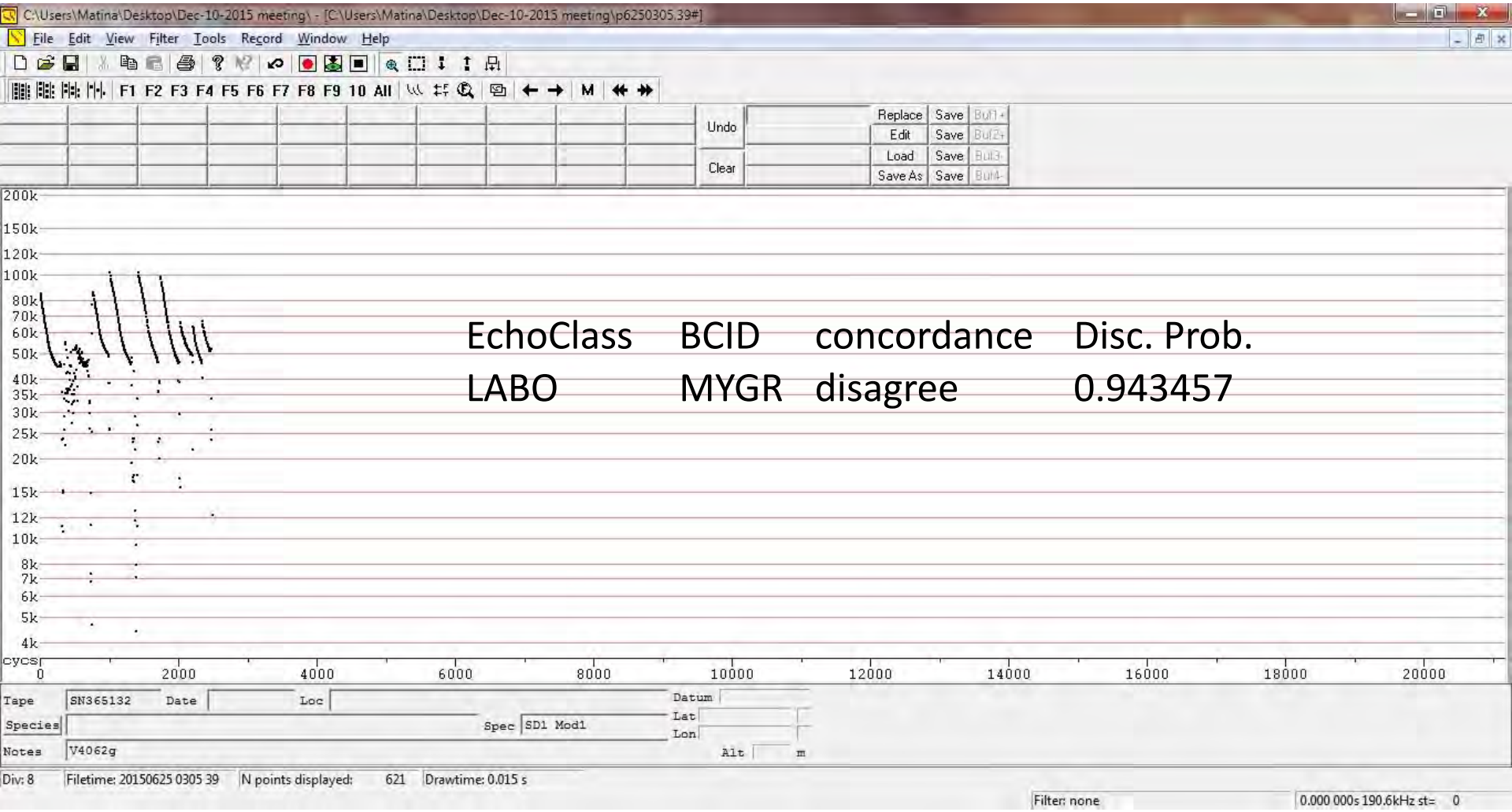
Step 6

- Manual verification/identification
  - Visual verification/identification is extremely important
    - Why?

High discriminant probability in BCID but not agreed by both programs

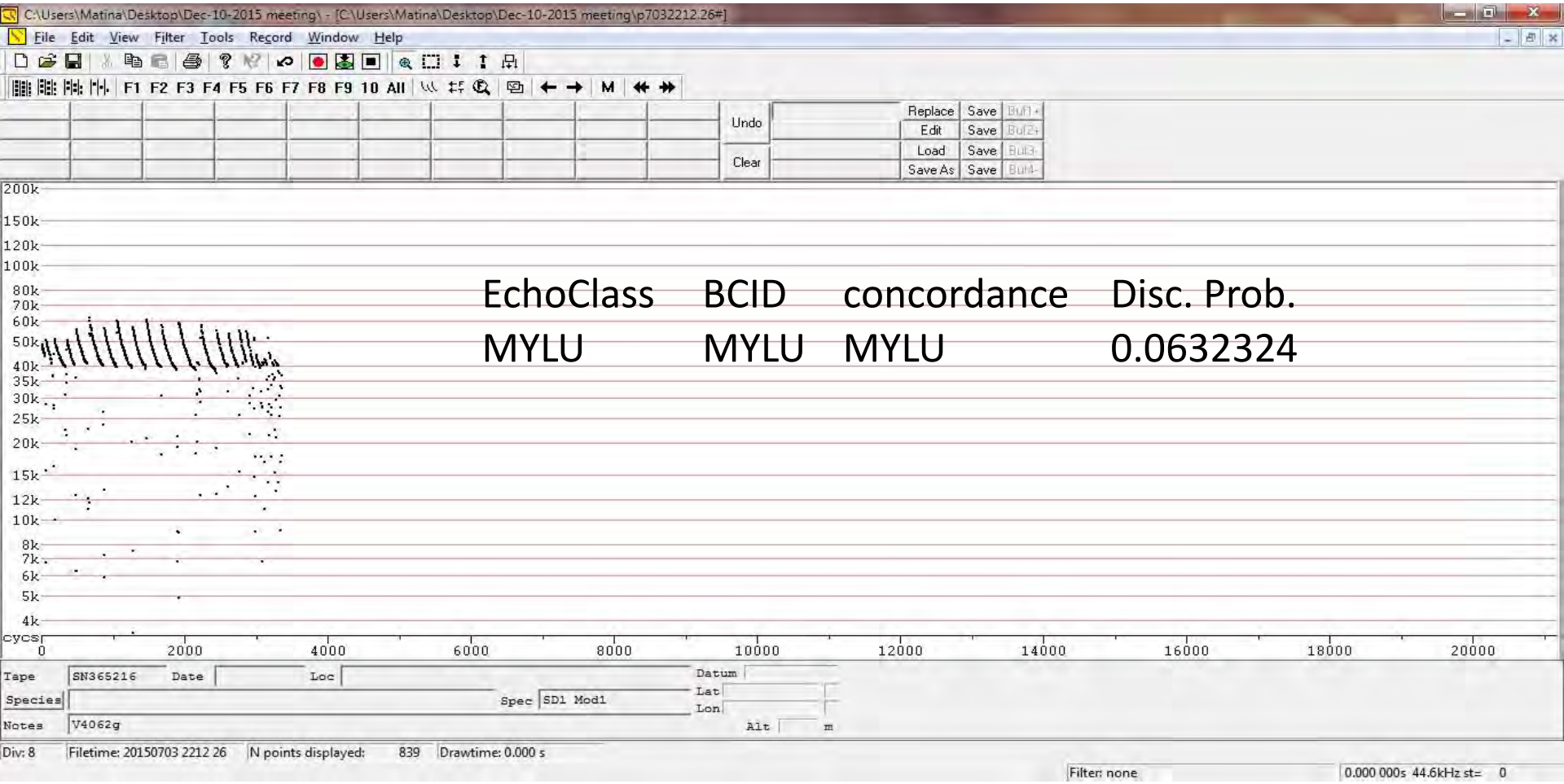


High discriminant probability in BCID but not agreed by both programs



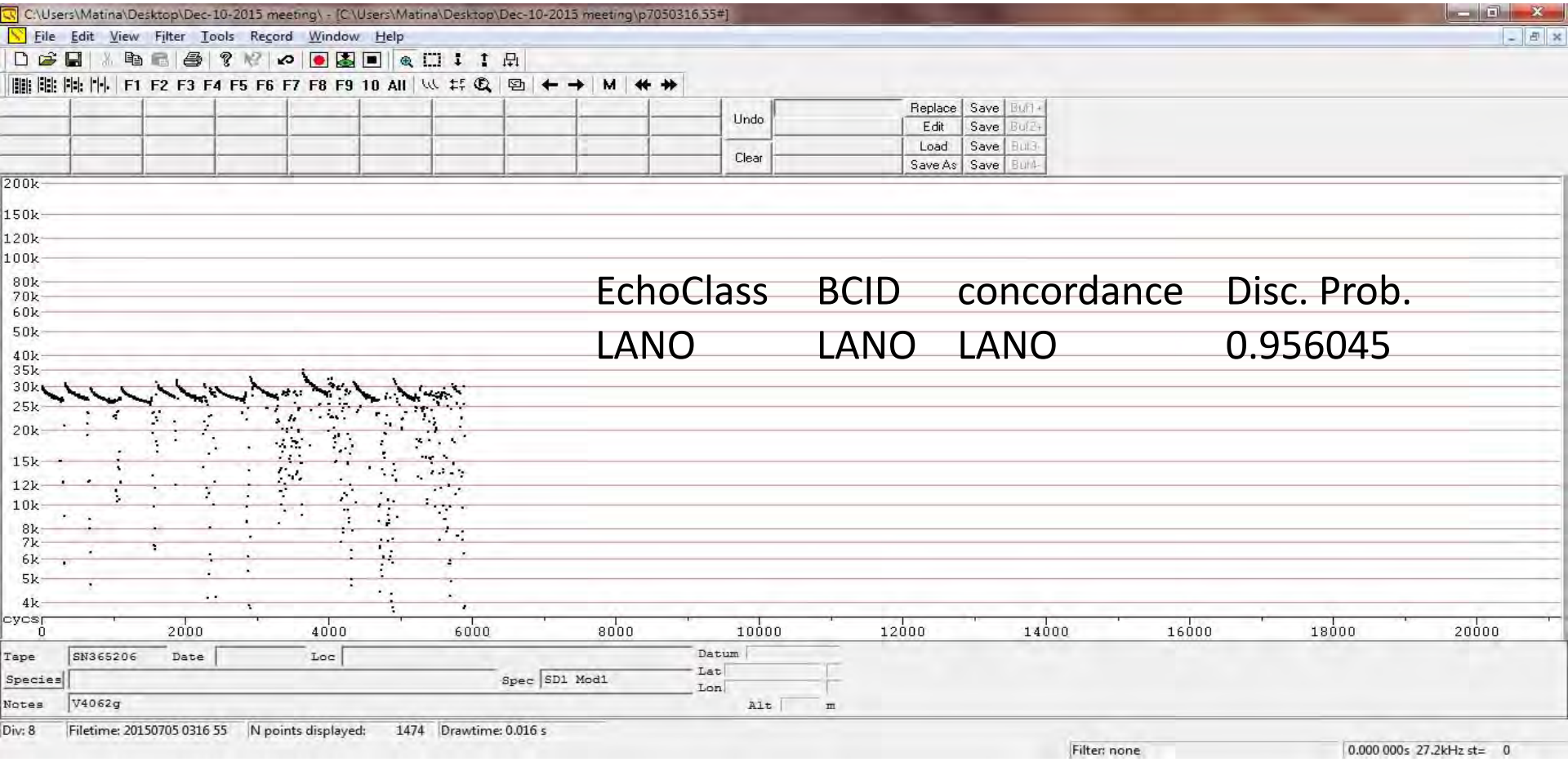
EchoClass	BCID	concordance	Disc. Prob.
LABO	MYGR	disagree	0.943457

Agreed by both programs but low (very low) discriminant probability in BCID



EchoClass	BCID	concordance	Disc. Prob.
MYLU	MYLU	MYLU	0.0632324

Agreed by both programs and high discriminant probability in BCID. But it still can be a different species



EchoClass	BCID	concordance	Disc. Prob.
LANO	LANO	LANO	0.956045

Step 6 (continue)

- Manual verification/identification
  - Pitfalls of automated identification
    - Software gets confused by bad calls/noises
    - Not all species available in the candidate list
    - Certain species are very similar to each other
      - LACI/TABR/LANO
      - TABR/LANO/EPFU
      - EPFU/NYHU/PESU/LABO
      - All Myotis species

Step 6 (continue)

- Manual verification/identification
  - Benefits of doing step 4 and 5
    - Narrow down the number of files to look at
    - Be more confident about a species' presence on a night at a site

Step 6 (continue)

- Typical species characteristics
  - EPFU: very straight vertical between 27-60 KHz
  - LABO: inconsistent minimum frequency varies a lot between 30-45 KHz
  - LACI: minimum frequency below 20 KHz
  - LANO: curvy calls between 25-45 KHz
  - NYHU: curvy calls with minimum frequency near 35 KHz
  - PESU: curvy calls with flat tails with minimum frequency above 40 KHz
  - TABR: flat calls between 20-30 KHz, can vary a lot
  - Myotis spp.: consistent straight vertical calls with minimum frequency near or above 40 KHz and high maximum frequency

Step 7

- Manage field notes
  - Complete weather data using nearest airport weather station data
  - Convert to Excel files or other formats preferred and add to the metadata

Step 8

- Manage GPS data
  - Original files will be a text file or Excel file
  - Manually split into nights/locations
  - Combine GPS data with recordings through matching time stamps

Step 9

- Submit all data to a national data server
- Data format will be determined by the national data server
- National data server will be available by the end of 2017