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Designing effective survey and sampling protocols for passive acoustic monitoring as part of the national bat monitoring

Newson, S.E., Boughey, K.L., Robinson, R.A & Gillings, S.

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For further information please contact:

Joint Nature Conservation Committee Monkstone House City Road Peterborough PE1 1JY www.jncc.gov.uk/

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Summary

The UK has one of the best developed bat monitoring programmes in the world, but despite this, six of 17 resident bat species in the UK are not currently monitored sufficiently to produce population trends. Large-scale deployment of passive acoustic methods using full spectrum bat detector technology offers a lot of potential for transforming routine monitoring of bat numbers, but it is essential that effective survey and sampling protocols are used if this potential is to be realised.

To guide these decisions, we analyse passive acoustic data collected by volunteers from a study area in south-west Britain to quantify the ability and power to detect population declines in bat numbers in relation to: i) whether recording is carried out across the whole night, ii) whether a record / sleep cycle is used, and iii) the number of sites surveyed.

Our results indicate that recording should be carried out over the full night to avoid producing biased estimates of decline. If recording is carried out according to a record / sleep cycle, then the sleep time should be short so that any biases, and reductions in precision and power to detect declines are small. A national scheme should aim to record target bat species on a much larger number of sites than considered in this study. This is likely to require stratification (e.g. by region and/or habitat), to maximise the number of sites that record the most range-restricted species to be able to produce trends for these.

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1 Introduction

British bat populations have been monitored by the Bat Conservation Trust (BCT) since 1996 through the National Bat Monitoring Programme (NBMP), which produces population trends for eleven resident bat species in the UK (Barlow *et al.* 2015) using a mixture of maternity and hibernacula roost counts, as well as field surveys. However, six resident bat species in the UK are not currently monitored sufficiently to produce any population trends, and for all species, there are parts of the UK which are poorly represented.

Many of the NBMP methods use skilled surveyors to conduct bat surveys, which can be resource intensive and limit the number of volunteers that can get involved, so there is an interest in looking at alternative approaches to improve coverage of both species and regions. Passive acoustic monitoring is a growing field of biodiversity monitoring which involves the use of acoustic sensors to collect large volumes of acoustic data (Browning et al. 2017). In the past, the cost of commercial ultrasonic acoustic sensors, and difficulties in automatically detecting (Mac Aodha et al. 2018) and identifying species from their calls (Rydell et al. 2017), have been prohibitive to a rollout of a national passive acoustic survey of bats. However, improvements in low-cost acoustic sensor technologies like the AudioMoth (Hill et al. 2017), and software to automatically detect and classify bat species from their echolocation calls have created new opportunities to monitor more species of bats, at a greater spatial and temporal resolution and coverage, and to engage more people, than is currently possible through the NBMP. With these developments, BCT proposes a new citizen science survey, the British Bat Survey (BBatS). The proposed survey protocol is to use full spectrum bat detector technology, where detectors are left out to record bat calls at their original frequency. When used in conjunction with call identification software and validation (see Newson et al. 2015), this approach offers great potential for transforming bat monitoring in the UK.

Most bat species in the UK can be identified with confidence from their calls, although bats in the genera *Myotis* and *Plecotus* can be challenging, but our knowledge for distinguishing these species is improving rapidly. By archiving recordings, there is the potential to reanalyse them as software and algorithms improve. Using passive detectors in this way, there is the potential to provide representative acoustic monitoring of bat species activity as a measure of distribution and a useful proxy for abundance. Specifically, bat activity represents bat usage of an area, which will be a combination of species abundance, and time spent in the area. This approach can generate a large volume of recordings per night as a measure of relative abundance, but the number of recordings can be highly variable depending on nightly weather conditions, local habitat and use of particular features in the landscape, for example proximity to a roost.

The concept of this study was to explore the potential of passive detector data, and of different sampling regimes for producing robust population trends. Specifically, we were interested in how different sampling regimes are likely to affect the ability and power to detect different levels of decline in bat activity of between 5% and 50%. We focus on bat activity defined as the number of 1-minute periods in a night within which the species was recorded as present. Where such measures of relative abundance are available, they should always be prioritised over occupancy data, as they provide much more sensitive measures of population change (Law *et al.* 2015).

We used data from a study in south-west Britain collected in a pilot project by BCT volunteers and consider two elements that relate to survey effort: timing, and sampling frequency through the night. Firstly, we examined how part-night sampling regimes are likely to influence power. It is well known that bat activity is greatest during the first hours after sunset (Collins 2016), so there may be opportunities for reducing the volume of data

collected, with little loss of power. For this, the following recommended protocols were taken from the Bat Conservation Trust's Good Practice Guidelines (p 57, Collins 2016).

- Recording from 30 minutes prior to sunset to three hours after sunset
- Recording from 30 minutes prior to sunset to three hours after sunset, and then from two hours before sunrise to thirty minutes after sunrise.
- Recording from 30 minutes before sunset to 30 minutes after sunrise.

Secondly we look at how timed sampling regimes (i.e. on / off sampling through a night) to reduce the volume of recordings (*.wav files), are likely to influence the power to detect specified levels of decline, and lastly, how the number of sites surveyed is likely to influence the power to detect decline.

2 Methods

2.1 Study area

This study focuses on the south west of England and south-east Wales (Figure 1). This area was chosen to include the geographic range of three target bat species, Leisler's bat, barbastelle and Nathusius' pipistrelle, that are not currently monitored by the National Bat Monitoring Programme (NBMP), and so are of particular interest to BCT (Table 1). In addition, it was likely that several additional secondary target species, for which improvements in monitoring would be useful, could be recorded in this area. Some of the target species are relatively widespread, while others have a more localised distribution.



Figure 1. Study area.

Table 1. Target bat species

Species

Primary target species

Leisler's bat, *Nyctalus leisleri* barbastelle, *Barbastella barbastellus* Nathusius' pipistrelle, *Pipistrellus nathusii*

Secondary target species

common pipistrelle, *Pipistrellus pipistrellus* soprano pipistrelle, *Pipistrellus pygmaeus* greater horseshoe bat, *Rhinolophus ferrumequinum* lesser horseshoe bat, *Rhinolophus hipposideros* noctule, *Nyctalus noctule* serotine, *Eptesicus serotinus* brown long-eared bat, *Plecotus auritus* Distribution in study area

Localised Widespread Localised during the survey period

Widespread Widespread Widespread Widespread Widespread Widespread

2.2 Survey design

Site selection was based on 1-km OS grid squares. This is a familiar unit of area for BCT volunteers, is used by other terrestrial biodiversity surveillance schemes, and considering the detection distance and movements of bats is a useful resolution at which to characterise the landscape for sample stratification purposes.

AudioMoth devices (<u>https://www.openacousticdevices.info/</u> recording in full spectrum) were available for the survey. The AudioMoths were housed in a weather-resistant modified junction box case and mounted at a height of about 2 m. Prior to deployment, the AudioMoths were configured to make quasi-continuous recordings from approximately 30 minutes before sunset to 30 minutes after sunrise, split into five-minute wav files, and were set to record with medium gain. A sample rate of 384 kHz was used to ensure adequate sampling for lesser horseshoe bat (needed to be over twice the highest frequency of interest), which has a peak frequency of about 115 kHz.

We focused on a short survey period (17 June – 11 August 2019) to coincide with the core activity period for adult bats, but before young are able to fly. To minimise biases in site selection, we created a stratified random sample of 1-km squares in the target area. Square selection was stratified by habitat class according to Centre for Ecology and Hydrology (CEH) Land Cover Map 2015 aggregate habitat classes, to ensure representative coverage of the broad habitats within the region. To allow some flexibility, 100 1-km squares were selected to help ensure that there were enough survey sites to be accessible to all volunteers that wished to take part, who were allowed to select a random 1-km. Because bats are highly mobile, to ensure that survey sites were independent, we set a minimum separation distance between 1-km squares of 9-km, which is 1.5 times the largest core sustenance zone of our target species (6-km for barbastelle).

To minimise biases in the choice of survey point location within 1-km squares, for example towards habitats that are believed to be more important for bats, volunteers were asked to select a location close to the centre of the 1-km square (i.e. a random location with respect to habitat). However, volunteers were allowed some flexibility to ensure that i) the location was safe to access, ii) secure enough to leave an AudioMoth out overnight, iii) not lit by artificial light at night, and iv) was at least 1.5 meters away from vegetation or other obstructions that could mask bat calls, hard surfaces such as tree trunks or walls, which cause echoes that can distort recordings, and water, where reflections of sound off the water surface can distort recordings. Volunteers were asked to try and survey the same location over a minimum of four non-consecutive nights where possible.

2.3 Uncertainty in species identification

Because monitoring on this scale can generate a very large volume of recordings, efficient processing is necessary, which is greatly aided by a semi-automated approach for assigning recording to species. In this study, we made use of an acoustic classifier that is being developed by University College London for use in national bat monitoring (Mac Aodha *et al.* 2018), which provides identification at the call level.

Each bat call was assigned to a species along with a confidence score (continuous value, 0– 1), which tells you how similar an unknown call is to the reference library of known species calls used to build the classifier. Whilst the confidence scores aim to be an indicator of the true success probabilities of the automated identifications, the values are strongly speciesspecific. To address this and following the recommendations in Barre *et al.* (2019), we used manual checking of a stratified random sample of bat calls to model success or failure of correct identification against the confidence score for each species. In doing this, we were able to determine the confidence score for each species that corresponds to the same level of identification error across species. This is illustrated in Table 2, where a confidence score for Leisler's bat of 0.90 and for common pipistrelle of 0.50, both represent a false positive rate of 50%. Calls with an identification error of above 50% were discarded (as advised in Barre *et al.* 2019). Calls with an identification error of more than 0.50 often relate to poor quality calls or calls that contain too little information to be able to assign them to species by any means, so little is lost by discarding these.

In addition, on BCT's advice, we removed identifications where there were fewer than 3 calls (see Table 2) assigned to the same species within a one-minute section, with the exception of lesser horseshoe bat, which produces weak calls, but which can normally be assigned on manual inspection to species with confidence.

Following this, manual checking of spectrograms using software BatSound

(<u>https://batsound.com/</u>) or Kaleidoscope Pro (<u>https://www.wildlifeacoustics.com</u>) was carried out as an independent check of the original species identities. For these analyses, bats in the genus *Myotis* (potentially Daubenton's, Brandt's, whiskered, Alcathoe, Natterer's and Bechstein's bats) and *Plecotus* (potentially brown long-eared or grey long-eared bat) which can be difficult to assign to species, were assigned to genus level only. Whilst there is likely to be some remaining error in species identification, for most calls this is less than 10%, so for the purpose of these analyses, we assume that remaining error is negligible.

Table 2. Bat species detected through the fieldwork, the confidence score threshold (equal to to a 50% false positive rate) below which calls were discarded, and the minimum number of calls assigned to the species within a one-minute section below which calls were discarded.

Species	Species code	Confidence score threshold	Minimum number of calls
Greater horseshoe bat, <i>Rhinolophus</i> ferrumequinum	Rhifer	0.531	3
Lesser horseshoe bat, Rhinolophus hipposideros	Rhihip	0.661	1
noctule, Nyctalus noctule	Nycnoc	0.500	3
Leisler's bat, Nyctalus leisleri	Nyclei	0.900	3
serotine, Eptesicus serotinus	Eptser	0.800	3
Nathusius' pipistrelle, Pipistrellus nathusii	Pipnat	0.900	3
common pipistrelle, Pipistrellus pipistrellus	Pippip	0.500	3
soprano pipistrelle, Pipistrellus pygmaeus	Pippyg	0.500	3
barbastelle, Barbastella barbastellus	Barbar	0.709	3
<i>Myotis</i> species	Myotis	0.500	3
Plecotus species	Plecot	0.865	3

2.4 Data preparation and sampling protocols

All data preparation and analysis were conducted in R (R Core Development Team, 2018). Data preparation involved truncating start times of bat calls (i.e. cutting off the decimal places, so that times were to the minute only.

After establishing whether the species is present in a minute based on the minimum number of calls in Table 2, we then produced a complete dataset for each species and site surveyed for each minute, with a count of 1 where the species was recorded, and 0 where it was not.

Columns for the time since sunset and time after sunrise were added to this. Data preparation then involved coding up a series of columns with 0/1 values to indicate rows that are to be used in different protocols.

Whilst the AudioMoths were set to record quasi-continuously, occasional equipment failure and some drift towards longer sleep periods between recordings when the battery became low, meant that recording was carried out for 92% rather than 100% of the time. Because battery failure occurred towards the end of the night when bat activity is low, we think that the impact on the results will be negligible.

The sampling protocols that we have tested operate at different levels (Table 3). These include the number of sites, which part of the night, and the on/off cycle ('sleep protocol').

	Protocol	Sub-category	Description
1	Which part of the night	All night	30 minutes before sunset to 30 minutes after sunrise
2		Dusk	30 minutes before sunset to 3 hours after sunset
3		Dusk and Dawn	30 minutes before sunset to 3 hours after sunset and 2 hours before sunrise to 30 minutes after sunrise
4		0 sleep	Record continuously
5	Configuration of on/off	1 minute	Record every other minute
6	cycle	4 minutes	Record first 1 minute in every 5 minutes
7		9 minutes	Record first 1 minute for every 10 minutes
8 9 10	Number of sites	25 sites 50 sites 100 sites	25 randomly selected sites (with replacement) 50 randomly selected sites (with replacement) 100 sites randomly selected sites (with replacement)

Table 3. Sampling protocols.

2.5 Limitations and constraints of the protocols and bat data

Before we describe the steps involved in the simulations that we have carried out, it is important to highlight how characteristics of these survey protocols, and limitations of the bat data available to us, constrain how we have been able to perform the power analysis:

- In total only 27 sites were surveyed, on which some species were absent. This
 constrains our ability to assess how the number of sites influences power. We have
 used sampling with replacement to utilise the data as best we can. However, for some
 species, we are essentially re-using the same small amount of data multiple times.
 Below we explain how we have configured the simulations to limit the effect of this
 non-independence.
- It would have been useful to look at power in relation to the number of visits, but the small sample size of sites surveyed, and the fact that only two visits were made to some sites, limits what could be done with the whole dataset without excessively reusing the same data multiple times with replacement, with implications for interpretation. For this reason, we did not explore this here.
- Switching from full night to part night sampling substantially reduces the sample size for some species. This is a valid and useful result, but it limits what we can do with certain species for which we have little data to work with.

- Applying strict filters based on identification uncertainty also substantially reduced the sample size for certain species, limiting the species for which these analyses could be completed.
- In these analyses, we used the number of occupied minutes as the measure of relative abundance. As activity is not uniform through the night, the total number of occupied minutes, or the average per hour, varies between full night, dusk and dawn and dusk-only sampling. Totals and averages are further modified by the sleep protocols. This means that relative abundance measures from these protocols cannot be compared directly. The only valid comparisons that can be made are between change in relative abundance derived from different protocols.
- As the sleep protocol operates at the minute level, any sampling and degrading of the data needs to be undertaken at this level to generate appropriate variation across iterations. This is particularly important across 'sites', where the data for individual sites may have been duplicated during resampling with replacement that is required to generate sample sizes larger than collected during the pilot study.
- With only a single year of data, and no similar independent data on which to base the variability in trend across sites, we have opted for scaling variance proportionally with the mean so that larger declines are more variable across sites than smaller declines. Specifically, to simulate a 25% decline, change values for each site are drawn from a normal distribution of mean 0.25, with variance set to one quarter of the mean. Plots of individual site trajectories (see Figure 2) show what we consider to be appropriate levels of inter-annual variability. Whilst we acknowledge this is a weakness of this study, that is difficult to overcome without further data, we also repeated the analyses with variance set to half of the mean i.e. double the variance (see Table S4). This demonstrates that the variance, within reasonable bounds is not likely to significantly change the power to detect a specified level of decline, or recommendations resulting from it.
- Ideally, we would know how the activity of a given number of bats translates into detected calls per minute, which would allow us to simulate the effect of declines directly. Instead we simulate declines indirectly as declines in bat activity. As defined above bat activity represents bat usage of an area, which will be a combination of species abundance, and time spent in the area.
- We cannot simulate overall population increases because it is not clear as to what procedure should be followed to turn minute-level absence into presences without a much greater understanding of the patterns of within-night presence. Nevertheless, the way we have generated declines allows for some positive inter-annual changes after the first year.

With these above constraints in mind, the steps of the simulations that we have carried out are described as following. The following example is for the Dusk protocol, with 9-minute sleep, 100 sites, all visits and testing for a 25% decline in relative abundance:

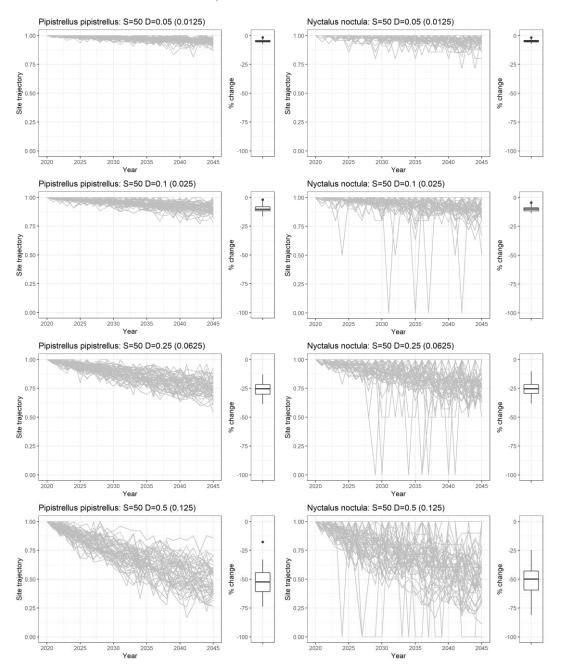
- 1. Obtain all data for a particular species. Each row relates to present (1) or absence (0) in each minute of recording at a site, on a visit, at a particular time.
- 2. Limit to night protocol: remove any minutes that fall outside the Dusk period.
- 3. Limit to sleep protocol: remove rows relating to the 2nd-10th minute in each 10-minute block, creating one minute of active listening and nine minutes of sleep.

- 4. Create sample size: resample with replacement to get a set of 100 sites. As sites need to be treated independently from here onwards, each instance of a site is renamed (e.g. instances of site 31 become 31A, 31B, 31C etc).
- 5. Assign each site a change value: to generate variation across sites, change values for each site are drawn from a normal distribution of mean 0.25 to simulate a 25% decline, with variance set to one quarter of the mean.
- 6. Convert each site's change value into an annual rate of change (λ) over a 25-year period
- 7. Generate relative abundance for each year, from t = 1 to 25:
 - a. Impose the change on minute-level data: for each row of data (= a minute of recording, with a 1 or 0 to indicate whether a bat was detected), generate a random binomial deviate with probability λ^t . This produces a 0 or 1 to indicate whether the minute should be zeroed (i.e. decline) or retained.
 - b. Sum over rows for each visit, site and year to get the total 'abundance' at site *s* on visit *v* in year *t*. Here 'relative abundance' is the number of minutes when detected.
 - c. For trend production, calculate the maximum relative abundance over visits to each site in each year. Note that a plot of the resulting values shows trend lines approximating to a 25% decline.
- 8. Check the actual sample size for trend production: filtering steps 1 & 2 can remove all non-zero counts for certain species, meaning the results of step 7 is a time series of zeroes at a site which is uninformative for trend production and causes problems in model convergence.
- 9. Delete any wholly unoccupied sites and report effective sample size.
- Calculate the trend estimate: fit a fixed effect generalised linear model of the form Count ~ factor(site) + year, with Poisson error distribution and log transformation. Determine whether the slope is significantly different from zero and use the slope estimate to derive a population change estimate over the 25 years.

The steps above are repeated 100 times for each species and protocol to produce:

- 100 estimates from which a trend can be estimated for a given species and protocol.
- The percentage of iterations in which the trend estimate (i.e. slope) was significantly different from zero. We compare this to a nominal value of 80% power.
- The mean population change estimate (and its distribution) to determine whether there is systematic bias in the trend measures returned.

Figure 2. Examples of site-level population trajectories and percent change measures for common pipistrelle and noctule. These are the results of simulated declines (D) (5%, 10%, 25% and 50% declines with the variance shown in brackets) at 50 sites (S) showing the level of inter-site and interannual variation this process generates. The site trajectories are baselined to year 1 for interpretation purposes. In reality the population trajectories would start at different intercepts depending on the relative abundance at each site in year 1.



3 Results

3.1 Survey coverage

Twenty-seven survey points / 1-km squares were surveyed for bats. This sample comprised 164 nights of recording, with a median of 5 nights of recording (range 2-16 visits) per site. After steps were taken to reduce error in species identification (see methods above), the revised dataset retained records for six bat species (including one species group, *Myotis*). Bat species were recorded on between 9 (33%) 1-km squares for lesser horseshoe bat, to 27 (100%) of 1-km squares for common pipistrelle (Table 4). Across species, there was a large difference in the recorded activity, measured as the number of 1-minute sections in which the species was recorded, and ranging from a total of 21 lesser horseshoe bats calls to 10,987 common pipistrelle calls across the survey (Table 4).

Night protocol	Sleep protocol	Metric	Barbar	Myotis	Nycnoc	Pippip	Pippyg	Rhifer
	0	Sites	10	25	25	27	<u>26</u>	9
		Calls	33	422	1754	10987	4357	21
	1	Sites	8	22	24	27	26	6
Full	1	Calls	15	228	863	5494	2170	12
1 dii	4	Sites	2	19	20	27	24	3
		Calls	2	90	330	2222	897	3
	9	Sites	1	16	20	27	23	3
	•	Calls	1	51	166	1107	432	3
	0	Sites	5	19	25	27	24	5
	0	Calls	13	102	958	3859	1663	11
	4	Sites	4	16	23	27	24	2
Duale & Davia	1	Calls	7	45	456	1927	831	4
Dusk & Dawn	4	Sites	1	13	20	27	21	1
	4	Calls	1	25	180	785	342	1
	0	Sites	0	8	20	26	18	1
	9	Calls	0	10	90	389	154	1
	0	Sites	7	23	25	27	26	6
	0	Calls	17	226	1368	6757	2654	16
	4	Sites	6	22	23	27	26	3
Durah	1	Calls	9	118	652	3375	1316	8
Dusk	4	Sites	1	15	20	27	24	2
	4	Calls	1	46	269	1387	547	2
	9	Sites	0	13	20	27	21	2
	Э	Calls	0	27	134	686	259	2

 Table 4. Sample sizes (number of 'occupied' sites) and total number of bat calls (summed across sites and visits) for each species under different sampling protocols. See Table 2 for species codes.

If we look at the number of sites with a non-zero count under the different sampling protocols, even without imposing a decline, the number of occupied sites is zero or much reduced under certain protocols for some species. For some species and protocols there were fewer than ten individual bat calls over less than ten sites, which we think is below the level at which a robust analysis can be carried out (Figure 3).

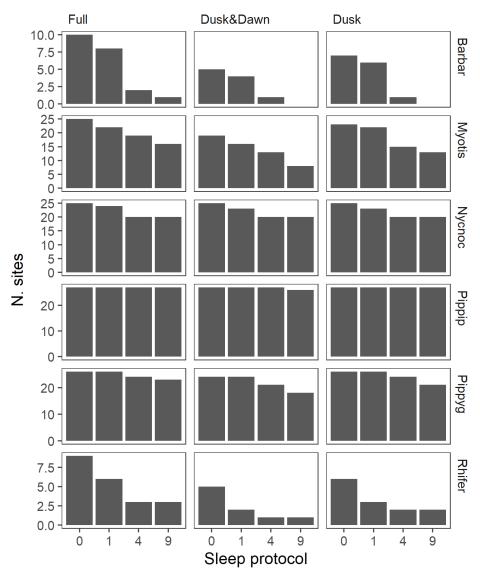


Figure 3. Number of sites with non-zero count under the different sampling protocols. See Table 2 for species codes.

Similarly, we can look at the total number of bat calls, summed across sites and all visits under the different sampling protocols. Even without imposing a decline, the number of calls is much reduced under certain protocols for some species. This further highlights the limitations of the pilot dataset, and how some protocols cannot be tested for some species (Figure 4).

For the reasons above, the current pilot dataset was only able to support a consideration of the sampling protocols for the three most widespread and abundant bat species: noctule, soprano pipistrelle and common pipistrelle.

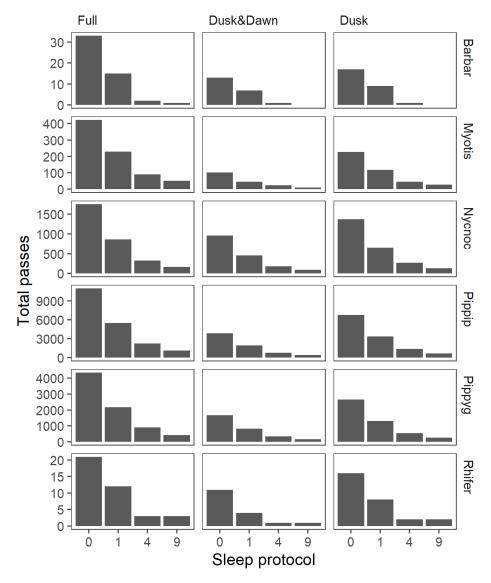


Figure 4. Total number of bat calls summed across sites and all visits under the different sampling protocols. See Table 2 for species codes.

3.2 Influence of sampling protocol on number of sites

Recording continually, i.e. with the greatest sampling effort, would result in a dataset with bat species recorded on the maximum number of sites. If a sampling protocol is used, that in some way produces a sub-sample of the whole dataset, the number of sites on which a species is recorded may be reduced (see top panel Figures 5–7 and Appendix 1, Table S1).

For common pipistrelle, which was recorded with a high level of activity across all surveyed sites, reducing the data by applying the sampling protocols here, had very little influence on the number of sites that would be available for analysis.

For soprano pipistrelle, which was recorded on about 88% of sites, applying the more extreme sampling protocol of recording for one minute in every ten, would result in a small reduction in the proportion of sites recording the species to about 85%.

For noctule, which was recorded on about 92% of sites surveyed, applying the more extreme sampling protocol of recordings for one minute in every ten, would result in a larger reduction in the proportion of sites recording the species, to about 74% of sites.

We expect that for other bat species recorded at lower levels of activity, applying the same sampling protocol would result in significantly fewer sites being retained and available for analysis.

3.3 Influence of sampling protocol on the accuracy of trend estimates

In section 3.2, we have seen that increasing the on/off cycle to have longer sleep durations between recording, reduces the number of sites available for analyses. A consequence of this is that it reduces the precision of the trend estimates, with wider confidence limits around the estimates. This is illustrated in the middle panel of Figures 5–7 (and Appendix 1, Table S2).

Reduced precision is to be expected, but perhaps a more important and nuanced finding, is that the trend estimates are biased for some permutations, with population declines being strongly underestimated. For example, with longer sleep periods of 4 and 9 seconds, there are few permutations of sample size and imposed declines where the analysis would not generate biased underestimates of population decline (middle panel of Figures 5–7 & Appendix 1, Table S2). This becomes a bigger problem for some species, if recording is not carried out over the whole night, but the extent of the problem is species-specific. For common pipistrelle, a dusk only survey is likely to result in a biased underestimate of trend regardless of sample size or imposed decline. For noctule, trend estimates are likely to be under-estimated, unless recording is carried out across the whole night. To explain this pattern, for species where activity is low, or low over some sites, when you impose a small decline, the species can become extinct at a site. If you are degrading some of the sites by imposing a protocol, sites can go to zero potentially in the first year, after which, the number of sites then cannot get lower in subsequent years with a continued decline imposed.

3.4 Influence of sampling protocol on survey power

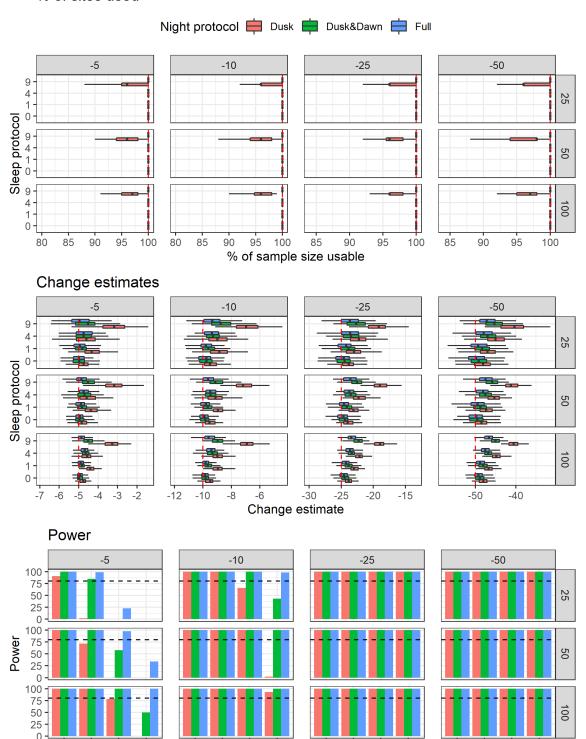
In the above, we have shown how the sampling protocol influences the number of sites available for analysis, and that the sampling protocol used can result in biased underestimates of decline. In this section, we look at the relationship between survey protocol, number of sites surveyed, and power to detect specified levels of decline over a 25-year period (bottom panel of Figures 5–7, see also Appendix 1, Table S3).

Importantly these analyses look at survey power in relation to the different sampling protocols for the three most commonly recorded bat species only, for which the power to detect a specified level of decline is likely to be greatest. The power to detect the same level of decline will be less for other bat species not considered here.

Keeping this in mind, it should be possible to detect a 25% decline (with 80% power or more) over 25 years for all three species given the assumed variances used to generate the trends. Whilst the analysis suggests that it should be possible to achieve this level of power, even given a fairly modest number of sites being surveyed, it would be important to ensure that a sufficiently large number of sites were surveyed to provide trends that are representative at the regional or national scale of interest. Detecting small declines of 10% for noctule or even 5% declines for common and soprano pipistrelle should be possible over a 25-year period, but the survey power is heavily influenced by the sampling protocol used, and dependent on the number of sites surveyed.

Importantly, whilst in theory there is the power to detect these specified levels of decline, as considered above, the sampled protocol used can result in biased underestimates of decline.

Figure 5. How the choice of sampling protocol influences the number of sites recording common pipistrelle, (top panel), the accuracy of trend estimates (middle panel), and the power to detect a specified level of species decline, where the black dotted line highlights 80% power (bottom panel). The variance in trend between sites was set to be one quarter of the mean. The red dotted line is the reference or true value. Each protocol is shown for 25, 50 and 100 sites.

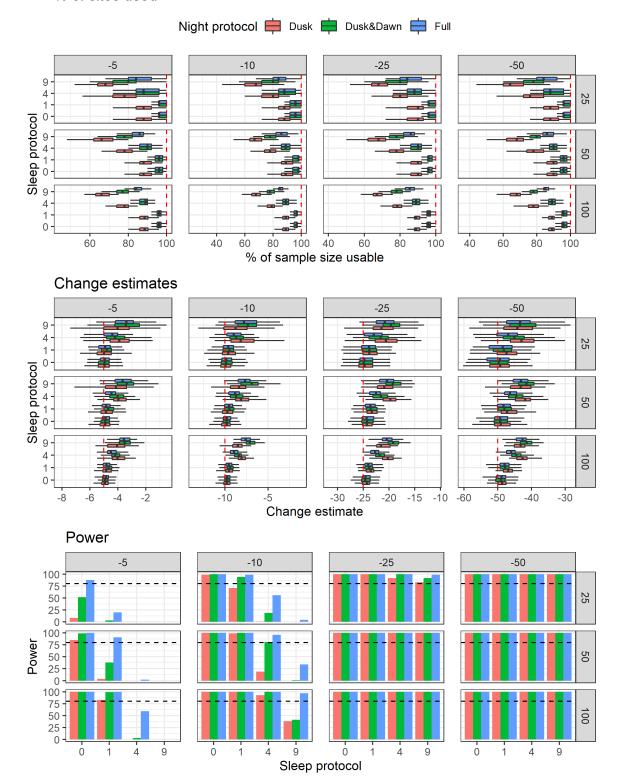


% of sites used

0 1

Sleep protocol

Figure 6. How the choice of sampling protocol influences the number of sites recording soprano pipistrelle, (top panel), the accuracy of trend estimates (middle panel), and the power to detect a specified level of species decline, where the black dotted line highlights 80% power (bottom panel). The variance in trend between sites was set to be one quarter of the mean. The red dotted line is the reference or true value. Each protocol is shown for 25, 50 and 100 sites.

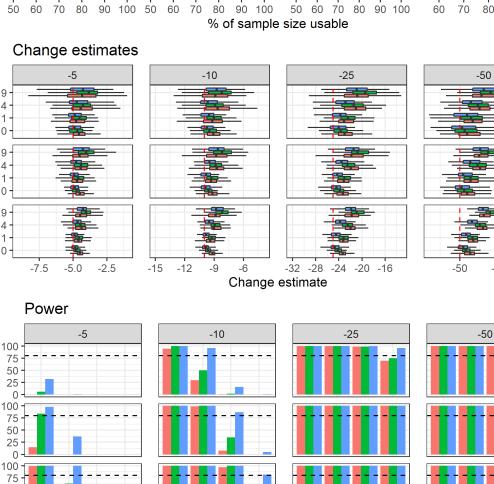


% of sites used

15

Figure 7. How the choice of sampling protocol influences the number of sites recording noctule, (top panel), the accuracy of trend estimates (middle panel), and the power to detect a specified level of species decline, where the black dotted line highlights 80% power (bottom panel). The variance in trend between sites was set to be one quarter of the mean. The red dotted line is the reference or true value. Each protocol is shown for 25, 50 and 100 sites.

Night protocol 🚔 Dusk 🚔 Dusk&Dawn 🚔 Full -5 -10 -25 -50 9 = 4 25 1 0 Sleep protocol 9 4 50 1 0 **1** 9 _____ ____ 4 100 1 0 -8 90 100 50 60 70 80 90 100 50 60 70 80 90 100 100 60 70 60 70 90 50 80 80 % of sample size usable Change estimates -5 -10 -25 -50 9 4 25 1 5 5-0 Sleep protocol 9 4 50 1 0 9 4 100 1 0 -7.5 -5.0 -2.5 -15 -12 -32 -28 -24 -20 -16 -50 -40 -30 -9 -6 Change estimate Power



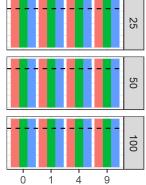
9 0 Sleep protocol 1

4

9

4

1



16

% of sites used

Power

50 25 0

0

1

4

9

0

4 **Discussion**

In total only 27 sites were surveyed, with 6 bat species (including one species group, *Myotis*) recorded on between 9 and 27 sites. If on/off or part-night sampling is used, this degrades the dataset, which for most bat species reduces the sample of sites further. For this reason, it has been necessary to restrict our analysis to the three most widely recorded bat species. For more information on the limitations and constraints of the protocols and bat data see section 2.5.

The survey area in the south-west was chosen to include the geographic range of three target bat species, Leisler's bat, barbastelle and Nathusius' pipistrelle, that are not currently monitored by the National Bat Monitoring Programme (NBMP), and so are of particular interest to BCT. Unfortunately, the limitations of the pilot data set meant that it was not possible to consider these species in the analysis here, although we think that some general recommendations relevant to these species can still be made.

With on/off sampling, increasing the sleep time between recording reduces the precision of trend estimates, resulting in wider confidence limits of trend estimates, and reduces the power to detect a specified level of decline. However, an important finding is that with longer sleep periods of 4 and 9 minutes, there are few permutations of sample size and imposed declines where the analysis would not generate biased underestimates of population decline. Whilst problems associated with a shorter sleep period (recording 1 in every 2 minutes), appear to be quite modest in many cases, whether a record / sleep cycle is a good option for the British Bat Survey, would depend on the gains in processing time, reduced storage costs, and whether there are additional costs or considerations in relation to battery life and on-board (SD card) storage. Unless these are significant, this is unlikely to be a useful approach, particularly as the survey effort for a volunteer to deploy an AudioMoth to record continuously, or according to a record / sleep cycle is the same.

Because bat activity is greatest during the first hours after sunset (Collins 2016), manual acoustic sampling of bats is most commonly conducted at dusk only, or at dusk and dawn. With part night sampling, the assumption is that this would record most bat activity, but with less effort than would be required if recording for the whole night. When deploying static detectors, which are left outside to automatically trigger and record bats, there is no difference in effort for the fieldworker who leaves out a detector for part of the night rather than a whole night, but there are additional costs of recording over a whole night in relation to time needed to process recordings, and the cost of storing a larger volume of large *.wav files.

We demonstrate here that part night sampling can result in biased underestimates of population decline, although the severity of the problem is likely to be species-specific. For common pipistrelle, a dusk only survey is likely to result in a biased underestimate of trend regardless of sample size or imposed decline. For noctule, trend estimates are likely to be under-estimated, unless recording is carried out across the whole night. With these findings, we would advise against a part-night sampling protocol. The recommendation for whole night recording is supported by Froidevaux *et al.* (2014) and Skalak *et al.* (2012), who showed that sampling the full night was essential to cover the bimodal peaks of bat activity, and have also shown that it is likely to be important to be able to detect rare species with low detection probabilities.

Whilst there was not a viable option at the time of writing to consider a 'trigger' for the AudioMoth as an alternative approach for reducing the volume of recordings, i.e. to only record the events that are of interest by recording sounds only above a defined frequency and amplitude, this is another option which should be considered if viable.

As would be expected, the more sites surveyed, the greater the power that there would be to detect a specified level of population decline. In this study we look at the power to detect specified declines considering a sample size of 25, 50 and 100 sites. In these analyses, we were only able to consider three bat species, for which the power to detect a specified level of decline is likely to be greatest. It is likely that a much larger number of sites would need to be surveyed for other, more localised and rare species, to achieve a comparable level of power. Whilst it is important to have sufficient power (in theory) to detect a specified level of decline, considering power alone is not useful if the sampling protocol results in biased estimates of decline.

For any national scheme, it would be important to stratify by region or habitat to maximise the number of sites that record the most range restricted species, in order to increase the probability of being able to produce trends for these. When thinking about the number of sites needed for a national scheme, it is important to have in mind a level of population decline or increase that you want to be able to detect.

4.1 **Recommendations**

Adopting effective survey and sampling protocols are essential for producing robust population trends for bats. Although limited to three bat species, our study constitutes an important analysis for informing the design of national bat monitoring. We propose the following recommendations: (1) record over the full night to avoid under-recording species that are active over much of the night; (2) recording according to a record / sleep cycle could be used if the sleep time is short (recording for 1 minute in every 2 minutes), but unless there are significant gains in doing this (e.g. reduced cost / time to process data) to compensate for the reductions in accuracy, precision and power to detect declines, this is not recommended; (3) a more extensive dataset is collected to revisit the analyses here for a broader suite of species; (4) it is likely that a national scheme would need to record target bat species on more than 100 sites a year, but the sample size should be determined by the power to detect specified declines for the scarcest species, which we were not able to look at here.

5 Author contributions

SEN led on the writing and interpretation of the analysis. SG led on the analysis and R code to support this, with input at all stages from RR. KB coordinated BCT volunteers to collect the bat data, processed the bat data to identify species, quantified uncertainty in species identification, and provided guidance on the use of the data. All four co-authors helped to shape the ideas, context and analysis of the project and read and commented on draft versions of the manuscript.

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Appendix 1

Table S1. How the choice of sampling protocol influences the number of sites recording the species, for a) common pipistrelle, b) soprano pipistrelle, and c) noctule. The variance in trends between sites was set to be one quarter of the mean.

a) common pi	pistrelle
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		Imposed level of species decline				
Night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	25	25	25	25
Dusk	0	50	50	50	50	50
Dusk	0	100	100	100	100	100
Dusk&Dawn	0	25	25	25	25	25
Dusk&Dawn	0	50	50	50	50	50
Dusk&Dawn	0	100	100	100	100	100
Full	0	25	25	25	25	25
Full	0	50	50	50	50	50
Full	0	100	100	100	100	100
Dusk	1	25	25	25	25	25
Dusk	1	50	50	50	50	50
Dusk	1	100	100	100	100	100
Dusk&Dawn	1	25	25	25	25	25
Dusk&Dawn	1	50	50	50	50	50
Dusk&Dawn	1	100	100	100	100	100
Full	1	25	25	25	25	25
Full	1	50	50	50	50	50
Full	1	100	100	100	100	100
Dusk	4	25	25	25	25	25
Dusk	4	50	50	50	50	50
Dusk	4	100	100	100	100	100
Dusk&Dawn	4	25	25	25	25	25
Dusk&Dawn	4	50	50	50	50	50
Dusk&Dawn	4	100	100	100	100	100
Full	4	25	25	25	25	25
Full	4	50	50	50	50	50
Full	4	100	100	100	100	100
Dusk	9	25	24.04	24.04	24.06	24.25
Dusk	9	50	48.12	48.22	48.08	48.17
Dusk	9	100	96.39	95.98	96.52	96.34
Dusk&Dawn	9	25	25	25	25	25
Dusk&Dawn	9	50	50	50	50	50
Dusk&Dawn	9	100	100	100	100	100
Full	9	25	25	25	25	25
Full	9	50	50	50	50	50
Full	9	100	100	100	100	100

b) soprano pipistrelle

			Impos	sed level of s	pecies declir	ie
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	22.13	22.19	22.36	22.24
Dusk	0	50	44.33	44.56	44.35	44.56
Dusk	0	100	88.51	89.04	89.17	88.55
Dusk&Dawn	0	25	24.03	24.07	24.12	24.25
Dusk&Dawn	0	50	48.13	48.24	48.07	48.14
Dusk&Dawn	0	100	96.19	96.27	96.13	96.06
Full	0	25	24.03	24.07	24.12	24.25
Full	0	50	48.13	48.24	48.07	48.14
Full	0	100	96.19	96.27	96.13	96.06
Dusk	1	25	22.13	22.19	22.36	22.24
Dusk	1	50	44.33	44.56	44.35	44.56
Dusk	1	100	88.51	89.04	89.17	88.55
Dusk&Dawn	1	25	24.03	24.07	24.12	24.25
Dusk&Dawn	1	50	48.13	48.24	48.07	48.14
Dusk&Dawn	1	100	96.19	96.27	96.13	96.06
Full	1	25	24.03	24.07	24.12	24.25
Full	1	50	48.13	48.24	48.07	48.14
Full	1	100	96.19	96.27	96.13	96.06
Dusk	4	25	19.28	19.46	19.67	19.46
Dusk	4	50	38.88	38.81	38.93	38.99
Dusk	4	100	77.33	78.35	78.13	77.62
Dusk&Dawn	4	25	22.37	22.19	22.25	22.32
Dusk&Dawn	4	50	44.43	44.63	44.54	44.7
Dusk&Dawn	4	100	88.37	89.22	89.04	88.56
Full	4	25	22.37	22.19	22.25	22.32
Full	4	50	44.43	44.63	44.54	44.7
Full	4	100	88.37	89.22	89.04	88.56
Dusk	9	25	16.66	16.58	16.94	16.36
Dusk	9	50	33.42	33.39	33.46	33.41
Dusk	9	100	65.92	67.54	67.18	67.2
Dusk&Dawn	9	25	19.53	19.36	19.72	19.24
Dusk&Dawn	9	50	39.01	39.26	38.79	39.24
Dusk&Dawn	9	100	77.04	78.19	78.44	78.08
Full	9	25	21.41	21.08	21.43	21.24
Full	9	50	42.64	42.86	42.76	42.84
Full	9	100	84.58	85.57	85.47	85.24

c) Noctule

			Impos	sed level of s	pecies declin	е
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	22.92	23.12	23.2	23.06
Dusk	0	50	46.26	46.22	46.24	46.06
Dusk	0	100	92.81	92.55	92.4	92.32
Dusk&Dawn	0	25	22.92	23.12	23.2	23.06
Dusk&Dawn	0	50	46.26	46.22	46.24	46.06
Dusk&Dawn	0	100	92.81	92.55	92.4	92.32
Full	0	25	22.92	23.12	23.2	23.06
Full	0	50	46.26	46.22	46.24	46.06
Full	0	100	92.81	92.55	92.4	92.32
Dusk	1	25	21.19	21.1	21.25	21.31
Dusk	1	50	42.49	42.78	42.63	42.28
Dusk	1	100	85.19	85.3	85.27	84.75
Dusk&Dawn	1	25	21.19	21.1	21.25	21.31
Dusk&Dawn	1	50	42.49	42.78	42.63	42.28
Dusk&Dawn	1	100	85.19	85.3	85.27	84.75
Full	1	25	22.12	22.07	22.14	22.12
Full	1	50	44.28	44.34	44.5	44.22
Full	1	100	88.96	88.9	88.79	88.71
Dusk	4	25	18.34	18.23	18.61	18.73
Dusk	4	50	36.82	37.18	37.06	36.75
Dusk	4	100	73.86	73.68	74.32	73.98
Dusk&Dawn	4	25	18.34	18.23	18.61	18.73
Dusk&Dawn	4	50	36.82	37.18	37.06	36.75
Dusk&Dawn	4	100	73.86	73.68	74.32	73.98
Full	4	25	18.34	18.23	18.61	18.73
Full	4	50	36.82	37.18	37.06	36.75
Full	4	100	73.86	73.68	74.32	73.98
Dusk	9	25	18.34	18.23	18.61	18.73
Dusk	9	50	36.82	37.18	37.06	36.75
Dusk	9	100	73.86	73.68	74.32	73.98
Dusk&Dawn	9	25	18.34	18.23	18.61	18.73
Dusk&Dawn	9	50	36.82	37.18	37.06	36.75
Dusk&Dawn	9	100	73.86	73.68	74.32	73.98
Full	9	25	18.34	18.23	18.61	18.73
Full	9	50	36.82	37.18	37.06	36.75
Full	9	100	73.86	73.68	74.32	73.98

Table S2. How the choice of sampling protocol influences the ability to detect an imposed specified level of species decline, for a) common pipistrelle, b) soprano pipistrelle, and c) noctule. The variance in trends between sites was set to be one quarter of the mean.

a) common pipistrelle

			Imposed level of species decline			
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	-4.76953	-9.43095	-23.928	-48.1928
Dusk	0	50	-4.77948	-9.50996	-23.9031	-48.649
Dusk	0	100	-4.77935	-9.55246	-23.8543	-48.043
Dusk&Dawn	0	25	-4.97551	-9.77307	-24.6067	-49.2394
Dusk&Dawn	0	50	-4.95051	-9.88817	-24.5453	-49.5877
Dusk&Dawn	0	100	-4.94788	-9.86582	-24.431	-48.9296
Full	0	25	-4.9992	-9.8165	-24.7902	-49.6783
Full	0	50	-4.96053	-9.90849	-24.7228	-49.9216
Full	0	100	-4.93718	-9.87967	-24.6107	-49.2883
Dusk	1	25	-4.33507	-8.83366	-23.0439	-47.0662
Dusk	1	50	-4.37066	-8.94315	-23.0532	-47.4028
Dusk	1	100	-4.39901	-8.95994	-22.9498	-46.7827
Dusk&Dawn	1	25	-4.86428	-9.55211	-24.06	-48.4774
Dusk&Dawn	1	50	-4.82157	-9.63872	-24.0937	-48.871
Dusk&Dawn	1	100	-4.83909	-9.63797	-23.8878	-48.1802
Full	1	25	-4.9551	-9.77036	-24.5359	-49.2723
Full	1	50	-4.9085	-9.81721	-24.4763	-49.5179
Full	1	100	-4.89525	-9.81031	-24.3569	-48.9007
Dusk	4	25	-4.64203	-8.93841	-22.2733	-44.9157
Dusk	4	50	-4.54803	-9.12107	-22.1638	-45.4545
Dusk	4	100	-4.61317	-8.97574	-22.2321	-44.798
Dusk&Dawn	4	25	-4.72346	-9.26709	-23.429	-46.9956
Dusk&Dawn	4	50	-4.71152	-9.48664	-23.4991	-47.5479
Dusk&Dawn	4	100	-4.65861	-9.36474	-23.3925	-46.8533
Full	4	25	-4.7237	-9.32991	-23.7713	-47.951
Full	4	50	-4.75778	-9.47661	-23.724	-48.3389
Full	4	100	-4.70729	-9.429	-23.654	-47.6421
Dusk	9	25	-3.20283	-6.92799	-19.2066	-40.5681
Dusk	9	50	-3.19674	-7.09821	-18.9293	-41.1899
Dusk	9	100	-3.3066	-6.90858	-19.008	-40.5686
Dusk&Dawn	9	25	-4.62343	-8.77918	-22.522	-45.358
Dusk&Dawn	9	50	-4.50565	-9.07553	-22.4863	-45.882
Dusk&Dawn	9	100	-4.53211	-9.0163	-22.3685	-45.1116
Full	9	25	-4.89701	-9.36237	-23.6468	-47.1223
Full	9	50	-4.86089	-9.61035	-23.4867	-47.4446
Full	9	100	-4.86166	-9.56621	-23.3997	-46.7724

b) soprano pipistrelle

			Imp	osed level of	species decl	line
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	-4.95511	-9.87789	-24.6784	-49.4474
Dusk	0	50	-4.92159	-9.81508	-24.1922	-48.6607
Dusk	0	100	-4.90085	-9.75292	-24.4163	-48.7346
Dusk&Dawn	0	25	-4.96228	-9.85319	-24.6737	-49.8054
Dusk&Dawn	0	50	-4.87505	-9.8283	-24.127	-49.165
Dusk&Dawn	0	100	-4.89289	-9.71965	-24.4798	-49.1271
Full	0	25	-4.93763	-9.77656	-24.6313	-49.7859
Full	0	50	-4.86466	-9.76501	-24.1285	-49.1269
Full	0	100	-4.86124	-9.72371	-24.4423	-48.9561
Dusk	1	25	-4.97954	-9.64809	-23.6938	-47.5752
Dusk	1	50	-4.85055	-9.65175	-23.3388	-46.8351
Dusk	1	100	-4.82127	-9.50815	-23.5835	-46.8679
Dusk&Dawn	1	25	-4.82472	-9.56618	-23.8859	-48.5513
Dusk&Dawn	1	50	-4.62977	-9.46613	-23.4737	-47.9496
Dusk&Dawn	1	100	-4.64295	-9.37499	-23.772	-47.8716
Full	1	25	-4.94614	-9.62185	-24.0369	-48.8132
Full	1	50	-4.84124	-9.58984	-23.6687	-48.2357
Full	1	100	-4.81425	-9.54816	-23.9481	-48.0294
Dusk	4	25	-3.96907	-7.96015	-20.4669	-42.8628
Dusk	4	50	-3.88545	-7.95846	-19.8591	-42.2372
Dusk	4	100	-3.96629	-8.02841	-20.2459	-42.5764
Dusk&Dawn	4	25	-4.21517	-8.6398	-21.7144	-45.1396
Dusk&Dawn	4	50	-4.07878	-8.62525	-21.6255	-44.3941
Dusk&Dawn	4	100	-4.14699	-8.47509	-21.7348	-44.4991
Full	4	25	-4.48197	-8.97351	-22.9696	-46.8483
Full	4	50	-4.47708	-8.92014	-22.5207	-46.234
Full	4	100	-4.43936	-8.9066	-22.7325	-46.0768
Dusk	9	25	-4.17373	-8.68052	-21.2195	-44.0155
Dusk	9	50	-4.16016	-8.38323	-20.821	-43.1028
Dusk	9	100	-4.15237	-8.52085	-21.1979	-43.2895
Dusk&Dawn	9	25	-3.43351	-7.20891	-19.3637	-42.5538
Dusk&Dawn	9	50	-3.36959	-7.0571	-19.209	-41.5366
Dusk&Dawn	9	100	-3.49168	-7.08849	-19.1758	-41.5862
Full	9	25	-3.51912	-7.60566	-20.5216	-43.5578
Full	9	50	-3.61719	-7.7099	-20.2793	-43.1497
Full	9	100	-3.57222	-7.56738	-20.378	-42.9502

c) noctule

			Imp	osed level of	species decl	ine
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	-4.51197	-9.10124	-22.9282	-47.4791
Dusk	0	50	-4.50152	-9.15161	-23.3525	-47.8793
Dusk	0	100	-4.50534	-9.20742	-23.3915	-47.4046
Dusk&Dawn	0	25	-4.76198	-9.46559	-23.7644	-48.3019
Dusk&Dawn	0	50	-4.77599	-9.6095	-23.9466	-48.7234
Dusk&Dawn	0	100	-4.7506	-9.58732	-24.13	-48.3263
Full	0	25	-4.87422	-9.73508	-24.1657	-48.8854
Full	0	50	-4.89514	-9.88292	-24.4613	-49.3955
Full	0	100	-4.88027	-9.84305	-24.6827	-49.0803
Dusk	1	25	-4.70379	-9.21761	-22.6697	-46.4982
Dusk	1	50	-4.67525	-9.27773	-23.2247	-47.0476
Dusk	1	100	-4.65121	-9.34711	-23.2452	-46.5544
Dusk&Dawn	1	25	-4.61038	-9.00954	-22.5196	-46.7028
Dusk&Dawn	1	50	-4.62952	-9.24841	-23.0129	-47.1587
Dusk&Dawn	1	100	-4.57773	-9.19794	-23.1995	-46.7652
Full	1	25	-4.90425	-9.65594	-23.8532	-48.1319
Full	1	50	-4.90171	-9.86123	-24.2263	-48.723
Full	1	100	-4.90669	-9.80017	-24.4833	-48.4153
Dusk	4	25	-4.34253	-8.73982	-21.221	-44.598
Dusk	4	50	-4.42169	-8.71109	-21.8732	-44.9828
Dusk	4	100	-4.39032	-8.86366	-22.0483	-44.1843
Dusk&Dawn	4	25	-4.34557	-8.93412	-21.6886	-45.0708
Dusk&Dawn	4	50	-4.43134	-8.80841	-22.3515	-45.3081
Dusk&Dawn	4	100	-4.48644	-8.94086	-22.5282	-44.8093
Full	4	25	-4.66731	-9.61358	-22.9791	-46.6577
Full	4	50	-4.76935	-9.40185	-23.5724	-47.0923
Full	4	100	-4.74855	-9.50941	-23.7199	-46.8397
Dusk	9	25	-4.41956	-9.03178	-21.0648	-43.8102
Dusk	9	50	-4.43025	-8.73706	-21.4662	-44.6081
Dusk	9	100	-4.42183	-8.98444	-21.8855	-43.7619
Dusk&Dawn	9	25	-3.9249	-8.41807	-19.6042	-41.9052
Dusk&Dawn	9	50	-4.026	-8.17562	-20.2991	-42.6789
Dusk&Dawn	9	100	-4.05158	-8.17068	-20.8916	-42.077
Full	9	25	-4.36592	-8.85902	-21.0005	-43.712
Full	9	50	-4.39811	-8.71322	-21.6439	-44.6429
Full	9	100	-4.33491	-8.68918	-22.0615	-44.1864

Table S3. How the choice of sampling protocol influences the power to detect a specified level of species decline, for a) common pipistrelle, b) soprano pipistrelle, and c) noctule. In the following analyses. The variance in trends between sites was set to be one quarter of the mean.

a) common pipistrelle

			Imposed level of species decline				
night	sleep protocol	number of sites	-5	-10	-25	-50	
Dusk	0	25	91	100	100	100	
Dusk	0	50	100	100	100	100	
Dusk	0	100	100	100	100	100	
Dusk&Dawn	0	25	100	100	100	100	
Dusk&Dawn	0	50	100	100	100	100	
Dusk&Dawn	0	100	100	100	100	100	
Full	0	25	100	100	100	100	
Full	0	50	100	100	100	100	
Full	0	100	100	100	100	100	
Dusk	1	25	2	100	100	100	
Dusk	1	50	72	100	100	100	
Dusk	1	100	100	100	100	100	
Dusk&Dawn	1	25	85	100	100	100	
Dusk&Dawn	1	50	100	100	100	100	
Dusk&Dawn	1	100	100	100	100	100	
Full	1	25	99	100	100	100	
Full	1	50	100	100	100	100	
Full	1	100	100	100	100	100	
Dusk	4	25	0	66	100	100	
Dusk	4	50	0	100	100	100	
Dusk	4	100	78	100	100	100	
Dusk&Dawn	4	25	0	100	100	100	
Dusk&Dawn	4	50	58	100	100	100	
Dusk&Dawn	4	100	100	100	100	100	
Full	4	25	23	100	100	100	
Full	4	50	98	100	100	100	
Full	4	100	100	100	100	100	
Dusk	9	25	0	0	100	100	
Dusk	9	50	0	2	100	100	
Dusk	9	100	0	93	100	100	
Dusk&Dawn	9	25	0	43	100	100	
Dusk&Dawn	9	50	0	100	100	100	
Dusk&Dawn	9	100	50	100	100	100	
Full	9	25	0	98	100	100	
Full	9	50	34	100	100	100	
Full	9	100	100	100	100	100	

b) soprano pipistrelle

			Imposed level of species decline			line
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	8	99	100	100
Dusk	0	50	85	100	100	100
Dusk	0	100	100	100	100	100
Dusk&Dawn	0	25	52	100	100	100
Dusk&Dawn	0	50	99	100	100	100
Dusk&Dawn	0	100	100	100	100	100
Full	0	25	88	100	100	100
Full	0	50	100	100	100	100
Full	0	100	100	100	100	100
Dusk	1	25	0	71	100	100
Dusk	1	50	4	99	100	100
Dusk	1	100	83	100	100	100
Dusk&Dawn	1	25	3	94	100	100
Dusk&Dawn	1	50	38	100	100	100
Dusk&Dawn	1	100	99	100	100	100
Full	1	25	20	99	100	100
Full	1	50	91	100	100	100
Full	1	100	100	100	100	100
Dusk	4	25	0	1	92	100
Dusk	4	50	0	19	100	100
Dusk	4	100	0	93	100	100
Dusk&Dawn	4	25	0	19	100	100
Dusk&Dawn	4	50	0	81	100	100
Dusk&Dawn	4	100	3	100	100	100
Full	4	25	0	56	100	100
Full	4	50	2	96	100	100
Full	4	100	59	100	100	100
Dusk	9	25	0	0	83	100
Dusk	9	50	0	0	100	100
Dusk	9	100	0	38	100	100
Dusk&Dawn	9	25	0	0	92	100
Dusk&Dawn	9	50	0	1	100	100
Dusk&Dawn	9	100	0	41	100	100
Full	9	25	0	4	99	100
Full	9	50	0	34	100	100
Full	9	100	0	97	100	100

c) noctule

			Imposed level of species decline			
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	0	95	100	100
Dusk	0	50	15	100	100	100
Dusk	0	100	99	100	100	100
Dusk&Dawn	0	25	6	100	100	100
Dusk&Dawn	0	50	84	100	100	100
Dusk&Dawn	0	100	100	100	100	100
Full	0	25	32	100	100	100
Full	0	50	98	100	100	100
Full	0	100	100	100	100	100
Dusk	1	25	0	30	100	100
Dusk	1	50	0	99	100	100
Dusk	1	100	21	100	100	100
Dusk&Dawn	1	25	0	50	100	100
Dusk&Dawn	1	50	0	100	100	100
Dusk&Dawn	1	100	63	100	100	100
Full	1	25	1	96	100	100
Full	1	50	37	100	100	100
Full	1	100	100	100	100	100
Dusk	4	25	0	1	99	100
Dusk	4	50	0	8	100	100
Dusk	4	100	0	97	100	100
Dusk&Dawn	4	25	0	2	99	100
Dusk&Dawn	4	50	0	35	100	100
Dusk&Dawn	4	100	0	100	100	100
Full	4	25	0	16	100	100
Full	4	50	0	87	100	100
Full	4	100	4	100	100	100
Dusk	9	25	0	0	70	100
Dusk	9	50	0	0	100	100
Dusk	9	100	0	28	100	100
Dusk&Dawn	9	25	0	0	75	100
Dusk&Dawn	9	50	0	0	100	100
Dusk&Dawn	9	100	0	32	100	100
Full	9	25	0	1	96	100
Full	9	50	0	5	100	100
Full	9	100	0	82	100	100

Table S4. How the choice of sampling protocol influences the power to detect a specified level of species decline, for a) common pipistrelle, b) soprano pipistrelle, and c) noctule. In the following analyses. The variance in trends between sites was set to be one half of the mean (double the above variance).

	sleep protocol		Imposed level of species decline			
night		number of sites	-5	-10	-25	-50
Dusk	0	25	88	100	100	100
Dusk	0	50	100	100	100	100
Dusk	0	100	100	100	100	100
Dusk&Dawn	0	25	100	100	100	100
Dusk&Dawn	0	50	100	100	100	100
Dusk&Dawn	0	100	100	100	100	100
Full	0	25	100	100	100	100
Full	0	50	100	100	100	100
Full	0	100	100	100	100	100
Dusk	1	25	4	100	100	100
Dusk	1	50	74	100	100	100
Dusk	1	100	100	100	100	100
Dusk&Dawn	1	25	81	100	100	100
Dusk&Dawn	1	50	100	100	100	100
Dusk&Dawn	1	100	100	100	100	100
Full	1	25	99	100	100	100
Full	1	50	100	100	100	100
Full	1	100	100	100	100	100
Dusk	4	25	0	70	100	100
Dusk	4	50	1	100	100	100
Dusk	4	100	66	100	100	100
Dusk&Dawn	4	25	1	99	100	100
Dusk&Dawn	4	50	53	100	100	100
Dusk&Dawn	4	100	100	100	100	100
Full	4	25	31	100	100	100
Full	4	50	95	100	100	100
Full	4	100	100	100	100	100
Dusk	9	25	0	0	99	100
Dusk	9	50	0	10	100	100
Dusk	9	100	0	92	100	100
Dusk&Dawn	9	25	0	57	100	100
Dusk&Dawn	9	50	0	100	100	100
Dusk&Dawn	9	100	41	100	100	100
Full	9	25	4	99	100	100
Full	9	50	39	100	100	100
Full	9	100	99	100	100	100

a) common pipistrelle

b) soprano pipistrelle

			Imposed level of species decline				
night	sleep protocol	number of sites	-5	-10	-25	-50	
Dusk	0	25	14	97	100	100	
Dusk	0	50	74	100	100	100	
Dusk	0	100	100	100	100	100	
Dusk&Dawn	0	25	52	100	100	100	
Dusk&Dawn	0	50	93	100	100	100	
Dusk&Dawn	0	100	100	100	100	100	
Full	0	25	79	100	100	100	
Full	0	50	99	100	100	100	
Full	0	100	100	100	100	100	
Dusk	1	25	0	64	100	100	
Dusk	1	50	5	99	100	100	
Dusk	1	100	80	100	100	100	
Dusk&Dawn	1	25	2	89	100	100	
Dusk&Dawn	1	50	39	100	100	100	
Dusk&Dawn	1	100	98	100	100	100	
Full	1	25	23	98	100	100	
Full	1	50	82	100	100	100	
Full	1	100	100	100	100	100	
Dusk	4	25	0	3	89	100	
Dusk	4	50	0	24	100	100	
Dusk	4	100	0	83	100	100	
Dusk&Dawn	4	25	0	15	99	100	
Dusk&Dawn	4	50	0	70	100	100	
Dusk&Dawn	4	100	2	99	100	100	
Full	4	25	0	58	100	100	
Full	4	50	3	98	100	100	
Full	4	100	50	100	100	100	
Dusk	9	25	0	0	67	99	
Dusk	9	50	0	2	99	100	
Dusk	9	100	0	35	100	100	
Dusk&Dawn	9	25	0	0	81	100	
Dusk&Dawn	9	50	0	2	99	100	
Dusk&Dawn	9	100	0	40	100	100	
Full	9	25	0	6	91	100	
Full	9	50	0	34	100	100	
Full	9	100	0	89	100	100	

c) noctule

			Imposed level of species decline			
night	sleep protocol	number of sites	-5	-10	-25	-50
Dusk	0	25	0	91	100	100
Dusk	0	50	20	100	100	100
Dusk	0	100	99	100	100	100
Dusk&Dawn	0	25	7	98	100	100
Dusk&Dawn	0	50	79	100	100	100
Dusk&Dawn	0	100	100	100	100	100
Full	0	25	32	100	100	100
Full	0	50	99	100	100	100
Full	0	100	100	100	100	100
Dusk	1	25	0	39	100	100
Dusk	1	50	0	97	100	100
Dusk	1	100	24	100	100	100
Dusk&Dawn	1	25	0	58	100	100
Dusk&Dawn	1	50	2	99	100	100
Dusk&Dawn	1	100	75	100	100	100
Full	1	25	0	92	100	100
Full	1	50	34	100	100	100
Full	1	100	99	100	100	100
Dusk	4	25	0	0	96	100
Dusk	4	50	0	12	100	100
Dusk	4	100	0	90	100	100
Dusk&Dawn	4	25	0	3	99	100
Dusk&Dawn	4	50	0	45	100	100
Dusk&Dawn	4	100	0	99	100	100
Full	4	25	0	19	99	100
Full	4	50	0	85	100	100
Full	4	100	10	100	100	100
Dusk	9	25	0	0	73	100
Dusk	9	50	0	1	100	100
Dusk	9	100	0	40	100	100
Dusk&Dawn	9	25	0	0	79	100
Dusk&Dawn	9	50	0	1	100	100
Dusk&Dawn	9	100	0	42	100	100
Full	9	25	0	0	94	100
Full	9	50	0	14	100	100
Full	9	100	0	87	100	100