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Acoustic monitoring: new applications for bird and human wildlife conflicts monitoring

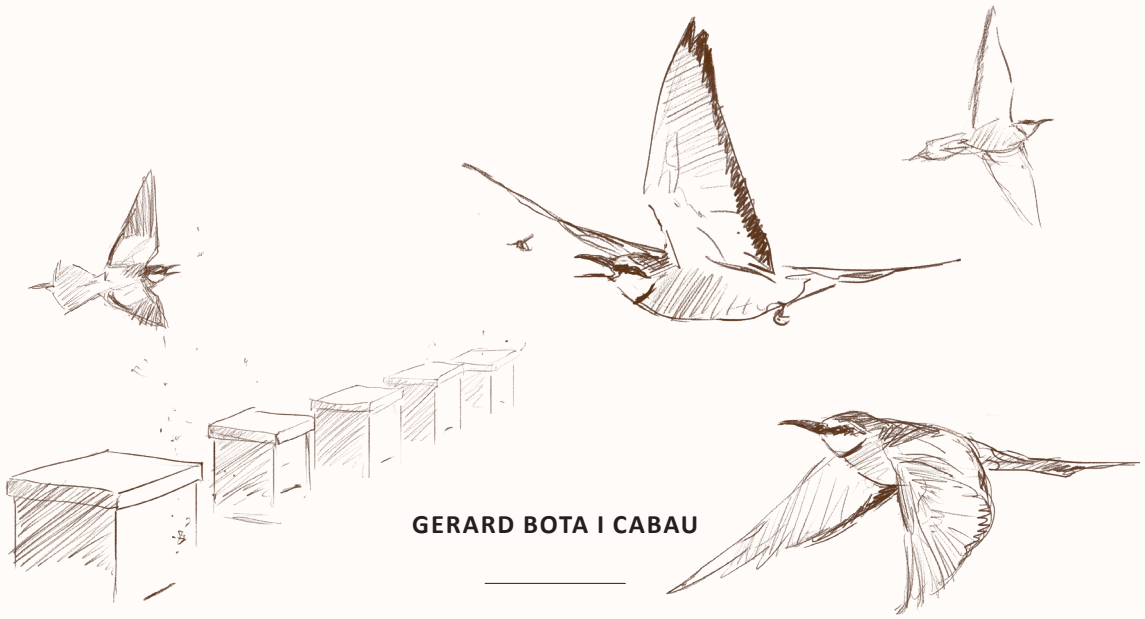
Gerard Bota Cabau



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GERARD BOTA I CABAU

ACOUSTIC MONITORING: NEW APPLICATIONS FOR BIRD AND HUMAN WILDLIFE CONFLICTS MONITORING

Monitoreig acústic:
noves aplicacions per al seguiment d'ocells
i de conflictes entre activitats humanes i fauna





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Programa de Doctorat en Biodiversitat- Universitat de Barcelona (UB)
Centre de Ciència i Tecnologia Forestal de Catalunya (CTFC)

Acoustic monitoring: new applications for bird and human wildlife conflicts monitoring

*Monitoreig acústic: noves aplicacions per al seguiment d'ocells i de conflictes
entre activitats humanes i fauna*

Memòria presentada per **Gerard Bota Cabau**
per optar al grau de Doctor per la Universitat de Barcelona

Gerard Bota i Cabau

Els directors de la tesi,

Cristian Pérez-Granados
(Universitat d'Alacant)

Juan Traba Díaz
(Universidad Autónoma de Madrid)

El Tutor de la tesi,

Santi Mañosa Rifé (Universtat de Barcelona)

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Aquesta tesi doctoral m'agafa ja una mica gran però no per això amb la sensació d'acabar content i satisfet pel camí recorregut. Des de molt al principi de la meua vida professional tenia molt clar que el que em motiva realment es treballar en la conservació de la biodiversitat i que la recerca n'és una eina, no un fi en si mateix. Fa temps que tinc molt clar que existeix un esvoranc massa gran entre la recerca sobre conservació i la conservació aplicada i per això sempre he volgut mantenir-me professionalment en aquesta interfase entre aquests dos mons. Ha calgut trobar el moment propici per fer-ho i espero que aquesta tesi i el doctorat que se'n deriva m'ajudi encara més a continuar teixint ponts entre aquest dos mons.

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RESUM

En un context de crisi global de pèrdua de la biodiversitat, com el que es troba el planeta, és essencial disposar d'informació precisa vinculada als sistemes de monitoreig de la biodiversitat per poder prendre decisions basades en l'evidència. El desenvolupament de noves tecnologies per a l'estudi de la biodiversitat obre noves oportunitats que fins fa pocs anys eren difícils d'imaginar. Una d'aquestes tecnologies en alça és la monitoreig acústic passiu (PAM en les seves sigles en anglès). Aquesta tecnologia ofereix millores respecte a metodologies de seguiment més convencionals. No obstant això, encara existeixen limitacions i oportunitats per explorar relacionades amb la disponibilitat de protocols de seguiment estandarditzats i adaptats a contextos reals, així com en aspectes relacionats amb l'ús d'índexs acústics per respondre a preguntes de conservació aplicada.

L'objectiu d'aquesta tesi és explorar noves aplicacions potencials del PAM, que van des del disseny i proves de noves unitats de gravació autònomes de so (ARU en les seves sigles en anglès), fins a la definició de protocols i l'avaluació d'índexs acústics per al seguiment d'ocells, per finalment explorar el seu potencial per a estudis sobre la migració i per avaluar l'impacte real de determinades espècies d'ocells en conflictes entre activitats humanes i la fauna salvatge.

El disseny de protocols PAM específics per a la detecció d'espècies d'ocells, basat en les característiques i rendiment de l'ARU que s'utilitzarà i l'ecologia de les espècies objectiu, ofereix grans oportunitats per maximitzar la detecció d'espècies i minimitza els esforços quan es dissenya un protocol de monitoreig acústic. Aquesta tesi posa de manifest la necessitat d'avaluar l'eficàcia dels equips d'enregistrament per a programes de monitorització acústica, ja que el seu rendiment influeix molt en l'àrea potencialment mostrejada. Tenint en compte els aspectes ecològics rellevants, com la densitat de població i el comportament de cant de l'espècie, aquesta tesi també desenvolupa un protocol PAM específic per a la detecció de l'alosa becuda (*Chersophilus duponti*) que s'ha utilitzat amb èxit posteriorment en contextos reals de seguiment. També es presenten cinc passos lògics per desenvolupar protocols de seguiment eficaços mitjançant l'ús d'ARUs per detectar la presència d'espècies concretes.

L'índex d'activitat vocal (VAR en les seves sigles en anglès) és un dels índexs acústics més utilitzats per estimar l'abundància d'ocells a partir de gravacions de so. Aquesta tesi mostra que el VAR és un indicador bo i factible per estimar l'abundància de l'alosa becuda i l'abellerol europeu (*Merops apiaster*), així com per estimar la pressió de depredació d'aquesta última espècie sobre els ruscs d'abelles. L'establiment de noves relacions sòlides entre l'índex VAR i l'abundància d'espècies presenta un seguit de noves oportunitats i aplicacions per al monitoreig de fauna salvatge que van més enllà de la pròpia estimació d'abundància. En aquest context, la tesi demostra la utilitat del PAM per establir el patró temporal migratori dels abellerols europeus i proporciona un mètode fiable per verificar i quantificar la pressió de depredació dels abellerols europeus en ruscs d'abelles i el seu potencial impacte en l'activitat de vol de les abelles. Aquests resultats proporcionen noves aplicacions per a l'ús de PAM, reforçant la idea del gran potencial d'aquesta metodologia per a l'estudi i el seguiment de la biodiversitat.

ABSTRACT

In a context of the global biodiversity crisis, such as the one in which the planet is found, having accurate information linked to biodiversity monitoring systems is essential to be able to make evidence-based decisions. The development of new technologies for the study of biodiversity is opening up new opportunities that until a few years ago were difficult to imagine. One of these technologies on the rise is passive acoustic monitoring (PAM). This technology offers improvements over more conventional monitoring methodologies. However, there are still some limitations and opportunities to explore related to the availability of standardized monitoring protocols adapted to real contexts, as well as related to the use of derived acoustic indices to answer applied conservation questions.

The objective of this thesis is to explore new potential applications of PAM, going from the design and testing of new Autonomous Recording Units (ARU), to defining protocols and assessing indexes for bird monitoring, to finally explore its potential for bird migration studies and evaluation and monitoring human-wildlife conflicts.

The design of specific PAM protocols for bird monitoring, based on the performance of the ARUS to be used and the ecology of the target species, offers great opportunities for maximizing species detection and minimizes survey efforts when an acoustic monitoring protocol is designed. This thesis highlights the need for assessing the effectiveness of potential recorders for acoustic monitoring programmes, since its performance greatly influences the area potentially sampled. Considering relevant ecological aspects, such as population density and vocal behaviour of the species, this thesis develops a species-specific PAM protocol for the detection of Dupont's lark (*Chersophilus duponti*) that has been successfully used in real monitoring contexts. Five logical steps to develop effective monitoring protocols for detecting species presence, using ARUs, are also presented.

The Vocal Activity Rate (VAR) index is one of the most used acoustic indices for estimating bird abundances from sound recordings. This thesis shows that VAR is a good and feasible indicator to estimate the abundance of the Dupont's lark and the European Bee-eater (*Merops apiaster*) as well as to estimate the hunting pressure of this last species on beehives. The establishment of new robust relationships between VAR index and species abundance presents a range of new opportunities and applications for wildlife population monitoring that goes beyond the abundance estimate itself. In this context, the thesis proves the usefulness of PAM in establishing the migratory temporal pattern of the European bee-eater and to provides a reliable method for verifying and quantifying the predation pressure of European bee-eaters in beehives and its potential impact on honeybee flying activity. These results provide new applications for the use of PAM, reinforcing the idea of the potential of this methodology for the study and monitoring of biodiversity.

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**GENERAL
INTRODUCTION**

GENERAL INTRODUCTION

Biodiversity monitoring and new technologies

Global biodiversity is declining especially in the last decades and is mainly threatened by human-induced pressures like climate change, overexploitation and habitat loss, pollution, and invasive species (Maxwell et al. 2016). In this context of global decline in biodiversity, there is a growing need for cost-effective, scalable, and integrated ecological monitoring techniques and programmes (Henry et al. 2008, Cardinale et al. 2012) able to provide, among other variables, fast and cheap data on biodiversity distribution and abundance. The design of biodiversity surveys integrated in monitoring programmes capable of providing reliable trends in biodiversity are key elements to: i) understand the changes in biodiversity composition and abundance, ii) take evidence-based conservation decisions, and iii) develop conservation programmes to mitigate or reverse biodiversity loss (Possingham et al. 2001). Biodiversity monitoring programmes are also expected to deliver information on trends in key aspects of biodiversity; to provide early warning of problems that might otherwise be difficult or expensive to reverse; to generate quantifiable evidence of conservation successes and conservation failures; to highlight ways to make management more effective; and to provide information on return of conservation investment (Lindemayer et al. 2012).

There are six relevant steps for implementing effective biodiversity monitoring programmes: (i) identifying the monitoring aims; (ii) identifying the key components, functions, and processes to monitor and the precision required to fulfil the goals identified in step (i); (iii) identifying the most suitable monitoring methods for these elements, (iv), carrying out the monitoring activities; (v) managing and analysing the resultant data; and (vi) interpreting the results (Schmeler et al. 2017). Step (iii) is especially critical since robust and suitable monitoring methods are critical to achieve pre-defined expected outputs and should meet the following criteria: select representative sampling locations, have sufficient sample size and sufficient detections of target species, and have a sound temporal sampling scheme (Buckland & Johnston 2017).

In recent decades we have witnessed an incredible increase on biodiversity data and monitoring programmes around the world, according to the development of new monitoring techniques aimed to help reverse the cited biodiversity loss. Nonetheless, there are still big information gaps on specific geographic areas, taxa groups and ecosystems (Pereira et al. 2012). This is reflected by the lack of quantitative data on biodiversity change in two-thirds of the 4th national reports

submitted by Parties to the United Nations Convention on Biological Diversity (CBD) (Bubb et al. 2011).

Traditionally, biodiversity monitoring programmes relied on humans to collect field observations both produced by researchers and/or by volunteers (citizen science) (Kelling et al. 2015). However, in the last decades, advances in technology have offered new opportunities for enhanced data collection and greatly expanded the amount of information collected, the scope, and the impact of biodiversity monitoring programs (Pimm et al. 2015, Schmeler et al. 2017, Stephenson 2020). Most of these new cutting-edge technologies are mainly based in different type of automated and non-invasive sensors like satellite-based sensors, environmental DNA (eDNA) analysis, image sensors (camera traps, thermal and multi-spectral cameras), and acoustic recording devices (e.g., Pettorelli et al. 2014, Beaudrot et al. 2016, Browning et al. 2017, Deiner et al. 2017, Díaz-Delgado & Muncher 2019, Sugai et al. 2019). These new technologies for acquiring biodiversity data provide some benefits compared to traditional human-based monitoring ones. For example, where the establishment of monitoring programmes is difficult, due to remoteness of a region or recurring to high costs of training or travel, new technology can play a key part in providing standardized methods for extensive monitoring of biodiversity (Schmeler et al. 2017). Likewise, these methodologies are especially effective for the study and monitoring of cryptic, elusive, and rare species or for those that live in environments inhospitable to humans like underwater ecosystems (Bohman et al. 2014, Lambert & McDonald 2014, Nowacek et al. 2016).

Although new technologies provide important improvements over traditional monitoring methods, its application is not exempt from considerations and limitations. For example, the technology considered should be selected taking into account the goals of the biodiversity monitoring scheme, the indicators to be measured, the target taxa, and the biome and habitat types involved, as well as by the available capacity and budget on equipment and human resources (Schmeler et al. 2017, Stephenson 2020). For example, while using camera traps might be especially well suited for detecting small mammals in rainforests, this method might be ineffective for detecting understory birds, for which sound recorders could be more suitable, or for monitoring the presence of aquatic wildlife, for which eDNA might be a more cost-effective approach (Bohman et al. 2014, Beaudrot et al. 2016, Deiner et al. 2017, Burivalova et al. 2019). Costs associated with equipment acquisition, the need for large storage and sharing capacity of huge volumes of data and time and expertise required for data analysis can be also great

challenges when using new technologies (Pimm et al. 2015, Browning et al. 2017, Stephenson 2020, Pérez-Granados & Traba 2021).

Terrestrial Passive Acoustic Monitoring, an increasing cut-edge technology for species monitoring

One of these new emerging and non-invasive technologies for biodiversity monitoring is the study of acoustic signals uttered by animal species through the use of sound recorders. Acoustic communication is widespread in the animal world, and very often individuals communicate using a sequence of distinct acoustic elements (Kershenbaum et al. 2014). Many animal species, ranging from small insects to whales, emit acoustic signals that encode information about their presence and activities with multiple purposes behind like communication (i.e., alarm calls, social cohesion, individual identity), echolocation, sexual display, or territorial defence (Bradbury & Vehrencamp, 1998).

Although acoustic signals propagate differently based on multiple intrinsic (e.g., dominant frequency, acoustic volume) and environmental factors (e.g., wind speed, background noise or air temperature, Forrest 1994, Penna & Solís, 1998, Ellinger & Hödl 2003, Darras et al. 2016, Titze & Palaparthi 2018), wildlife vocalizations can be detected remotely under a wide range of circumstances, even with poor visibility or when direct views of target species are impossible. Indeed, researchers have detected and estimated, over decades, the presence or abundance of wildlife based on their acoustic signals. Therefore, the study of wildlife vocalizations offers great opportunities to improve the current biodiversity monitoring programs. Based on that, during the last years there has been an increase in the number of studies recording acoustic signals of animals in the field using sound recorders, followed by their posterior processing; giving place to a new monitoring technique: the Passive Acoustic Monitoring (PAM).

Browning et al. (2017) defined PAM as a methodology that involves surveying and monitoring wildlife and environments using sound recorders (acoustic sensors). The devices, mainly Autonomous Recording Units (ARUs), are deployed in the field for a time to obtain recorded acoustic data using a specified recording schedule. ARUs are sound recorders that can be programmed with specific time schedules to be unattended while operating in the field, with great battery autonomy and storage capacity and built to operate in outdoor conditions (Digby et al. 2013, Shonfield & Bayne, 2017). After acoustic data collection, the recordings are stored and processed to extract useful ecological data. This process consists mainly of identifying and classifying species calls (Darras et al. 2019) or quantifying biotic sound levels (Sueur & Farina

2015) using available acoustic analysis software. Although the recordings can be processed manually (i.e., hearing or visualizing the recordings), the development of sophisticated automated signal recognition software, with the ability to automatically detect and recognize the acoustic signature of a target species' vocalization on sound recordings, have extended the use of PAM at large spatial and temporal scales (Knight et al. 2017). Afterwards, these results are analyzed similarly to other types of survey data using acoustic indexes (e.g., estimate population abundance (Marques et al. 2013), detecting the presence of animal species of interest (Schroeder & McRae 2020) or human impact on wildlife occurrence (Astaras et al. 2017)).

Since 2010, PAM has emerged as an increasingly important and widely used tool for studying wildlife in terrestrial environments (see review in Sugai et al. 2019), although in other ecosystems, such as marine environments, this technology have already been used for many more years before (Sousa-Lima et al. 2013). Some methodological and technical improvements are behind the success of PAM-based research in terrestrial environments. In the last years, several low-cost ARUs (< 100 €) models have been launched (e.g., Atkins & Johnson 2016, Hill et al. 2018, Beason et al. 2019), which have been a great improvement since the cost of previously available ARUs was quite high (> 1000 €). Another multiplier factor for PAM success has been the rapid development of automated and semi-automated signal recognition software, included the development of user-friendly software, which have improved in accuracy and efficiency due to innovations in signal processing and machine learning algorithms (Gibb et al. 2019). Current sound detection and classification tools ranges in analytic complexity from simple thresholding (Digby et al. 2013) to complex statistical machine learning models including artificial and convolutional neural networks (Walters et al. 2012, LeCun et al. 2015), random forest (Ross & Allen 2014), Hidden Markov Models (Zilli et al. 2014) and support vector machines (Heinicke et al. 2015). Although methods are fast improving, variable accuracy of auto-ID tools remains a major issue (Gibb et al. 2019, but see Kahl et al. 2021). Multiple factors like target species vocal repertory variability (Walters et al. 2012), background noise (Stowell et al. 2019), or temporal overlap between calls of different species (Stowell & Plumbley 2014), among others, can affect performance of detection and classification algorithms. Both proprietary commercial and open-source software tools and packages for analysis of sound recordings data are available. The first ones often contain intuitive user-friendly interfaces that enable users to process audio data with less statistical computing skills (e.g., Kaleidoscope Pro from Wildlife Acoustics, Raven Pro from Cornell Lab of Ornithology). However, licenses expenditure can be costly, and limitations of these tools are often not clearly reported by the manufacturer (Browning et al. 2017). In contrast,

open-source software packages are freely available, their limitations are often better documented, and many have very active online communities that may offer assistance with problems (Browning et al. 2017). However, most of the open-source software require greater experience with statistics and computer programming (Gibb et al. 2019). Finally, the development of acoustic indices able to estimate species ecological parameters around recorders (e.g., see review for estimating wildlife density around recorders in Marques et al. 2013, Pérez-Granados & Traba 2021, for birds) or to characterize the acoustic complexity of soundscapes (Pieretti et al. 2011, Sueur & Farina 2015) have also been an important factor to extend the use of such technique (Gibb et al. 2019).

PAM addresses and gives alternative solutions to many of the biases associated to traditional field surveys (based on field observers). For example, PAM may: (i) increase the spatial and temporal scale of the studies at reduced human cost, (ii) minimizes disturbance due to human presence during monitoring, which allows sampling of elusive species or in remote areas, and (iii) offers a great degree of standardization in data collection if common protocols are applied (Browning et al. 2017, Shonfield & Bayne 2017, Pérez-Granados et al. 2018a, Gibb et al. 2019). Another advantage is that acoustic recordings can be stored, reanalyzed, and reinterpreted as new questions arise (the same recordings can be used to answer different ecological questions for the same species). For example, the same sound database was used for studying differences in seasonal vocal activity, the role of nocturnal vocal behavior, or how climatic conditions affects the singing activity of the Undulated Tinamou (*Crypturellus undulatus*) (Pérez-Granados et al. 2020, Pérez-Granados & Schuchmann 2021a, 2021b). Furthermore, incidental standardized sound recordings generated initially by a specific species objective can be reanalyzed for other monitoring species purposes. For example, Newson et al. (2017) showed how extensive acoustic recordings obtained through a bat monitoring volunteer-based project can be also used for Orthoptera monitoring reanalyzing the same recordings.

However, some limitations for their potential use should not be overlooked. Obviously, non-acoustic species or with low acoustic activity can be missed or can remain unidentified (Acevedo & Villanueva-Rivera 2006, Alquezar & Machado 2015). Very noisy environments (such as highly biodiverse areas or with high presence of anthropogenic noise) can mask target sounds, making signal recognition more challenging or even impossible (Pieretti et al. 2011, Knight et al. 2017, but see Deichmann et al. 2018, Burivalova et al. 2019). Moreover, ARUs may have a lower sensitivity than a human listener (Yip et al. 2017), which may reduce the sampling radius of the surveys designed, but this limitation seems to be beginning to be overcome, especially if the

Effective Detection Radius (EDR; defined as the radius at which as many vocalizations are undetected within that distance as are detected beyond that distance, Buckland et al. 2001) of the ARU is estimated (Van Wilgenburg et al. 2017). As other new technologies, PAM requires a large amount of storage capacities and analytic power together with expert time for the analysis of recordings (Roch et al. 2016, Gibb et al. 2019). Although the costs of purpose-designed acoustic sensors have been rapidly decreased in the last few years (e.g., Hill et al. 2018, Beason et al. 2019), state-of-the-art sensors are still costly. So, it is needed to consider that might be a large initial expense associated with establishing an acoustic survey programme, whose cost will vary according to the model used or/and number of ARUs needed.

Passive Acoustic Monitoring methodological frameworks, from sensors and protocols to indexes

PAM methodological frameworks must follow, like any monitoring system, basic general rules that can be synthesised in the clear definition of objectives, the use of the most appropriate methodologies to obtain the data, and correct analysis adjusted to the objectives and interpretation of the results (Schmeler et al. 2017). However, PAM should also follow specific requirements adapted to its own characteristics and the target species to be monitored like: (i) testing the recording equipment and performing pilot surveys, (ii) designing surveys and protocols for determining adequate recording schedules and sampling efforts, which might be useful to optimize the set of audio settings and autonomy in PAM stations, and (iii) identifying the best signal processing and acoustic recognition analysis to generate indexes able to be interpreted from an ecological point of view (Browning et al. 2017, Sugai et al. 2019).

Sound waves attenuate as they travel further through the environment. The distance at which a sound can be detected above ambient background noise is highly influenced for many factors being the main ones the sound's amplitude and frequency (lower frequencies reach higher distances), the caller's relative position to the recorder, and environmental factors such as topography, air temperature, surrounding vegetation characteristics, etc. (Forrest 1994, Penna and Solís 1998, Ellinger & Hödl 2003, Darras et al. 2016, Titze & Palaparathi 2018). These factors have strong implications for monitoring wildlife through PAM, since they affect the likelihood that a calling animal will be detected (Darras et al. 2016). Studies using PAM should evaluate how far the ARU employed is able to record the vocalizations of the target species on the selected habitat, since that distance may greatly differ among taxa (Llusia et al. 2011). Therefore, understanding the distance at which species monitoring are detected and how sound source level, emission conditions and ARUs performance affect the species detectability are crucial factors for quantifying the species use of space. Previous estimates of detection areas can be

achieved using focal signals played back at varying distances and directions from the recorder (Llusia et al. 2011, Hagens et al. 2018). All these aspects will further facilitate the comprehension of the data registered and analyzed.

The distance at which a target species is detected is also highly influenced by the recording equipment selected. ARUs vary in price and can also vary in sound sensitivity, signal-to-noise ratio (SNR), and directionality and quality of microphones (Rempel et al. 2013, Browning et al. 2017, Turgeon et al. 2017), among others. Between these factors, SNR (defined as 10 times the logarithm of the ratio of a standard signal's power to the noise power of the microphone created by its self-noise) can impact acoustic monitoring surveys by affecting the probability that a bird singing or calling will be audible and identifiable on a recording (Rempel et al. 2013, Darras et al. 2020). Microphone sensitivity decreases with field use, adding more potential variability in the effective area sampled even using the same ARU model equipment and thus inducing distance-related biases in detection probability (Turgeon et al. 2017). So, knowing the performance of the ARUs for the specific context where it is going to be used is relevant.

In order to improve possibilities for PAM, there is still special need to develop standardized survey and analysis protocols (Gibb et al. 2019). Sampling design in PAM surveys is influenced by the researchers' knowledge and prior experience on target species (Gibb et al. 2019), resulting in a variety of recording protocols, not necessarily based on scientific results nor directly transferrable between biological groups and research goals (Darras et al. 2018). For example, close related species living in the same area can present very different sound activity patterns (Pérez-Granados & Schuchmann 2020), affecting the potential transferability of the designed protocols between species. In this context, when the objective is monitoring a terrestrial target species, there are few examples of designing and defining detailed protocols adapted to the habitat conditions, population density and ecology of the target species (but see Hagens et al. (2018) and Pérez-Granados et al. (2019)).

Determining an appropriate level of survey effort entails trade-offs between the resolution and accuracy of the data and the costs and logistical complexity of collecting and storing it. Sampling effort in PAM can be optimized through spatial distribution of ARUs and recording schedules (Sugai et al. 2020). When focused on particular species, optimal spatial distribution of acoustic sensors relies on the home range, species density, habitat use and calling behavior of focal taxa. Measurement of detection area or the EDR should also be required in advance to define the number of recorders per site (Sugai et al. 2020). Recording schedules based on behavioural and

ecological aspects of focal taxa and research goal are also critical (Hagens et al. 2018, Sugai et al. 2020). Additionally, a greater autonomy and reduction of audio storage needs can also be achieved by optimizing scheduling recordings to objectives and high activity levels of the target species (Browning et al. 2017, Gibb et al. 2019).

After acoustic data be collected using pre-defined protocols, the obtained recordings have to be stored and processed to extract useful ecological data. Following acoustic processing using signal recognition software, typical sound identification outputs are spatially (linked to the ARU position in the field) and temporally explicit record of species call detections. But these outputs, most of times, have not direct ecological interpretation, so acoustic indexes should be calculated. A core application of species survey programs is to calculate abundance and population trend estimation, and so these aspects have been in the focus of studies based on acoustic monitoring since the beginning. In the case of bird monitoring, density estimation from PAM count data has now several methodological options (Marques et al. 2013, Pérez-Granados & Traba, 2021). Up to eight different methods to estimate bird density using ARUs have already been used, most of them with very few application examples (1-2 case studies), being the approach most often used tested in 20 studies (see review in Pérez-Granados & Traba, 2021). Thus, it seems crucial exploring more deeply existing methods to test specific, easy-to-interpret, and cost-efficient approaches for inferring population densities using ARUs in specific contexts, in order to broaden the scope of these emerging methodologies.

Exploring new applications of Passive Acoustic Monitoring for species in terrestrial ecosystems

Linking technological and methodological advances and analysis improvements, new applications always emerge to answer new and old questions, and PAM is not an exception. This methodology has already been used for monitoring many taxonomic groups (Sugai et al. 2019), being bats, birds, and anurans the animal groups most studied, in this order. However, if we consider just those studies that used ARUs, birds are the group most commonly surveyed. In relation to research topics investigated, studies on species activity patterns are the most frequent (25% of references), followed by habitat use (16%), survey techniques (15%), and species assessments (9%) (Sugai et al. 2019). More specifically, PAM has been used to face challenges that are difficult to approach with more traditional methodologies, such as the detection of highly threatened species with very cryptic behaviours (Bobay et al. 2018, Schoeder & McRae 2020), for early detection of invasive species (Juanes 2018) or to monitor anthropogenic activities potentially damaging for wildlife (Astaras et al. 2017). Two potential

fields in which PAM still have a long way to go and explore are the study of bird migration and the evaluation and monitoring of human-wildlife conflicts.

Many bird species utter flight calls, that is, species-specific vocalizations given primarily during sustained flight, especially during migration (Farnsworth 2005). Despite existing uncertainty on other potential roles of these calls, scientific literature suggests that flight calls help to maintain group cohesion and stimulate migratory restlessness in conspecifics (Farnsworth 2005). Patterns of call counts across seasons and years are often consistent and probably represent some behavioural and biological patterns (e.g., migration timing of different species). Indeed, nocturnal call counts of migrating birds have been used as indices of nocturnal bird migration intensity (Larkin et al. 2002, Farnsworth et al. 2004). Nonetheless, extensive variation in calling rates (within and among species) poses a major challenge for measuring bird abundance from flight calls alone (Farnsworth et al. 2004), which may difficult the use of ARUs for monitoring species-specific patterns of bird migration. Likewise, atmospheric conditions like air temperature, air humidity, or precipitation as well as the time of day may affect call rates independently of migration intensities (Farnsworth 2005, Hüppop & Hilgerloh 2012, Horton et al. 2015). In addition, anthropogenic factors like artificial light may alter the propensity of nocturnal migrants to produce flight calls (Gillings & Scott 2021). Therefore, it is desirable to have independent and complementary data (like radar data, ringing data or direct counts of birds) to compare with the data obtained through PAM and to validate the results obtained using such technique (Farnsworth 2005, Salamon et al. 2016).

Furthermore, all prior studies using PAM for monitoring bird migration have focused on nocturnal bird migration surely due to the potential of this methodology to monitor migration at times when the birds cannot be seen. However, the effectiveness of PAM for monitoring the diurnal migration of bird species is a field to be explored. Diurnal recording has intrinsic potential analytic challenges when compared to the nocturnal period, since background noise (mainly due to human activities, but also other vocalizing animals) is usually higher during the day, and environmental conditions (e.g., higher air temperature) could make more difficult recording the signal as well as the identification by signal recognition software (Knight et al. 2017). Therefore, there is still room to develop new application focusing on the study of the migration of diurnal bird species.

The evaluation and monitoring of human-wildlife conflicts using PAM is another early emerging field. The frequency of harmful interactions between wildlife and humans has widespread in the

last decades due to exponential increase in human population and the resultant expansion of human distribution range and activities (Sanderson et al. 2002, Anand & Radhakrishna 2017). In parallel, scientific papers referencing human–wildlife conflict and wildlife-damage compensation schemes have rapidly grown in recent years (Nyhus 2016, Ravenelle & Nyhus 2017). These interactions are bidirectional, since certain human activities (like poaching) can impact on threatened species and habitats, while certain species can affect human assets such as people's own lives (due to predatory or poisonous species) or productive activities (like crops, cattle, etc.). Development and use of new tools and technologies have been pointed out as a relevant future research need to solve and mitigate human-wildlife conflicts (Nyhus 2016). PAM has already been used as law enforcement tool to control wildlife poaching in protected areas promoting detail on spatiotemporal gun poaching patterns through acoustic gunshot detection (Astaras et al. 2017). It has also been applied to establish an elephant early warning system for human populations who regularly experience serious conflict with the species (Zeppelzauer & Stoeger 2015). Furthermore, PAM has been pointed out as a potential tool for early detection of damaging invasive species (Juanes 2018, Martínez et al. 2020). In this context, it has also been incipiently used with pest invasive species like common starlings (*Sturnus vulgaris*) (Campbell et al. 2017) or cane toads (*Bufo marinus*) (Hu et al. 2009, Brodie et al. 2021) in Australia. Despite these studies, the use of PAM still offers many possibilities within the field of human-wildlife conflict studies.

OBJECTIVES AND STRUCTURE

OBJECTIVES AND STRUCTURE

The objective of this thesis is to explore new potential applications of Passive Acoustic Monitoring (PAM), going from the design and testing of new Autonomous Recording Units (ARU), to defining protocols and assessing indexes for bird monitoring, to finally explore its potential for bird migration studies and evaluation and monitoring human-wildlife conflicts.

The specific objectives of this thesis are:

1. To assess the cost-effectiveness of different audio recording systems considering relevant factors for the design of passive acoustic monitoring protocols.
2. To develop a species-specific protocol for passive acoustic monitoring of an elusive and threatened passerine, the Dupont's lark (*Chersophilus duponti*).
3. To assess the Vocal Activity Rate (VAR) index as a useful acoustic method to infer bird abundance from sound recordings.
4. To explore the potential use of passive acoustic monitoring for new applications like the study of diurnal bird migration and for the evaluation and monitoring of human-wildlife conflicts, using in both cases the European bee-eater (*Merops apiaster*) as model species.

These specific aims are addressed in detail by the five following chapters:

Differences between ARUs may cause bias in, for example, probability detection, estimated population size or trend estimates (Rempel et al. 2013), and so it is essential to know how the characteristics of ARUs can influence the sampled area (Browning et al. 2017). In **Chapter I**, we evaluated the performance of five different audio recording systems, ranging from low- to high-cost multipurpose digital recorders, aiming to elucidate the effect of playback distance and singing direction in their ability to record bird vocalizations. This evaluation included one low-cost audio recorder (LCR) that was later adapted to become a fully functional ARU (see Annex I for technical details), which was used in the following four chapters.

The development of effective, standardized, scalable, and financially sustainable monitoring protocols are key issues for the success of monitoring programs, such as those based on PAM

(Celis-Murillo et al. 2009, Venier et al. 2012, Honrado et al. 2016, Stephenson 2020). In **Chapter II**, we tested and described an acoustic monitoring protocol for detecting the presence of the Dupont's lark using ARUs. This species is especially suitable for passive acoustic monitoring since it has a characteristic and loud song (Pérez-Granados et al. 2018b), its singing habits are nocturnal and it is very difficult to detect visually (Pérez-Granados & López-Iborra 2017), and it has a patchy and regressing distribution (Gómez-Catasús et al. 2018). This protocol was designed under different plausible density scenarios for the species in order to determine the minimum number of recorders, the minimum recording length per night and the number of monitoring nights needed to detect the species in potential monitoring programmes.

The combination of low-cost but good-performance ARUs together with protocols and indexes for estimating abundance that are relatively easy to calculate and interpretate, are key aspects to create new real opportunities for wildlife population monitoring based on passive acoustic monitoring. In **Chapter III**, we examined whether the VAR index (defined as the number of songs uttered per time unit for the target species) can be used as a cost-efficient method to infer abundance of two terrestrial bird species using passive acoustic monitoring. VAR is expected to be a surrogate of birds' abundance around ARUs, and thus the VAR index should increase as the species abundance does (Farnsworth et al. 2004, Oppel et al. 2014). We deployed ARUs in different areas of known estimated abundance for two bird species to elucidate whether there was a significant positive relationship between VAR index and the number of individuals estimated around recorders.

As exposed above, the use of ARUs for wildlife monitoring has increased in recent years and has been used for a wide range of research topics (Sugai et al. 2019), but it has scarcely been used for monitoring bird migrations. Acoustic monitoring of flight calls during the night has proven to be an effective method to study bird night migration (see Farnsworth et al. (2004), Farnsworth & Russell (2007), Sanders & Mennill (2014) and Gillings & Scott (2021)). But, as far as we know, such methodology has never been used before for monitoring the migration pattern of a diurnal bird. In **Chapter IV**, as a new potential application, we evaluated the use of acoustic monitoring to characterise the daily and seasonal diurnal migration pattern of the European Bee-eater in Catalonia (NE Spain). We used VAR as an index of the abundance of bee-eaters to describe migration patterns and citizen science data as a qualitative independent data for comparison to our acoustic monitoring method.

The study and resolution of human-wildlife conflicts needs the development of fast and accurate protocols for the reliable estimation of the competitiveness between humans and protected potential damaging species (Lopéz-Bao et al. 2017). One of this human-wildlife conflicts is the one that occurs between bee-eaters and beekeeping. The impacts of bee-eaters on apiculture production have been relatively well-studied in Europe (e.g., Galeotti & Inglisa 2001, Farinós-Celdrán et al. 2016, Moreno-Opo et al. 2018), and rely on both direct predation, and through the inhibition of honeybees foraging behaviour due to their hunting pressure around the apiaries. In **Chapter V**, and with European Bee-eater as model species, we aimed to assess the utility of PAM as a new and efficient technique to measure bee-eater predation pressure at beehives and its impact on honeybees foraging activity. We examined whether VAR could be used as a reliable index of the bee-eater hunting pressure around honeybee hives. Furthermore, coupled ARUs with automatic hive monitoring systems in apiaries allowed us to analyse the effect of the presence of bee-eaters on the flying activity of honeybees. Our ultimate goal was to demonstrate that PAM can be a useful technique for potential wildlife damage claims validation, especially in situations where direct observation of damage or estimation of the pressure of the causative species can be complicated.

The five chapters of this thesis constitute the following original publications and submitted manuscripts:

Chapter I:

Pérez-Granados, C.; **Bota, G.**; Giralt, D.; Albarracín, J.; Traba, J. (2019) Cost-effectiveness assessment of five audio recording systems for wildlife monitoring: differences between recording distances and singing direction. *Ardeola*, 66 (2): 311-325

Chapter II:

Pérez-Granados, C.; **Bota, G.**; Giralt, D.; Traba, J. (2018) A cost-effective protocol for monitoring birds using autonomous recording units: a case study with a night-time singing passerine. *Bird Study*, 65 (3): 338-345

Chapter III:

Pérez-Granados, C*.; **Bota, G***.; Giralt, D.; Barrero, A.; Gómez-Catusus, J.; Bustillo-de la Rosa, D.; Traba, J. (2019) Vocal activity rate index: a useful method to infer terrestrial bird abundance with acoustic monitoring. *Ibis*, 161(4): 901-907. * Equally contribution.

Chapter IV:

Bota, G., Traba, J., Sardà-Palomera, F., Giralt, D., Pérez-Granados, C. (2020) Acoustic monitoring of diurnally migrating European Bee-eaters agrees with data derived from citizen science. *Ardea*, 108 (2): 139-149

Chapter V:

Bota, G., Traba, J., Sardà-Palomera, F., Giralt, D., Pérez-Granados, C. New technologies for monitoring human-wildlife conflicts: evaluation of passive acoustic monitoring for measuring European Bee-eater predation pressure at beehives. *Ecological Applications* (submitted)

**SUPERVISORS
REPORT**

SUPERVISORS REPORT

Com directors de la tesi doctoral titulada “Monitoreig acústic: noves aplicacions per al seguiment d'ocells i de conflictes entre activitats humanes i fauna” realitzada per Gerard Bota Cabau, presento el següent informe sobre la contribució del doctorant en les publicacions en coautoría que componen la tesi:

Capítol 1. Pérez-Granados, C., **Bota, G.**, Giralt, D., Albarracín, J., Traba, J. (2019) Cost-effectiveness assessment of five audio recording systems for wildlife monitoring: differences between recording distances and singing direction. *Ardeola*, 66 (2): 311-325

Contribució del doctorand: Participació en el disseny de l'experiment, realització de la presa de dades, anàlisi parcial de dades i redacció parcial i revisió del manuscrit.

Sobre la revista: *Ardeola* en el SCR Journal Impact Factor té un índex d'impacte de 0.970 (2019). Es troba en el Q2 del àrea de Ciències Animals i Zoologia i en el Q3 en Ecologia, Evolució, Comportament i Sistemàtica.

Capítol 2. Pérez-Granados, C., **Bota, G.**, Giralt, D., Traba, J. (2018) A cost-effective protocol for monitoring birds using Autonomous Recording Units: a case study with a night-time singing passerine. *Bird Study*, 65 (3): 338-345

Contribució del doctorand: Participació en el disseny de l'experiment, realització parcial de la presa de dades, anàlisi parcial de dades i redacció parcial i revisió del manuscrit.

Sobre la revista: *Bird Study* en el SCR Journal Impact Factor té un índex d'impacte de 1.155 (2018). Es troba en el Q3 del àrea de Natura i Conservació del Paisatge i en el Q3 en Ecologia, Evolució, Comportament i Sistemàtica.

Capítol 3. Pérez-Granados, C*, **Bota, G***, Giralt, D., Barrero, A., Gómez-Catusus, J., Bustillo-de la Rosa, D., Traba, J. (2019) Vocal Activity Rate (VAR) index: a useful method to infer terrestrial bird abundance with acoustic monitoring. *IBIS*, 161(4): 901-907. * Equally contribution.

Contribució del doctorand: Participació en el disseny de l'experiment, realització parcial de la presa de dades, anàlisi de dades i redacció del manuscrit.

Sobre la revista: IBIS en el SCR Journal Impact Factor té un índex d'impacte de 2.088 (2019). Es troba en el Q1 del àrea de Ciències Animals i Zoologia, en el Q1 en Ecologia, Evolució, Comportament i Sistemàtica i Q1 del àrea de Natura i Conservació del Paisatge

Capítol 4. **Bota, G.**, Traba, J., Sardà-Palomera, F., Giral, D., Pérez-Granados, C. (2020) Acoustic monitoring of diurnally migrating European Bee-eaters agrees with data derived from citizen science. *Ardea*, 108 (2): 139-149

Contribució del doctorand: Participació principal en el disseny de l'estudi, anàlisi de dades i redacció del manuscrit. Realització parcial de la presa de dades.

Sobre la revista: ARDEA en el SCR Journal Impact Factor té un índex d'impacte de 1.026 (2020). Es troba en el Q2 del àrea de Ciències Animals i Zoologia, en el Q3 en Ecologia, Evolució, Comportament i Sistemàtica.

Capítol 5. **Bota, G.**, Traba, J., Sardà-Palomera, F., Giral, D., Pérez-Granados, C. New technologies for monitoring human-wildlife conflicts: evaluation of passive acoustic monitoring for measuring European Bee-eater predation pressure at beehives. *Ecological Applications* (enviat)

Contribució del doctorand: Participació principal en el disseny de l'estudi, anàlisi de dades i redacció del manuscrit. Realització parcial de la presa de dades.

També fem constar que cap dels articles abans esmentats ha estat utilitzat per cap dels coautors com a part d'una altra tesi doctoral.



Dr. Cristian Pérez-Granados
Universitat d'Alacant



Dr. Juan Traba Díaz
Universidad Autónoma de Madrid

Barcelona, a 15 de setembre de 2021

CHAPTER I

Cost-effectiveness assessment of five audio recording systems for wildlife monitoring: differences between recording distances and singing direction

Cristian Pérez-Granados, Gerard Bota, David Giralt,
Josep Albarracín & Juan Traba

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Cost-effectiveness assessment of five audio recording systems for wildlife monitoring: differences between recording distances and singing direction.

Cristian Pérez-Granados^{1,2}, Gerard Bota³, David Giralt³, Josep Albarracín³, Juan Traba^{1,4}.

¹ *Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain.*

² *National Institute for Science and Technology in Wetlands (INAU), Federal University of Mato Grosso (UFMT), Computational Bioacoustics Research Unit (CO.BRA), Cuiabá, Mato Grosso, Brazil.*

³ *Biodiversity and Animal Conservation Lab. Landscape Dynamics and Biodiversity programme. Forest Science and Technology Center of Catalonia (CTFC), Solsona, Catalonia, Spain.*

⁴ *Centro de Investigación en Biodiversidad y Cambio Global (CIBC-UAM), Universidad Autónoma de Madrid, Madrid, Spain.*

ABSTRACT

Audio recording systems coupled with automated song recognition are commonly being used for monitoring wildlife. Recorders usually differ in cost and effectiveness, and their performance may vary with source distance, wind speed and acoustic source direction, among other factors. We here assess the cost-effectiveness of five recording systems considering such factors as distance and singing direction. We developed field tests using playback of Dupont's Lark *Chersophilus duponti* songs from nine fixed locations at distances of 1 to 256 m, played towards or away from the recorders' position. We selected this species because its very characteristic song should be easily identified by automated signal recognition software. Field tests were carried out during March 2016 in level dwarf-shrub steppe (mean height < 40 cm) in NE Spain. We found large differences in effectiveness between recorders. The number of songs detected by an automated signal recognition algorithm significantly decreased with distance and when playback was angled away from the recorder position, a factor never previously tested. Finally, we give the design of a cost-effective Autonomous Recording Unit, based upon the most effective recorder. We recommend researchers working with acoustic recorders to evaluate the performance of several devices before making a selection for long-term monitoring programmes, and to consider such factors such as singing direction in their analyses.

Key words: Acoustic monitoring, automated signal recognition, Autonomous recording unit, *Chersophilus duponti*, classification software, playback.

INTRODUCTION

In recent years, researchers have used Autonomous Recording Units (hereafter ARUs) as a tool to monitor a wide range of taxa based on the sounds they produce. Such taxa include mammals (Heinicke et al. 2015), amphibians (Acevedo & Villanueva-Rivera 2006, Shearin et al. 2012), insects (Brandes 2005) and birds, the last of these being the most commonly surveyed group (e.g., Digby et al. 2013, Ganchev et al. 2015, Alquezar & Machado 2015). This methodology requires the placement of one or several ARUs in the field to record sounds during the time of interest (e.g., dawn, Gil et al. 2014, Zwart et al. 2014) or for 24-hour periods (Jahn et al. 2017) followed by interpretation of the recordings. Several studies have demonstrated that ARUs are able to offer a suitable alternative to traditional field survey methods for detecting species presence and for describing habitat occupancy and community composition of animals that produce sounds (see review for birds in Leach et al. 2016).

Field surveys performed by observers are subject to intrinsic biases, notably those due to interpersonal variation in the ability to detect and identify songs, resulting from differences in observer age, experience and hearing acuity (Cyr 1981, Kepler & Scott 1981). Observer presence may also influence vocal activity and natural behaviour or may provoke non-natural displacements of the studied species (Acevedo & Villanueva-Rivera 2006). Moreover, field surveys are usually short and time-restricted and thus prone to temporal biases due to weather conditions, moon phase, daily vocal activity variation, etc. (Bibby et al. 2000, Catchpole & Slater 2008, Pérez-Granados & López-Iborra 2017). The use of ARUs offers an efficient alternative independent of many of the biases of field surveys, since the technique is non-invasive, consistent over time and creates a permanent and archivable record of surveys (Acevedo & Villanueva-Rivera 2006, Brandes 2008). Recordings can be re-examined by experienced observers (Rempel et al. 2005), re-analysed using song identification programs (de Oliveira et al. 2015) and can provide useful information for future studies and comparisons (Alquezar & Machado 2015).

In the last decade, the use of ARUs for monitoring biodiversity has increased in popularity. However, some shortcomings associated with their use should not be overlooked. Species with low sound output can be missed or can remain unidentified (Acevedo & Villanueva-Rivera 2006, Alquezar & Machado 2015). Moreover, ARUs usually have a lower sensitivity than a human listener (Hutto & Stutzman 2009, Yip et al. 2017) that decreases with distance (Jahn et al. 2017). Sound recordings obtained by ARUs provide little or no ability to determine sound direction in

three dimensions and to distinguish between individuals, which makes it difficult to estimate wildlife abundance (but see Drake et al. 2016 and Pérez-Granados et al. 2019). Furthermore, ARUs require a large amount of expert time for the analysis of recordings (Hutto & Stutzman 2009, Digby et al. 2013), although automated signal recognition and classification software for processing large data sets promptly have favoured their use (Heinicke et al. 2015, de Oliveira et al. 2015).

ARUs differ in price, size, weight, sound sensitivity, signal-to-noise ratio, quality and the directionality of microphones, among other factors (Venier et al. 2012, Frstrup & Mennitt 2012, Rempel et al. 2013, Turgeon et al. 2017). Differences between ARUs may cause bias in, for example, estimated population size or trend estimates (Rempel et al. 2013), and so it is essential to test variation in effectiveness between different ARUs. Only Rempel et al. (2013) have empirically tested the differences between six different recording systems. They found differences in their sensitivity (response to different frequencies) and in the signal-to-noise ratio, as well as in the number of bird species detected by a listener after song analyses, irrespective of cost (Rempel et al. 2013). Yip et al. (2017) also evaluated the detection distances of four different ARUs but offer no comparisons or data regarding the differences found between them. Likewise, there are no studies assessing the effectiveness of different recording systems using automated signal recognition software.

Animals use directionality of sound to avoid the risk of being detected by unintended listeners (Larsen & Dabelsteen 1990) and also to focus the sound intensity on the direction of intended listeners. The direction of sound propagation and the head and body position are important factors determining sound transmission efficiency in birds (Titze & Palaparthi 2018). The sensitivity of ARUs is therefore expected to vary with the direction in which sound is being propagated, a prediction so far untested.

The main goal of this study was to evaluate the performance of five different recording systems, ranging from low- to high-cost multipurpose digital recorders with realistic field tests, aiming to elucidate the effect of playback distance and singing direction their effectiveness. We used automated song detection software to estimate the number of signals automatically detected at different distances both with playback broadcasted directly towards recorders (favourable singing direction, hereafter) and also in the opposite direction (unfavourable singing direction, hereafter), to determine an index of their effectiveness. We predicted that more vocalisations would be detected at closer source locations compared to more distant ones. Also there would

be greater detection distances and a higher number of vocalisations detected at the same distance with favourable than with unfavourable singing directions. For each recorder, we also calculated the total amount of software processing time and we made a cost-effectiveness assessment to identify the most adequate recorder to use as an ARU for long-term monitoring studies. Playback consisted of a series of Dupont's Lark songs and were carried in a dwarf-shrub steppe, a typical habitat for the species (Seoane et al. 2006, Pérez-Granados & López-Iborra 2017). We selected this species because its song consists of a discrete number of song types (Pérez-Granados et al. 2016) that end with a very species specific and common sequence (see Supplementary material appendix 1), that should be easily identified in the spectrogram. Our ultimate goal was to build our own weatherproof and programmable ARU, equipped with long-life batteries, for use in further field studies (see Pérez-Granados et al. 2018a, 2018b).

METHODS

Field tests

Field tests were carried out on the sunny and cloudless morning of 24 March 2016 in the Timoneda d'Alfés (Lleida, Catalonia, North-eastern Spain, 45.50°N, 77.76°W). Tests started at 9:28 a.m. and finished at 10:10 a.m. (local time, GMT +1). The Timoneda d'Alfés is a continuous and homogeneous dwarf-shrub steppe (mean height < 40 cm) of 1 km² dominated by *Thymus vulgaris*, *Sideritis scordioides* and *Helianthemum* spp. We used five sets of recording equipment ranging in cost from 35 to 1,100 Euros (2016 prices, Table 1). These were the Wildlife Acoustics Song Meter™ SM2 with pencil microphones (SM2 hereafter), Olympus DM650 16-Bit PCM Stereo Recording (Olympus), Sony ICD-P320 with compact 6-mm element microphones (Sony), Sytech Digital recorder SY-1707 with compact 25-mm element microphones (Sytech), and a Mini USB Voice Recorder SK-001 with AC1517D72772-C processor and integrated microphones (USB, Table 1). All recorders remained available in March 2019, unless SM2 has been updated by SM4. Recorders were attached to a 50cm-tall wooden stick and remained in the same position and above natural vegetation throughout the tests. Recorders were located with microphones in an up-position, separated by 1m in order to minimise any blockage of sound (Rempel et al. 2013). All recordings were made at 44 kHz and 16 bits and using a bit rate of 1411 kbps. Recordings for SM2 and Sytech were made in wav format while recordings for the other units were made in mp3 format and needed post-transformation to wav format for data analyses in Song Scope (see below). Although compression into mp3 format affects the spectral and temporal composition

of the signal (Obrist et al. 2010), and its conversion to wav format does not improve recording quality, this should not influence the signal recognition results (Rempel et al. 2005).

Table 1. Names and specifications of the five recorders used in the study. The approximate cost per unit (€) at the time of purchase is also shown. Costs and specifications for the USB recorder are shown for the customised recorder (Supplementary material appendix 6). *At maximum recording quality and storage capacity.

Abbreviation	Digital recorder	Manufacturer	Storage Capacity (GB)	Recording time* (h)	Programmable	Weatherproof	Cost (euros)
SM2	Wildlife Acoustic Song Meter™ SM2	Wildlife Acoustics	4 x 32	185	Yes	Yes	1,100
Olympus	Olympus DM650 16-Bit PCM Stereo	Olympus	16	107	Yes	No	160
Sony	Sony ICD-P320	Sony	2	7	Partially	No	60
Sytech	Digital recorder Sytech SY-1707	Sytech	8	48	No	No	35
USB	Mini USB Voice Recorder SK-001	QFRR009	16	260	Yes	Yes	180

We broadcast a digital and standardised recording (hereafter playback) of Dupont's Lark songs to aid in interpreting the results and facilitate automatic signal recognition by the classification software. The playback lasted for 70 seconds and comprised 13 Dupont's Lark songs recorded at different sites. Playback equipment consisted of a digital player (CAPADI, ref. MR102) with a coupled tweeter (CAPADI, ref. AB105). Playback volume and height above the ground (1m) were constant throughout the tests. The intensity of the playback, measured as Leq (Equivalent Continuous Sound level) at 2m, was 76.3 dB similar to a normal singing volume of the study species (authors' own data). The recording equipments had omnidirectional microphones and recorded simultaneously. Likewise, device locations did not vary throughout the field tests and were under the same environmental conditions in order to avoid biases.

Distance and singing direction response

The playback was broadcast from nine fixed locations, 32m apart and from 1m to 256m away. We carried out field tests broadcasting the playback both directly towards and away from the recorders, to estimate recorder performance under the most favourable field conditions – a bird singing towards the recorders, and under the most unfavourable conditions – a bird singing

directly away from the recorders. We only broadcast the playback once at each distance and singing direction. Wind speed during field tests was very low (< 2 m/s) and so was disregarded in our analyses.

Sound analyses

Field recordings were analysed by the same observer (CPG) and using the same laptop (Intel(R) Celeron(R) 2.16 GHz, 4096 MB RAM), automated signal recognition software and headphones. Automated song detection was performed using Song Scope 4.1.5 (Wildlife Acoustics 2011), with the help of the spectrogram visualisation tool of the software. Song Scope is able to create a target signal from the characteristics of the set of signals used for training, and uses it as a recogniser file to compare when a recorded sound matches the target signal (Waddle et al. 2009). We built viable and customised recognisers for each recorder (Towsey et al. 2012), always using the same settings after following the software recommendations (see Supplementary material appendix 2). We used a specific recogniser for each recorder after comparing the number of songs detected under favourable singing conditions per recording by both a recorder-specific and a neutral recogniser (the latter using those calls broadcast at playbacks, see Supplementary material appendix 3). In our analyses, we only considered the final sequence of the Dupont's Lark song as a target signal, since this is easily identified in the spectrogram and should be easily detected by automated classification software (see Supplementary material appendix 1).

For building recognisers, we aimed to adjust sample rate, frequency range and minimum frequency to help isolate the target signal, and remove all lower and higher-amplitude events, which are not likely to be part of the target signal (Waddle et al. 2009). In this way, we annotated selected songs in a clean spectrogram (Digby et al. 2013, de Oliveira et al. 2015). We chose and annotated 12 Dupont's Lark songs as models to create one specific recogniser for each recorder (Waddle et al. 2009). We selected six songs from the tests performed under favourable singing directions and another six performed under unfavourable singing directions. In both cases, three songs were selected at 1m, two at 32m and one at 64m. We selected songs from different distances and singing directions to create a more accurate recogniser, able to detect the target signal under imperfect recording conditions. All recognisers were created using at least ten of the 12 selected songs, and in the cases in which a particular song could not be used (e.g., bad spectrogram quality), another song from the same distance and singing direction was selected to minimise biases between recognisers. The most important setting to be considered when building a recogniser is the *cross training* value, which is a measure of how well the recogniser

is expected to perform (Wildlife Acoustics 2011). A low score (e.g., < 50%) may indicate that the generated recogniser may not accurately find the target signal within a recording (Wildlife Acoustics 2011). Recordings were scanned with their own recognisers using algorithm 2.0 in Song Scope (Waddle et al. 2009). Recogniser scanning reported a series of events identified as a target signal by the recogniser. All events were visually and/or acoustically checked, and a true positive was considered when the software correctly matched a Dupont's Lark song, while a false positive was noted when a non-Dupont's Lark song was recognised (Wolfgang & Haines 2016).

For each distance and singing direction we estimated the total number of events detected, the number of true positives and the number of false positives. For each recorder and singing direction we calculated: (1) the maximum detection distance at which at least one Dupont's Lark song was detected, and (2) the effective continuum distance, as the distance at which songs were detected in all shorter distance intervals. We also estimated: (3) detection rate: the percentage of Dupont's Lark songs detected in relation to the total number of songs played, (4) success rate: the percentage of Dupont's Lark songs correctly classified relative to the total number of events recognised, and (5) the time needed by automated signal recognition software to complete recording analyses for each recording system.

Cost-effectiveness assessment

To objectively identify the most effective recorder to be used as an ARU, we proposed a methodology based on scoring each device according to its detection and success rate at each distance and singing direction. Three points were awarded when detection or success rate was > 50%, two points when detection or success rate varied between 50% and 25%, one point when detection or success rate was < 25% and zero points when no songs were detected or correctly classified. We also estimated the number of units of each ARU needed –according to their continuum distance under unfavourable singing directions, limiting distance – to monitor simultaneously a potential habitat patch of 1 km²: this number was multiplied by their estimated price to estimate the total cost.

Statistical analyses

We used GLMM with family binomial to test the probability of a song being detected, using type of recorder (five levels), distance (nine levels) and singing direction (two levels) as fixed variables, and detection success (detected/undetected) as the dependent variable. As the playback employed 13 different Dupont's Lark songs, all of them being broadcast at all distances and directions, we included song type (13 levels) as a random factor. If a fixed effect was significant,

a Tukey's *post hoc* test was performed to test for differences between levels. Model performance was evaluated by plotting standardised residuals versus fixed variables, normal QQ-plots and histogram of residuals. No concrete pattern was found in any case. We tested the difference in the number of songs detected at each specific distance between the tests performed with favourable and unfavourable singing directions using Mann-Whitney U tests. Data analyses were conducted in R 3.4.1 (R Core Team 2016). We used packages "lme4" (Bates et al. 2015) for logistic GLMM and "multcomp" (Hothorn 2008) for post-hoc comparison tests.

RESULTS

Recording analyses

Cross-training values for recognisers created for each recorder were similar and ranged between 72.2 and 78.1 (Table 2). Software processing time differed among devices. Sytech recordings were the fastest to be analysed (226 sec.) while Sony were the most time-consuming (910 sec., Table 2).

Table 2. Cross training of each recogniser and time needed to perform sound-analyses in lab for each recorder. Total number of detections, total number of true positives (% of songs recognised in respect to total songs broadcast by playback is shown between brackets), total number of false positives, maximum detection distance and effective continuum distance per recorder are shown separately for tests performed with favourable and unfavourable singing directions.

	Cross training	Time (s)	Unfavourable singing direction				
			Total events	True positives	False positives	Max. distance	Effect. distance
SM2	78.1 ± 5.8	451	30	17 (14.5%)	13	96	96
Olympus	73.9 ± 14.6	235	10	5 (4.3%)	5	128	1
Sony	75.6 ± 8.1	910	48	11 (9.4%)	37	96	96
Sytech	75.8 ± 12.1	226	10	9 (7.7%)	1	64	64
USB	72.2 ± 13.4	525	33	18 (15.4%)	15	128	128
TOTAL			131	60 (10.3%)	72		

	Cross training	Time (s)	Favourable singing direction				
			Total events	True positives	False positives	Max. distance	Effect. distance
SM2	78.1 ± 5.8	451	46	34 (29.1%)	12	256	192
Olympus	73.9 ± 14.6	235	20	12 (10.3%)	8	128	64
Sony	75.6 ± 8.1	910	67	18 (15.4%)	49	256	192
Sytech	75.8 ± 12.1	226	22	19 (16.2%)	3	224	224
USB	72.2 ± 13.4	525	62	35 (29.9%)	27	256	256
TOTAL			217	118 (20.2%)	106		

Recorder effectiveness

According to GLMM, there were significant differences among recorders in the total number of songs automatically detected (Table 3). SM2 and USB detected a significantly higher number of songs than the rest of recorders (c.22%), while Olympus detected the lowest (c.8%) (Table 2). SM2 and USB did not differ significantly in the Tukey *post-hoc* test (Figure 1).

The maximum detection distance differed greatly among recorders and was strongly affected by singing direction (Table 2). For example, the maximum detection distance in tests performed with favourable singing direction ranged between 256m (SM2, Sony, USB) and 128m (Olympus). Under unfavourable singing direction these values decreased and varied between 128m for the best case (Olympus and USB) and 64m for the worst (Sytech, Table 2). We also detected large differences between recorders and singing direction in relation to effective continuum distance (Table 2). When singing direction was favourable, this value was relatively high and varied between 256m (USB) and 64m (Olympus), but it decreased under unfavourable singing directions up to 128m (USB) and 1m (Olympus, Table 2).

Table 3. Estimates of a general mixed model (GLMM) testing the effect of recorder type, distance to playback source and singing direction on the probability of detecting a Dupont's Lark song. Summary table of type-II partitioning of variances performed for each factor is also shown.

Response variable	Estimate	Std. Error	Z value	Pr(>z)	
(Intercept)	-0.469	0.381	-1,231	0.218	
Distance-32	-0.970	0.289	-3,356	< 0.001	
Distance-64	-1.767	0.317	-5,574	< 0.001	
Distance-96	-2.376	0.353	-6,731	< 0.001	
Distance-128	-2.545	0.367	-6,935	< 0.001	
Distance-160	-3.739	0.515	-7,260	< 0.001	
Distance-192	-3.366	0.456	-7,382	< 0.001	
Distance-224	-4.710	0.750	-6,280	< 0.001	
Distance-256	-3.980	0.561	-7,094	< 0.001	
Recorder-SM2	1.714	0.344	4,983	< 0.001	
Recorder-Sony	0.763	0.362	2,108	0.035	
Recorder-Sytech	0.710	0.368	1,929	0.051	
Recorder-USB	1.822	0.343	5,312	< 0.001	
Singing direction-Unfavourable	-1.081	0.203	-5,325	< 0.001	
Fixed effect	df	Sum Sq	Mean Sq	F	P
Distance	8	113.8	14.2	14.2	< 0.001
Recorder	4	35.7	8.9	8.9	< 0.001
Singing direction	1	28.3	28.3	28.3	< 0.001

The GLMM showed that number of songs detected decreased significantly with distance (Figure 2 and Table 4). Distances greater than 128m showed similar detection success, attending to Tukey *post-hoc* tests (see Supplementary material appendix 4). Tests performed under favourable singing directions detected a significantly greater number of songs when compared to those carried out with unfavourable singing direction (Figure 2). More specifically, tests performed under favourable singing directions detected significantly more songs at four of the considered distances (64, 128, 160 and 192m). Conversely, the number of songs detected for the remaining distances was unrelated to singing direction (Supplementary material appendix 5).

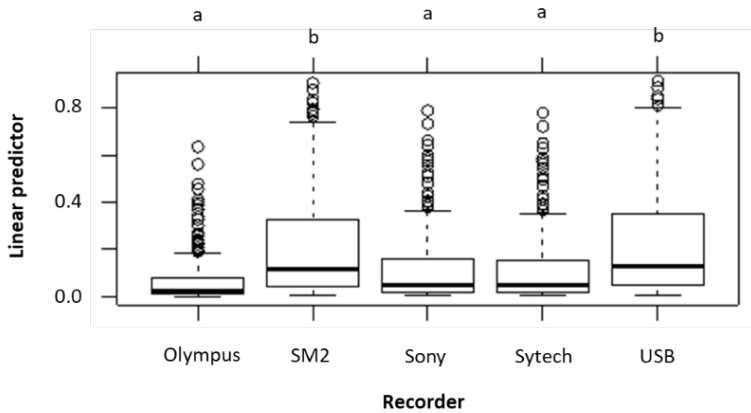


Figure 1. Results of the Tukey post-hoc test for the factor 'Recorder'. Detection significantly varied among recorders, SM2 and USB detecting significantly more songs than the other recorders, and with no differences between them. Different letters mean significant differences in the detection success after Tukey test.

Cost-effectiveness assessment

There were large differences in the total scores obtained per recorder. USB and SM2 had the highest scores, while the other three recorders compared poorly (Table 4), and thus were excluded from the cost-effectiveness assessment. According to their effective continuum distance under an unfavourable singing direction (limiting distance for monitoring studies) 16 USB or 25 SM2 recorders would be needed for monitoring a potential habitat patch of 1 km². This would imply a total cost of 2,880 Euros using USB and 27,500 Euros using SM2.

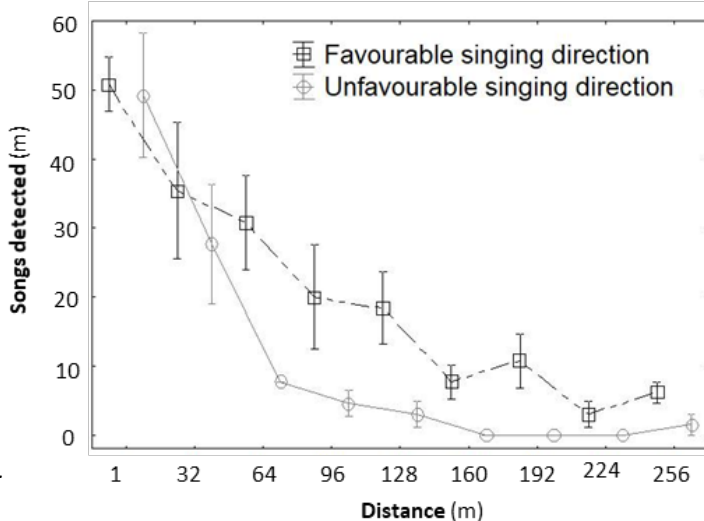


Figure 2. Mean percentage of songs detected (\pm SE) as a function of playback distance. Results are shown separately for tests performed with favourable and unfavourable singing directions

Table 4. Number of songs detected per recorder and distance. Percentage of songs recognised in respect to total songs broadcast by playback is between brackets. Results are shown separately for tests performed with favourable and unfavourable singing directions. The total Score obtained per recorder is also shown. A score was given at each distance and singing direction according to detection and success rate: 3 points when detection or success rate was > 50%, 2 points when detection or success rate was between 50% and 25%, 1 point when detection or success rate was < 25% and 0 points when no songs were detected or correctly classified.

Recorder	Total score	Singing direction	Distance									
			1m	32m	64m	96m	128m	160m	192m	224m	256m	
SM2	56	Favourable	7 (53.8%)	6 (46.2%)	6 (46.2%)	6 (46.2%)	5 (38.5%)	1 (7.7%)	2 (15.4%)	0 (0%)	1 (7.7%)	
		Unfavourable	9 (69.2%)	6 (46.2%)	1 (7.7%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
Olympus	31	Favourable	7 (53.8%)	2 (15.4%)	2 (15.4%)	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
		Unfavourable	3 (23.1%)	0 (0%)	1 (7.7%)	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
Sony	35	Favourable	6 (46.2%)	3 (23.1%)	2 (15.4%)	2 (15.4%)	2 (15.4%)	1 (7.7%)	1 (7.7%)	0 (0%)	1 (7.7%)	
		Unfavourable	6 (46.2%)	3 (23.1%)	1 (7.7%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
Sytech	47	Favourable	5 (38.5%)	3 (23.1%)	4 (30.8%)	2 (15.4%)	2 (15.4%)	1 (7.7%)	1 (7.7%)	1 (7.7%)	0 (0%)	
		Unfavourable	5 (38.5%)	3 (23.1%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
USB	57	Favourable	8 (61.5%)	9 (69.2%)	6 (46.2%)	3 (23.1%)	2 (15.4%)	2 (15.4%)	3 (23.1%)	1 (7.7%)	1 (7.7%)	
		Unfavourable	9 (69.2%)	6 (46.2%)	1 (7.7%)	1 (7.7%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)	

DISCUSSION

Our results revealed large differences among different recorders. In general, and in agreement with our predictions and previous studies, recorder performance decreased with increasing distance from sound source (e.g., Rempel et al. 2013, Yip et al. 2017). Maximum and effective detection distance differed greatly among devices, and that difference may be greater than 100m in some instances. This result highlights the need for assessing the effectiveness of selected recorders before considering an effective distance for monitoring programmes. We have also found that the probability of detecting songs differed with singing direction, as expected. More vocalisations were detected at intermediate–long distances (64–192m) in tests performed at favourable than with unfavourable singing directions. This could be because singing direction had no effect on recording quality at short distances, while at distances greater than 192m, songs were uttered too far and therefore only occasionally detected even with favourable singing direction. To our knowledge, this is the first study showing the strong influence of singing direction on detection distance by recorder, which highlights the need to include this factor in further research and to consider it when estimating effective detection radius of acoustic recorders. We are aware, however, that our results are based on one replicate per song type at each distance, and in a single-species study, and that detection distances would differ between species and habitat type according to their signal characteristics and sound propagation (Oppel et al. 2014, Yip et al. 2017). Therefore, relative differences found between recorders could also change according to the different habitats and species tested.

The cost-effectiveness assessment also showed large differences between recorders and revealed that a medium priced unit (USB) performed as well as the costliest device (SM2). Rempel et al. (2013) also detected similar performance between the cheapest (Zoom H2) and the most expensive unit (IR-C1). Our cost-effectiveness assessment suggests that a USB recorder could be a good choice for long-term studies, where costs are a limiting factor and sound quality recording (USB does not record on wav format) may not be a priority. However, cost performance should not be the only parameter used to select a recording system for research and monitoring studies (Rempel et al. 2013). According to our results, USB was also the device with the largest effective continuum distance, thereby confirming its suitability for long-term studies, given the lower number of devices, and thus shorter period for analysis, needed, to cover certain areas.

We are aware that we based our cost-effectiveness assessment on success and detection rate across distance. This variable is quite important to avoid biases when estimating animal abundance or site occupancy and to estimate how many devices are needed to survey a habitat patch (Pérez-Granados et al. 2018a). A low detection distance can be a handicap when covering large areas (Efford et al. 2009) but, depending on the objectives and/or the limitations of each study, other factors, such as recording time, battery life, available memory, programmability and portability should also be considered. After selecting the USB as the most effective recorder to build our own recorder, we needed to make a significant number of alterations to turn it into a functional ARU (see procedure and final recorder in Supplementary material appendix 6). However, it is not available for purchase by the general public. Moreover, in the last few years, new ARUs that are smaller, cheaper, available to the general public and with more functions and capabilities than self-adapted USB, have appeared (Hill et al. 2018, Beason et al. 2018).

ARUs are a useful tool for monitoring wildlife and there are many recorders with very different configurations (ability to make different recordings in each channel, GPS location, automatic modification of starting time according to sunrise, sample rate selection, etc.). Researchers will probably base their selection on cost, expert opinion or previous experience, among other factors, but our study suggests that this may not be the only approach. Instead, we also recommend that the field performance of a set of different recorders be evaluated before choosing one for a monitoring program, since effectiveness may greatly differ among them.

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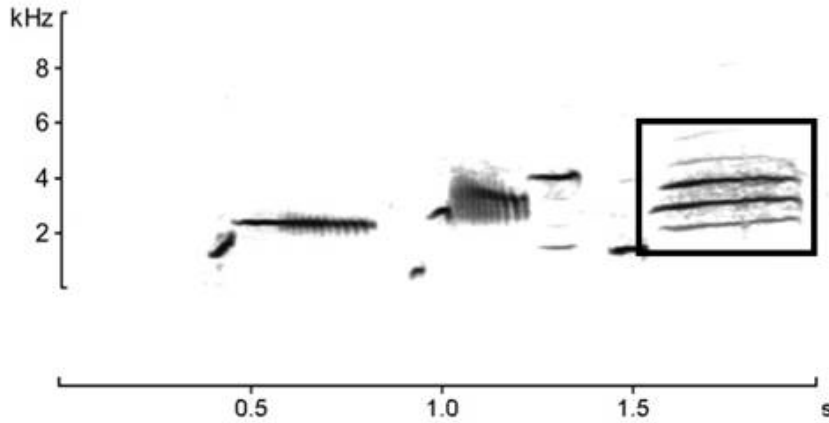
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APPENDIX 1

Supplementary material S1. *Sonogram of a typical Dupont's Lark song. Rectangle shows the final song of the species, which was used for building recognisers, due to its very particular, recognisable, and consistent characteristics.*



Supplementary material S2. *Settings used for recogniser creation in Song Scope.*

Display

Brightness: 0

Contrast: 0

Inverse video: No

Hue: 0

Saturation: 255

Luminosity: 128

Mixer

Sample rate: 10.000 Hz

Playback speed: Normal

Max sample delay: 64

First channel Gain (dB): 0

First channel delay (1/32,000 s): 0

Spectrogram

FFT Size: 256

FFT Overlap: ½

Frequency minimum: 46 (1,796 Hz)

Frequency range: 75 (4,726 Hz)

Amplitude Gain (Db): 0

Background filter: 1 s

Detector

Max syllable (ms): 600

Max syllable gap (ms): 600

Max song (ms): 500

Dinamic range: 20

Algorithm: 2.0

Recognisers

Minimum quality: 20

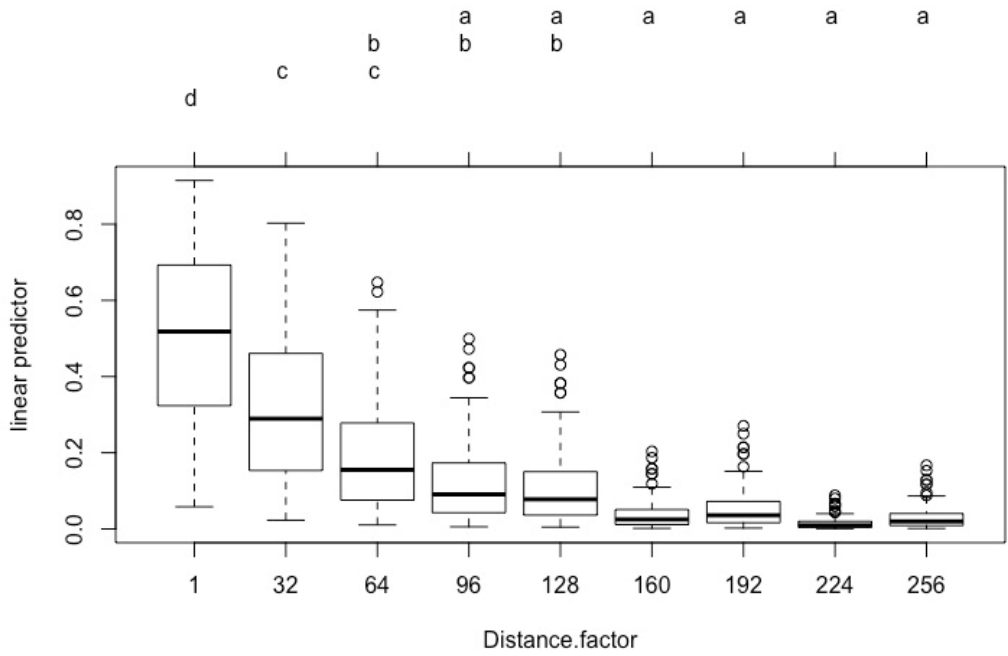
Minimum score: 50

Show top: 1 match

Supplementary material S3: *Total number and percentage of true positives of songs recognised in respect to total songs uttered by playback under favourable singing direction. Results are shown for customised recognisers built for each recorder and for a neutral recogniser built using the playback song.*

Recorder	Specific recogniser	Neutral recogniser
SM2	34 (29.1%)	21 (17.9%)
Olympus	12 (10.3%)	14 (12.0%)
Sony	18 (15.4%)	6 (5.1%)
Sytech	19 (16.2%)	13 (11.1%)
USB	35 (29.9%)	33 (28.2%)
TOTAL	118 (20.2%)	87 (14.8%)

Supplementary material S4. Results of the Tukey post-hoc test for the factor Distance. Detection decreased significantly with increasing distance between playback and recorders. Different letters mean significant differences in the detection success after Tukey test.



Supplementary material S5. *Summary table of Mann-Whitney U tests for the comparison between the total number of songs detected at each distance by all ARUs with favourable and unfavourable singing directions.*

Distance	U	Z	p-value
1	12.0	0	1
32	11.0	0.208	0.834
64	0.0	2.507	0.007
96	4.0	1.670	0.094
128	1.0	2.298	0.021
160	2.5	1.985	0.047
192	2.5	1.985	0.047
224	7.5	0.940	0.347
256	5.0	1.462	0.143

CHAPTER II

A cost-effective protocol for monitoring birds using autonomous recording units: a case study with a night-time singing passerine

Cristian Pérez-Granados, Gerard Bota, David Giralt & Juan Traba

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A cost-effective protocol for monitoring birds using Autonomous Recording Units: a case study with a night-time singing passerine.

Cristian Pérez-Granados¹, Gerard Bota², David Giralte², Juan Traba¹.

¹ *Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain.*

² *Biodiversity and Animal Conservation Lab. Landscape dynamics and biodiversity Programme. Forest Sciences Center of Catalonia (CTFC), Solsona, Catalonia, Spain.*

ABSTRACT

We describe for the first time an effective monitoring protocol for detecting wildlife presence using Autonomous Recording Units (ARUs) under different densities scenarios.

Aims: To describe an effective protocol for monitoring a night-time singing passerine, the Dupont's Lark (*Chersophilus duponti*), using ARUs.

Methods: We estimate, using both simulations and field-collected data, the number of devices needed to reliably detect the species under different density scenarios, and to assess recording time and the number of working days needed to ensure species detection. We placed between four and six ARUs in three Dupont's Lark populations with different bird densities. Devices were programmed to record for 90 minutes per day for four consecutive days. ARUs were deployed between April-June of 2017.

Results: We found large differences in the number of recorders needed to detect species presence under different density scenarios, with larger numbers of ARUs required in less dense populations. The number of ARUs needed differed between estimates obtained by simulations and with field data. This could be related to movements of the monitored species while they were singing. According to our results, the monitoring period for detecting the Dupont's Lark could be as little as one hour of recording (from one hour before dawn to dawn) and two monitoring days, the minimum monitoring time needed to detect the species in all populations surveyed, regardless of density scenarios.

Conclusion: Our results cannot be directly extrapolated to other singing species since singing behaviour and characteristics greatly differ between species. We describe five logical steps to develop effective wildlife monitoring protocols using ARUs for detecting species presence, which may be helpful for future studies and with different species.

Keywords: Acoustic monitoring, ARUs, monitoring protocol, presence, signal recognition software.

INTRODUCTION

Due to the global decline in biodiversity, there is an urgent need for rapid and effective monitoring programs to assess the conservation status of species and to maximize the effectiveness of conservation effort (Brandes 2008, Potamitis et al. 2014). The minimum knowledge needed for species conservation is whether target species are present within an area, and how they are distributed (Li et al. 2010). Furthermore, well-designed monitoring programs with standardized equipment and protocols can provide highly repeatable and reliable data to estimate population trends (Pereira & Cooper 2006, Buxton et al. 2013). In this context, the use of acoustic Autonomous Recording Units (ARUs hereafter) as a tool for monitoring a wide range of taxa have rapidly increased in the last several years (e.g., Heinicke et al. 2015, Alquezar & Machado 2015, Hedley et al. 2017, Van Wilgenburgh et al. 2017). ARUs function on their own while offering a suitable and efficient alternative for wildlife monitoring (see review for birds in Leach et al. 2016). Moreover, monitoring programs based on ARUs are not subjected to many of the biases of traditional field monitoring (detection differences between observers, temporal bias, etc.), since this is a non-invasive technique that creates a permanent, repeatable and archivable record of surveys (Acevedo & Villanueva-Rivera 2006, Brandes 2008), which can be checked by different observers.

The use of ARUs has some disadvantages that must not be overlooked and may even rule out their use for monitoring purposes. Recorders usually have less sensitivity than a human listener (Hutto & Stutzman 2009, Yip et al. 2017), which may be relevant for monitoring rare species or those with reduced vocal activity, and they have costs related to device acquisition. However, the main obstacle that has hampered the widespread use of ARUs is the large amount of expert time needed to analyse recordings (Hutto & Stutzman 2009, Digby et al. 2013). In recent years significant progress has been made in audio signal processing and automated signal recognition, making it possible to process large data sets in a timely manner (see review in Knight et al. 2017), and therefore facilitating the use of ARUs as an automated non-invasive monitoring technique (Buxton et al. 2013).

The number of ARUs deployed per site for wildlife monitoring depends on the radius within which it is possible to effectively detect distant songs of monitored species. This radius can differ greatly among recorders, habitats and species (e.g., Digby et al. 2013, Bota et al. 2017, Yip et al. 2017). Likewise, monitoring time using ARUs, including recording time per day and number of days of monitoring, greatly differs between model species and study objectives. Monitoring

periods in previous studies have varied between a few minutes to several hours per day (e.g., 10 min in a single day, Celis-Murillo et al. 2012; 6 h per day during seven consecutive months; Heinicke et al. 2015), making data analysis difficult even with the help of automated signal recognition software. The monitoring protocol can even differ between studies with a similar purpose, causing bias and making comparisons difficult. For example, Holmes et al. (2015) monitored bird presence obtaining eight recordings each day (recording length varied from 15 to 75 minutes) during a mean number of 10 work-days, while Goyette et al. (2011) monitored bird presence with a single recording but at least 48 h of continuous recording. The development of effective and standardized monitoring protocols will lead to greater time and economic efficiency and when possible, to a greater degree of standardization in data collection (Celis-Murillo et al. 2009, Venier et al. 2012). This may allow repeatable and robust data comparable over time and between sites, which may be used as an effective technique for monitoring habitat quality (Lin et al. 2017) and changes in species presence or abundance (Buxton & Jones 2012, Buxton et al. 2013).

Although a protocol for the use of ARUs should be a pre-requisite for any monitoring program, we found no study assessing the recording time and number of ARUs needed for monitoring species presence within a patch. In this paper, we aim to describe an effective protocol for detecting the presence of a night-time singing passerine using ARUs. We chose the Dupont's Lark (*Chersophilus duponti*) as a study model. Traditional field surveys developed for monitoring the Dupont's Lark have always been based on auditory contacts due to its mainly nocturnal singing and evasive day-time behaviour (Pérez-Granados & López-Iborra 2017). Moreover, European Dupont's Lark populations are patchily distributed over a large number of remote sites throughout peninsular Spain at different densities (Suárez 2010), which makes a spatially homogenised and well-distributed sampling effort difficult. The species is classified as 'Near Threatened' in the IUCN Red List (BirdLife International 2017) and as 'Vulnerable' in the European Red List of Birds (BirdLife International 2015). Therefore, we consider acoustic monitoring with ARUs as especially well-suited to improving monitoring programs of this species. This is evidenced by the fact that some managers have begun to suggest the use of ARUs as an aid for monitoring the Dupont's Lark (authors' own data).

The main objectives of this paper were to: (1) test and describe the procedure of an effective monitoring protocol using ARUs and automated song analyses for a night-time singing passerine, the Dupont's Lark, under different plausible density scenarios; (2) determine the minimum number of recorders needed to detect species presence using virtual and field data under

different density scenarios; and (3) estimate the minimum recording length per night and number of nights of monitoring needed to detect species presence under three different bird density scenarios. While our study was focused on the Dupont's Lark, we describe a series of steps to develop an effective monitoring protocol that may be useful for different taxa.

MATERIAL AND METHODS

Simulations under different density scenarios

We mathematically estimated the minimum number of ARUs needed to reliably detect the presence of the Dupont's Lark within a virtual 100 ha habitat patch. For the experiment, we considered an effective continuum distance of 128 m, which is the effective continuum distance estimated for our ARU for the studied species for opposite singing direction (see Field Recording section for how continuum distance was estimated). By using a virtual space, we sequentially added virtual ARU devices one by one in up to 16 locations (constrained to not overlap in their effective continuum distance). In this way, the entire virtual area would eventually be covered. We simulated the presence of singing individuals under four different plausible bird density scenarios (0.1, 0.25, 1, and 4 males/10 ha) and built accumulation curves of detectability rate, as a function of the number of ARUs placed. We assumed that each virtual male was a singing male. We considered 0.1 males/10 ha as an example of habitat patches occupied with a much reduced presence of the species. The density scenarios of 0.25 and 1 males/10 ha correspond to the average Dupont's Lark density estimated in Spain (0.27 males/10 ha, Suárez 2010), and to that found in core areas, such as the Layna moorland (1 males/10 ha, Garza et al. 2005), respectively. We used the density of 4 males/10 ha as an example of the highest densities estimated for the species in specific habitat patches (Suárez 2010).

Study area

The study area comprised three Dupont's Lark populations located in north-eastern Spain (Fig. 1). The Timoneda d'Alfés (Alfés, hereafter) population was located in Lleida (Catalonia, 41.30°N, 0.37°E, Fig. 1), occupying a low shrub-steppe dominated by thyme (*Thymus vulgaris*), shepherd's tea (*Sideritis scordioides*) and rushrose (*Helianthemum* spp.). The Barcones (41.17°N, 2.49°W) and Alcubilla de las Peñas (Alcubilla, hereafter; 41.15°N, 2.31°W) populations were located in Soria (Castile and León, Fig. 1), both on natural steppes dominated by small shrubs, such as thyme (*Thymus* spp.), broom (*Genista* spp.) and lavender (*Lavandula* spp.). We selected these sites because they had a similar patch size (range 70-100 ha) but different Dupont's Lark densities during 2017 (see below).

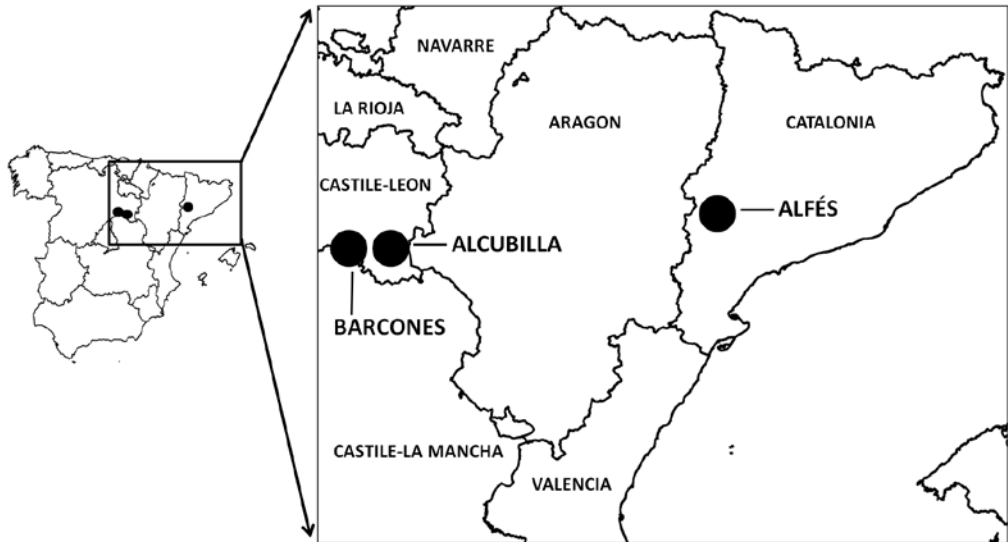


Figure 1. Location in north-eastern Spain of the three Dupont's Lark populations surveyed during the breeding period of 2017.

Bird data

The abundance of Dupont's Lark males at each site was estimated by a mapping method, based on four visits following line transects with a 500-m maximum detection band on each side of the observer (Pérez-Granados & López-Iborra 2017). Censuses were performed from 10th April to 20th June 2017 and only males were considered since females are presumably vocally inactive in this species. The distance of singing males from the observer was estimated acoustically and its location was recorded by GPS. The location of singing males detected in each visit were mapped (using ArcGIS 9.3, ESRI 2008) and a territory was defined when at least two registrations of a singing male (Bibby et al. 2000) were within a radius of 100 m, following the proposed methodology for counting Dupont's Lark (Pérez-Granados & López-Iborra 2017). We assumed a probability of detection equal to 1, since Dupont's Lark songs may be heard up to 1 km (Laiolo et al. 2007), and the entire habitat patches were covered during the censuses. Therefore, species' density (males/10 ha) in each population was calculated by dividing the total number of males estimated by patch size of suitable habitat. Censuses were carried out by walking at a constant speed (1-3 km) on dry and windless days. Census time was from 60 minutes before dawn to dawn, the maximum singing activity period for the species (Pérez-Granados & López-Iborra 2017). In Alcubilla, we estimated a mean density of 0.29 males/10 ha (2 males in 70 ha), 0.9 males/10 ha in Alfés (9 males in 100 ha) and 3.62 males/10 ha in Barcones (29 males in 80

ha). Therefore, we used these as study models of Dupont's Lark populations with low (Alcubilla), medium (Alfés) and high density (Barcones).

Field recording using autonomous recording units

We custom-designed our ARUs after performing field tests and a cost-effectiveness assessment for five acoustic recorders (Traba et al. 2017). In the assessment, we broadcasted Dupont's Lark songs from nine fixed locations from 1 to 256 m, either towards or against the recorder positions aiming to estimate parameters like maximum detection distance or effective continuum distance (distance at which at least one Dupont's Lark song was automatically detected in all distance intervals, with no distance lag) (own data). ARUs consisted of a USB Voice Recorder SK-001 with processor AC1517D72772-C and one integrated and single-channel electret microphone. Recorders were powered by 12V/1.8 mAh Lipo batteries (minimum 15-hour autonomy recording continuously), and were also connected to a digital timer to program recorder activation and registration at selected times. Recordings were collected on 4 Gb microSD memory cards capable of storing 60 hours of continuous data. Equipment was protected in easily portable and weatherproof plastic boxes (60x80x160 mm) made cryptic by painting them greenish-brown with spots.

We placed six ARUs in each monitored population during the breeding season of 2017 (April-May). However, only four units worked properly in the Alfés population due to technical issues. Locations of recorders were selected to cover as much of the patch patch area as possible but were constrained to not overlap in their radius of effective continuum distance. ARUs were ground-located with omnidirectional microphones in an upward position and horizontally separated by at least 50 cm from natural vegetation to minimize sound blockage (Rempel et al. 2013). ARUs were left in each population for four consecutive days, and a digital timer was programmed to record for 90 minutes, from one hour before dawn to 30 minutes after dawn (Pérez-Granados & López-Iborra 2017). Recording time covers the maximum singing activity period of the species. Recordings were split into 30-minute length files for analytical purposes. Daily times of sunrise at the geographic location of the studied areas were obtained from the Spanish Ministry of Development (<http://www.fomento.es>). We used a sample rate of 44.1 kHz and 16 bits in stereo mode for all recordings. Recordings were obtained in mp3 format and needed a post-transformation to wav format for analysis in Song Scope (see below).

Recording analyses

Automatic song recognition of Dupont's Lark presence was performed using Song Scope 4.1.5 (Wildlife Acoustics 2011), which is one of the most efficient programs available for automatic song recognition (Knight et al. 2017). We created a target signal for Dupont's Lark songs with the help of the spectrogram visualization tool in the software, which was used as a recogniser (Waddle et al. 2009). To build the recogniser, we used the final sequence of the species' song (the 'whee-ur-wheeee' song described by Cramp (1988), which is easily identified in a spectrogram and thus, can be easily detected by an automated classification software (Fig. 2). To build the recogniser, we adjusted sample rate and frequency ranges aiming to isolate the target signal, and removed all lower and higher amplitude events (Waddle et al. 2009, Towsey et al. 2012). Song Scope output reported a number of events that matched the target signal. These events were visually and/or acoustically checked by the same researcher, to confirm Dupont's Lark presence when needed.

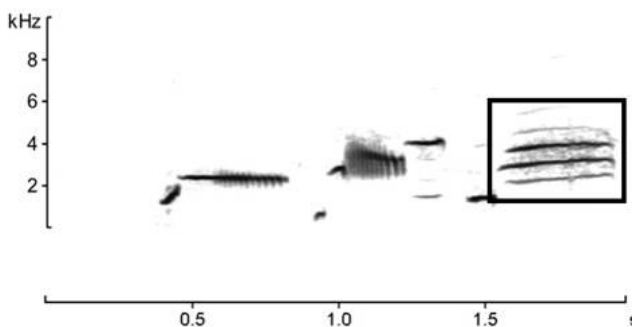


Figure 2. *Sonogram of a typical Dupont's Lark song. Rectangle shows the final song of the species, which was used for building the recogniser.*

Statistical analyses

We mathematically assessed the minimum number of ARUs needed to reliably detect the presence of the Dupont's Lark under the four different density scenarios. We considered that the presence of the species was reliably detected when we reached 90% probability of detection. We estimated the number of times an individual was detected through 1,000 randomizations using random locations for individuals and fixed locations for ARUs. We considered detection as the event when a virtual male fell within the effective radius of an ARU (128 m). Males were created to be static (i.e., not moving within a buffer), so these results should be considered as a minimum approximation to detection probability. The probability of detecting species presence

for each simulated density scenario was evaluated through logistic regression using presence/absence of the species as a dependent variable and number of ARUs as a predictor variable. Data analyses were conducted in R 3.4.1. (R Core Team 2016). We also created accumulation curves of detectability rate with field data collected in the three populations surveyed as a function of the number of ARUs deployed.

RESULTS

Simulations under different density scenarios

The number of ARUs needed to detect species presence in the considered virtual habitat patch (100 ha) differed greatly between the four density scenarios (Fig. 3). In the lowest density scenario (0.1 males/10 ha), 16 units were needed to ensure the detection of the species, while in those scenarios with a mean density of 0.25 and 1 males/10 ha, nine and four ARUs were sufficient to detect species presence with 90% of confidence, respectively (Fig. 3). Only one ARU was required to reach a similar confidence percentage when density increased to 4 males/10 ha, respectively (Fig. 3).

Estimation of ARUs needed to detect the species under real situations

We found large differences in the number of ARUs needed to accurately detect the presence of the species between the three monitored populations. In the high density population, the species was detected in all locations where ARUs were placed (Fig. 4). However, two and four devices were needed to reliably detect Dupont's Lark presence in the medium and low density populations, respectively (Fig. 4).

Recording time and number of nights

Dupont's Lark presence was detected in 39 out of 64 ARUs/night. In 37 of the cases (95%), the species was detected during the first night after ARUs were deployed. However, twice (5%) the species was not detected until the second night of monitoring, both cases in the low-density population. Therefore, the species was always detected within the first two nights of monitoring in all studied populations.

In relation to recording time, the species was always detected in the first hour of recording (N = 39), from one hour before dawn to dawn, with no new detections in the third recording phase (from dawn to 30 minutes after dawn). The best time for detecting the species was the period between one hour to 30 minutes before dawn, when the species was detected in 95% of the cases with known presence (N = 37), followed by the period between 30 minutes before dawn to dawn (82%, N = 32) and lastly the 30 minutes after dawn (46%, N = 18).

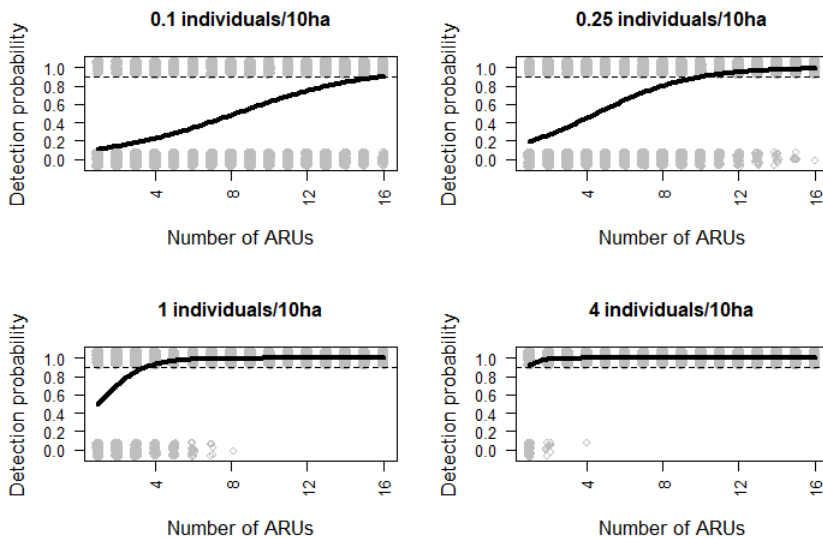


Figure 3. Estimates of the probability to detect the presence of the Dupont's Lark under four density scenarios in a 100 ha patch, given a different number of Autonomous Recording Units (ARUs) deployed. Estimates were obtained through logistic regression using presence/absence as a dependent variable and the number of ARUs as a predictor variable. Grey points show the estimates (presence/absence) derived from 1,000 randomizations using different random locations per individuals and regular locations for ARUs. The dotted line marks 90% probability to detect the species.

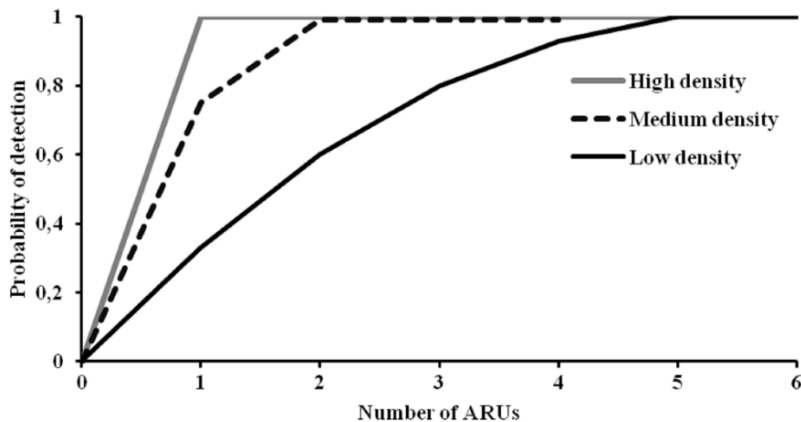


Figure 4. The probability of detection of Dupont's lark as a function of the number of Autonomous Recording Units (ARUs) deployed in three populations surveyed during the 2017 breeding period in Spain. Populations had different densities and curves were built for each density scenario separately. In the population of medium density only four ARUs were used and thus this curve is shorter.

DISCUSSION

In this paper, we describe for first time a protocol for songbird monitoring using ARUs, in which we estimate the number of devices, the number of nights and recording time needed to ensure species detection. Our results suggest that this validation is needed and useful before starting a monitoring program with automatic acoustic recorders, since the probability of detection of the monitored species may vary greatly depending on bird singing characteristics, habitat and the number of ARUs deployed, recording time and the number of nights of monitoring. Moreover, we present a test of the number of ARUs needed to detect species presence based on simulations and field data. Our simulations showed that the number of devices needed to detect species presence depends on bird density ranging from one ARU per 100 ha under a high bird density scenario (4 males/10 ha), to 16 ARUs per 100 ha under the lowest density scenario (0.1 males/10 ha). We also found large differences in the number of ARUs required to detect species presence in the three surveyed populations according to mean bird density, with an increasing number of devices needed under lower bird density scenarios. The estimated number of ARUs needed to detect the monitored species in the highest density scenarios by both simulations and field data were in agreement and indicate that Dupont's Lark or a hypothetical species may be properly detected with a low number of devices and effort nights. However, estimates of the number of ARUs needed to reliably detect species presence in simulations performed with 0.25 and 1 male/10 ha were around two times greater than those obtained in the field in populations with similar densities. This contradiction highlights the need for field-testing results obtained by simulations under controlled situations. We are aware that much of the variation found can be explained by the fact that we considered virtual static individuals in simulations, while Dupont's Lark tend to move while singing (Pérez-Granados & López-Iborra 2017, David Serrano, pers. com.), which may increase the probability of individual detection.

Our results also highlight that a period of one hour of recording, from one hour before dawn to dawn, and two monitoring nights is sufficient time for detecting presence in all surveyed Dupont's Lark populations, regardless of population density. However, singing activity and singing behaviour differ greatly between species, which suggests a necessity to adequately estimate the maximum period of singing activity for monitored species before planning any monitoring programme, so as to increase the effectiveness and reduce time and cost of data analyses. For example, the first 30 minutes after dawn, when many of the European passerines are most vocally active, was the worst time for detecting the Dupont's Lark during the monitoring period, due to its mainly nocturnal singing behaviour (Laiolo et al. 2007, Pérez-

Granados & López-Iborra 2017). We also found differences in the number of nights of monitoring needed to reliably detect Dupont's Lark as a function of bird density. The species was always detected during the first night of monitoring in the high and medium density populations, but it was not detected until the second night in the lowest density population. This could be related to different singing behaviour between populations according to mean density, since Dupont's Lark song rate and song diversity are positively related to population size (Laiolo & Tella 2005, 2007, Laiolo et al. 2008). Likewise, the number of monitoring days needed to detect species presence also differs between species according to their singing behaviour. During the spring 2017, we used the same ARU for detecting the presence of Western Capercaillies (*Tetrao urogallus*) in singing leks, and in some leks the species was not detected until the seventh day of monitoring. In other leks, however, males were detected during the first day of monitoring (D. Guixé, personal comm.). Our results show that field tests to determine recording time and number of nights needed for effective monitoring must be conducted before starting any monitoring programme using ARUs with other species, since they seem to differ greatly between considered species. Our results may be used as a source of comparison and protocol.

The use of ARUs for wildlife monitoring has rapidly increased in recent years (e.g., Heinicke et al. 2015, Hedley et al. 2017, Van Wilgenburgh et al. 2017), and with the rapidly decreasing costs of advanced technology (Hill et al. 2018), their use has already become an important tool in applied field studies (see review for birds in Shonfield & Bayne 2017). Therefore, the description of effective monitoring protocols can be useful to future studies and to improve monitoring programmes. We are aware that our study involved only a single model species, and surveys were carried out only during the breeding season, when detection probability is maximum. Therefore, we propose a minimum protocol for detecting the presence of a songbird species using ARUs. Our protocol is based on five steps that could be easily adapted to other species or contexts: 1) Estimate the effective distance at which the ARU is able to detect the species selected for monitoring; (2) estimate the number of nights/days of monitoring required to detect species presence with high probability; (3) assess the number and cost of devices needed for species detection under real field conditions; (4) limit the recording time to the period when singing activity is at a maximum; and (5) evaluate the cost and time required for data analyses. All recommendations should be assessed before beginning any monitoring programme, to reduce costs, keep the amount of recording time at a minimum and maximise monitoring effectiveness.

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CHAPTER III

Vocal activity rate index: a useful method to infer terrestrial bird abundance with acoustic monitoring

Cristian Pérez-Granados*, Gerard Bota*, David Giral, Adrián Barrero,
Julia Gómez-Catasús, Daniel Bustillo-de la Rosa & Juan Traba

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*Equal contributors



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Vocal Activity Rate (VAR) index: a useful method to infer terrestrial bird abundance with acoustic monitoring

Cristian Pérez-Granados^{1,2*}, Gerard Bota^{3*}, David Giralto^{3,2}, Adrián Barrero¹, Julia Gómez-Catasús¹, Daniel Bustillo-de la Rosa¹, Juan Traba^{1,4}

¹ Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain.

² National Institute for Science and Technology in Wetlands (INAU), Federal University of Mato Grosso (UFMT), Computational Bioacoustics Research Unit (CO.BRA), Cuiabá, Mato Grosso, Brazil.

³ Biodiversity and Animal Conservation Lab. Forest Science and Technology Center of Catalonia (CTFC), Solsona, Catalonia, Spain.

⁴ Research Center in Biodiversity and Global Change (CIBC-UAM), Universidad Autónoma de Madrid, Madrid, Spain.

* Equally contributed

ABSTRACT

Autonomous Recording Units have been widely used in a large number of bird studies in recent years, but challenges remain in estimating abundance based on acoustic monitoring. We tested whether or not Vocal Activity Rate index (the number of songs per unit time for a species), recorded using Autonomous Recording Units, was related to population abundance in two terrestrial bird species, the European Bee-eater *Merops apiaster* and the Dupont's Lark *Chersophilus duponti*. We took recordings at sites where censuses were also carried out to estimate local populations around recorders. We found a positive and significant relationship for the two monitored species. While our results are not conclusive, the strong and significant relationship found for both monitored species suggests that Vocal Activity Rate index may be used to infer bird abundance around recorders in terrestrial species. We describe five logical steps for using the Vocal Activity Rate index with Autonomous Recording Units in other species to guide future studies.

Keywords: Autonomous Recording Units, *Chersophilus duponti*, *Merops apiaster*, passive monitoring, population estimates.

INTRODUCTION

The use of Autonomous Recording Units (ARUs hereafter) for monitoring wildlife has increased widely in recent years (see review in Sugai et al. 2019). ARUs are a suitable alternative to traditional field surveys for detecting species' presence and estimating species richness or population densities (Darras et al. 2018). ARUs have some advantages over traditional field surveys: (1) devices can be deployed and retrieved at any time making fieldwork more flexible and avoiding disturbances to vocal activity, as in human presence surveys (Venier et al. 2012); (2) researchers can cover large spatial and temporal scales simultaneously, eliminating temporal differences between samples, at a relatively low cost (Alquezar & Machado 2015); (3) they can operate unattended in remote locations or areas with limited visibility (e.g., rain forest) to monitor cryptic species (e.g., nocturnal animals) and regardless of weather conditions (Pérez-Granados et al. 2018a); (4) recordings can be automatically scanned avoiding biases due to researcher ability, leading to a greater degree of standardization in data collection (Venier et al. 2012); and (5) ARUs create a permanent and archivable record of surveys, that can provide useful information for future studies and comparison (Alquezar & Machado 2015).

Despite these advantages, there are some obstacles that have hampered the widespread use of ARUs, such as extra costs for acquiring devices. The recent development of open source, low-cost detectors may overcome these cost barriers (Hill et al. 2018). However, the time needed for post-recording analyses and the difficult-to-estimate abundances remain key obstacles to their use (Knight et al. 2017). Advances in computation and automated signal recognition software have led to the development of species-specific recognizers, including machine learning processes, which allow researchers to manage the large volumes of acoustic data recorded (see review in Knight et al. 2017).

Commercial ARUs typically only include one, or at best two, microphones (Hill et al. 2018). Therefore, sound recordings obtained with ARUs provide little or no ability to determine sound direction in three dimensions, hindering the assessment of animal abundance due to the difficulty in mapping individual locations. For this reason, ARUs have been widely used to document community composition or species presence/absence, but scarcely used for monitoring programmes, especially at large spatial scales, due to the difficulty of inferring densities for monitored species (Dawson & Efford 2009). To extend acoustic monitoring beyond activity monitoring, there is a need to estimate the relationship between acoustic activity and

population density (Desjonquères et al. 2018). Different methods have been described and tested to infer population densities from sound recordings (e.g., Dawson & Efford 2009, Hedley et al. 2017). Some authors have used an array of microphones to locate precise sounds, ‘capturing’ time in each microphone allowing the generation of an ‘observed distance’, which can be analysed by conventional distance sampling methods (e.g., Dawson & Efford 2009). Other authors have used complex microphone systems to estimate sound direction with high precision (Hedley et al. 2017). However, the measurement of sound location, even with the use of multiple or complex microphones, is still imprecise and does not directly lead to an estimate of density (Hedley et al. 2017). Sebastián-González et al. (2018) have evaluated a new method to estimate bird population densities by using omnidirectional ARUs. This method requires measuring cue rate from the target species, environmental conditions and an estimate of the distance of the individual to the recorder based on the power of the received sound (Sebastián-González et al. 2018). This study represents an effective method for estimating animal density, but a large number of parameters are needed to develop the method, so this approach is still resource-intensive. Moreover, its implementation is hampered in nocturnal and elusive animal species due to difficulties in obtaining quality recordings at precise distances.

An alternative, rapid and cheap method described to infer population densities using ARUs with omnidirectional microphones is the use of the Vocal Activity Rate index (hereafter VAR), defined as the number of songs per time unit for the target species (Oppel et al. 2014). VAR has been used as an indicator of species’ activity and is expected to increase with population density (density-dependent; Farnsworth et al. 2004, Oppel et al. 2014). Previous studies have confirmed the existence of a positive relationship between VAR and abundance in different taxa, such as mammals or anurans (Nelson & Graves 2004, Barlow & Taylor 2005). In birds, VAR has been used to assess migration intensity (e.g., Farnsworth et al. 2004). Furthermore, that index has been successfully employed to assess changes and estimate population size in seabirds (Buxton et al. 2013, Borker et al. 2014, Oppel et al. 2014). However, Zwart et al. (2014) found no relationship between the amount of vocalisation recorded per ARU and the abundance of the European Nightjar *Caprimulgus europaeus* in the only known study focused on VAR of breeding terrestrial birds.

In this paper, we examined whether VAR, estimated by acoustic monitoring, can be used as an alternative method to infer abundance of two terrestrial bird species, European Bee-eater *Merops apiaster* and Dupont’s Lark *Chersophilus duponti*, with very different singing behaviour,

diurnal activity patterns and habitat selection. We deployed ARUs in areas of known estimated population size for each species to elucidate whether there is a significant relationship between vocal activity rate and the number of individuals around recorders. We predicted that vocal activity rate would increase with population size.

METHODS

Study species and singing behaviour

The European Bee-eater, a summer migrant, is a very vocal species that mostly calls at low frequencies (1-3 kHz) during daytime when foraging or migrating and can be heard at long distances. The contact call (see Appendix Fig. S1) is the most frequent vocalization of the species and it is a short, pleasant rolling sound ('pruuk') uttered for both sexes. It is simply structured, but with some subtle differences between calls or individuals (Valera 2016). The Dupont's Lark is a resident, territorial passerine whose song (see Appendix Fig. S1) ranges from low (<2 kHz) to high frequencies (>6 kHz). Songs are largely shared and repeated between neighbouring males before dawn, when they usually engage in countersigning disputes. Females are presumed to be vocally inactive in this species (Pérez-Granados et al. 2018b). The Dupont's Lark song usually ends with a common sequence, the so-called 'whee-ur-wheeee' (Pérez-Granados et al. 2018b). The use of ARUs has been assessed previously for monitoring the presence of Dupont's Lark (Pérez-Granados et al. 2018a, 2018c).

Study area

The study area comprised sites located in both central and north-eastern Spain. The European Bee-eater recordings were made in 10 different apiaries (composed of 30-50 beehives each) separated by 2-20 km located in farmland areas dominated by irrigated orchards, alfalfa *Medicago sativa* and maize *Zea mays* in western Catalonia (41.31°N 0.55°W, north-eastern Spain). The study area surveyed for the Dupont's Lark comprised 24 breeding sites located in central Spain (40°37'N, 3°09'W, Guadalajara and Soria provinces) and three sites in western Catalonia (45.50°N, 77.76°W, north-eastern Spain). The habitat patches monitored for Dupont's Lark were flat areas dominated by low scrubs or shrubs, such as thyme (*Thymus* spp.), broom (*Genista* spp.) and lavender (*Lavandula* spp.).

Acoustic recording

We used an ARU consisting of a USB Voice Recorder SK-001 with a AC1517D72772-C processor and one integrated omnidirectional microphone. ARUs were built after a cost-effectiveness comparison of five different recording devices (Bota et al. 2017). Recorders were powered by a 12V/8.0 mAh battery (> 300 hour-autonomy), and were also connected to a digital timer that allowed us to program the ARUs to record at selected times and days. Recordings always used a sample rate of 44.1 kHz and 16 bits in stereo mode, and were stored on microSD memory cards capable of storing 520 hours of continuous data. Equipment was protected in easily portable and waterproof boxes (171x100x100 mm) made cryptic by painting them greenish-brown with spots (Bota et al. 2017). These ARUs have previously shown reliability and effectiveness for bird song recording in similar conditions (Pérez-Granados et al. 2018a, 2018c).

Microphones were located in an upward position and horizontally separated by at least 50 cm from natural vegetation in order to minimize any sound blocking (see Appendix Fig. S2). To reduce variation in vocal activity, we limited recording analyses to time periods when singing activity was expected to be highest for each studied species (Oppel et al. 2014). In the case of the European Bee-eater, an ARU was located on each apiary from 15th July to 15th September 2017, a period of maximum presence of the species around apiaries in the study area (Bota et al. 2018). ARUs were programmed to record during the daytime, when individuals were calling while foraging or migrating in the vicinity of the beehives. Each recording was limited to 30 minutes and was taken every 2 hours from 9:15 am to 7:15 pm. Although ARUs were active during two consecutive months, we analysed a subsample of recordings from which we gathered information about European Bee-eater abundance during the recording schedule (see below). Dupont's Lark recordings were carried out by placing one ARU per site. ARUs were programmed to record continuously for 30 minutes starting one hour before sunrise, which is the highest singing activity period for the species. This allowed sufficient time to detect the presence of the species in 95% of cases (Pérez-Granados et al. 2018c). Each site was monitored for one day between 10th April and 15th May 2017.

Acoustic data analyses

Recordings were automatically scanned using Song Scope 4.1.5 (Wildlife Acoustics 2011) which creates a target signal from the feature characteristics of the example songs used for training. This target signal can then be used as a recognizer file for comparison to determine when a sound within a recording matches these characteristics (Wildlife Acoustics 2011). We built

customized species-specific recognizers for each studied species after training by setting parameters (e.g., adjusting sample rate, frequency range, minimum frequency) to isolate targeted signals and reduce the inclusion of false positives, such as background noise or other uninteresting songs incorrectly identified as a target signal. Recordings of each species were always scanned with their own species-specific recognizer and using algorithm 2.0 in Song Scope. We selected results with a score > 40% and quality above 20. The results of automated song recognition were visual and/or acoustically checked by the same observer (CPG) to separate false positives from true positives and calculate the true positive rate, estimated as the number of true positives found within all events detected (Knight et al. 2017). Therefore, posterior analyses about vocal activity rate were only based on true positives (i.e. correct detections made by the recognizer). Recordings were not checked to find targeted songs not automatically detected by recognizers, since this might be more time consuming than identifying songs by manually scanning spectrograms, and we aimed to propose a rapid method to infer relative bird abundances.

We calculated the recall for each recognizer after randomly checking 12 European Bee-eater and nine Dupont's Lark recordings. Recall is an index that represents the proportion of species' calls that were automatically detected and is a typical metric for assessing recognizer performance (Knight et al. 2017). Recall was determined for each recognizer by dividing the number of true positives by the total number of calls uttered by monitored species during the recording (Knight et al. 2017). To estimate the total number of calls uttered by monitored species per recording, an experienced observer (CPG) checked visually and acoustically the selected recordings and annotated the total number of calls of monitored species within them.

Bird data censuses

Methods for estimating bird abundances around ARUs differed among studied species according to counting method and previous tests to assess the distance at which our ARU was able to record vocalisations of the monitored species (Bota et al. 2017, Pérez-Granados et al. 2018c).

We conducted a total of 35 visual censuses of European Bee-eaters in the apiaries coinciding with the ARUs recording schedule (mean number of censuses per apiary = 3.5). European Bee-eaters closer than 100 m to each apiary (and ARU) were counted at 10-minute intervals within each 30-min long recording. We chose that distance because we have estimated that it was the distance at which our ARU was able to detect the European Bee-eater calls (unpubl. data). We

used the mean number of European Bee-eaters counted every 10 minutes during the 30-min counting period as bird abundance, because we considered that mean values are well suited for accounting for normal variations in abundance between intervals.

Dupont's Lark censuses were carried out in the four days after recordings in order to avoid modifying natural singing behaviour while recording. In each of the 27 monitored sites, we performed a census following the line transect method with a 500-m maximum detection band on each side of the observer, within which we assumed a probability of detection equal to 1 for singing males (Pérez-Granados & López-Iborra 2017). The distance of singing males from the observer was estimated acoustically and its location was recorded by GPS. Censuses were carried out by walking at a constant speed (1-3 km) on dry and windless days. Census time was from 60 minutes before dawn to dawn, a period during which the detection probability of the species remains broadly constant (Pérez-Granados et al. 2018b). According to field tests, we considered a 200 m buffer around each ARU to estimate the number of Dupont's Lark males potentially recorded at each ARU, since the probability of detecting the species beyond that distance under favourable singing conditions is always lower than 15% (Bota et al. 2017). Therefore, we used the total number of Dupont's Lark males detected by surveyors within the 200 m buffer around recorders as an index of abundance.

Statistical analyses

VAR was considered as the total number of songs per minute for each species (Garamszegi et al. 2007, Pérez-Granados et al. 2016). To estimate VAR, we divided the total number of true positives automatically detected per recognizer by recording length (Oppel et al. 2014, Zwart et al. 2014). We fitted a linear and logarithmic regression for each of the studied species to estimate the most appropriate relationship between vocal activity rate and abundance (Borker et al. 2014). We used linear regressions because we expected that VAR may increase linearly with abundance (Farnsworth et al. 2004), but also tested logarithmic regressions because signal recognition software may become overloaded under large bird density scenarios (Oppel et al. 2014).

RESULTS

We estimated VAR and bird abundance for 62 different recordings in 2017 (35 recordings and 17.5 hours of recording for the European Bee-eater and 27 recordings and 13.5 hours of

recording for the Dupont's lark). Actual bird abundance extracted from bird censuses around ARUs differed greatly among studied species and sites, and ranged from 0-8 males for the Dupont's Lark to 0-36 individuals for the European Bee-eater.

ARUs detected the species in all cases where at least one individual was censused by human surveyors. The true positive rate differed greatly between studied species. The true positive rate for the European Bee-eater was 97.34 (7,049 calls in 7,241 events detected) and 38.18 for the Dupont's Lark (4,407 calls in 11,542 events detected). Recall rate for the European Bee-eater recognizer was 46.4 % (2,112 calls detected of the 4,555 calls annotated in the validation data set) and 63.0 % for the Dupont's Lark (1,177 calls detected of the 1,868 calls annotated). We estimated a mean number of 12 and 10 minutes for scanning and checking one hour recordings of European Bee-eater and Dupont's Lark, respectively.

We found a positive and significant relationship between VAR and estimated bird abundance for both monitored species. For the European Bee-eater, the linear regression (parameter estimate \pm SE = 0.031 ± 0.004 , $F_{1,34} = 72.54$, $P < 0.001$, $R^2 = 0.68$) was found to be a better fitting model than the logarithmic model ($F_{1,34} = 37.31$; $P < 0.0001$; $R^2 = 0.52$), according to R^2 . For Dupont's lark, logarithmic regression (parameter estimate \pm SE = 1.876 ± 0.163 , $F_{1,26} = 132.70$, $P < 0.001$, $R^2 = 0.84$) was better fitting than the linear model ($F_{1,26} = 73.48$, $P < 0.0001$, $R^2 = 0.74$; Fig. 1).

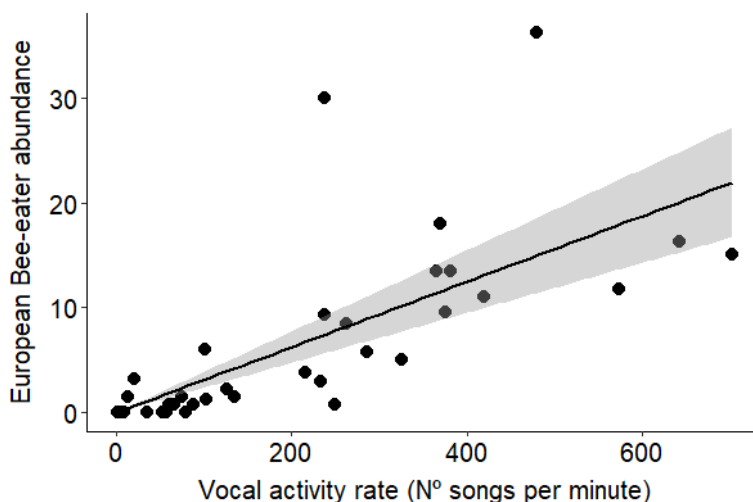


Figure 1a. Relationship between Vocal Activity Rate (number of songs per minute) index and number of European Bee-eaters. The observed values (black dots), fitted regression (black line) and 95% confidence interval (grey surface), are depicted.

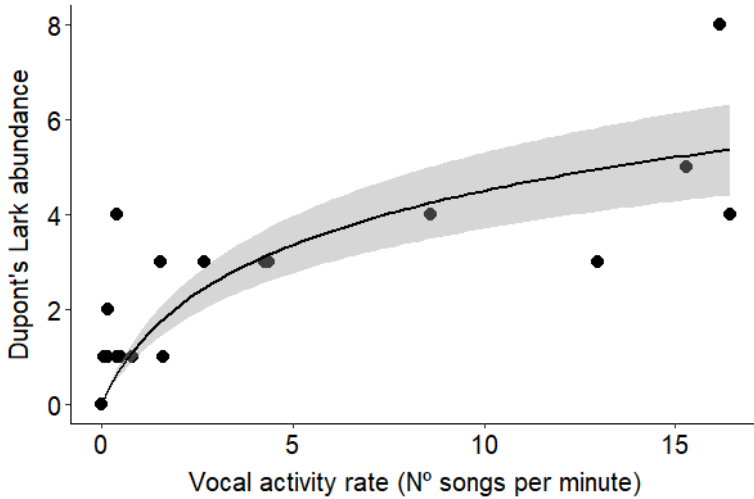


Figure 1b. Relationship between Vocal Activity Rate (number of songs per minute) index and number of Dupont's Larks. The observed values (black dots), fitted regression (black line) and 95% confidence interval (grey surface), are depicted.

DISCUSSION

Our study is the first to explore and to find a positive and significant relationship between VAR and estimated abundance for terrestrial bird species. The strong relationship found for monitored species that suggests that VAR could be a cheap and rapid method to infer an abundance estimate relative to other sampled sites and/or evaluate changes over time from recordings obtained with ARUs using omnidirectional microphones. Our study was focused on terrestrial birds, but the use of the VAR has potential wider applications and could be used, based on a previous assessment, for other acoustically active taxa.

Despite significant relationships found between VAR and estimated bird abundance, we are aware that our results are not fully conclusive in that VAR may not be useful to estimate an accurate absolute abundance for bird species, and that some variation in VAR remains unexplained. Several factors could be responsible for such variation, such as declining detection probabilities with distance of individuals to ARUs. For example, we monitored a site with four Dupont's Lark males within the effective detection radius, but with a very low vocal activity rate (0.4 songs/minute). However, three of the four males were located at greater distances than 160 m from the ARU, a distance at which detectability for detecting the Dupont's Lark can decline by up to 20% (Bota et al. 2017), which may have influenced our results.

As for censuses performed by human observers, VAR results depend on a large number of factors not accounted for in our study, such as breeding seasonality, number of conspecifics, mating success, weather conditions or time of year, among others (Catchpole & Slater 2008) which may compromise the utility of VAR to infer bird abundance from sound recordings. We encourage researchers to record over several monitoring days in order to obtain an averaged VAR per site (Buxton et al. 2013, Oppel et al. 2014), which may help to control the variability in bird singing activity. The relationship between VAR and censused bird abundance can be also affected by observers' ability to detect individuals, as in traditional field censuses. Further research should focus on this topic, including imperfect detection and associated errors when estimating the relationship between VAR and estimated bird abundance.

The creation of a good recognizer is also a key aspect to approximate bird abundances from sound recordings. In this case, recognizers successfully accomplished objectives, despite variations in recall and true positive rate between monitored species. Differences in true positive rate have no influence on estimated relationships since automated species identifications were verified to remove false positives prior to analyses. However, a small true positive rate can preclude the use of recognizers for automated species recognition at large spatial and temporal scales due to the large amount of effort and expertise required to remove false positives. Differences in true positive rate between monitored species may be related to specific conditions under which recordings were taken. In the European Bee-eater, recordings were made on summer days, outside most species' breeding seasons when general bird singing activity is low in temperate zones. On the contrary, Dupont's Lark recordings were taken during dawn choruses in the breeding season. A large number of birds, including several lark species such as the Eurasian Lark *Alauda arvensis* and the Greater Short-toed Lark *Calandrella brachydactyla*, share the dawn chorus with the Dupont's Lark (pers. obs.), which may partly explain the low true positive rate found in this species.

Recall rate may have a great impact or even preclude the use of VAR to infer bird abundances if it is too low. Recall of recognizers is highly variable among species, but in our case they can be considered high when compared to previous studies (e.g., Digby et al. 2013, Shonfield et al. 2018, but see Potamatis et al. 2014 and de Oliveira et al. 2015). The Dupont's Lark recognizer had a higher recall rate than that for the European Bee-eater, which may partly explain the poorer relationship found between VAR and European Bee-eater abundance. In both cases, a large number of calls from long distances may not be well recognized, but plausibly fairly well detected

by the researcher on the spectrogram viewer or when hearing the recordings. In the European Bee-eater case, numerous calls can be made in a continuous manner for a large of group of birds. In those cases, a visual check seems to be more effective for counting calls than automated scanning due to song overlap.

Little is known about the species, study conditions and vocalisations (contact calls, territorial calls, display calls, etc.) for which the VAR may be useful to infer abundances. Previous studies (Oppel et al. 2014, Borker et al. 2014) found a strong and significant relationship for seabird species with high VAR while breeding in their colonies. However, Zwart et al. (2014) did not find any relationship for the European Nightjar, also a nocturnal species, but one that vocalises during short periods while breeding, and which usually sings while flying and foraging. In agreement with those and our results, we believe that candidate species for which VAR might be useful can be identified based on their behaviour and ecology. Candidate species mainly include those living and singing or calling in groups, such as seabirds (Buxton et al. 2013, Oppel et al. 2014) or the European Bee-eater, which suggests that the VAR might also be useful for monitoring other colonial, vocally-active species. VAR can also be used to infer the abundance of territorial birds, such as passerines, at least in those species whose singing behaviour does not vary with population density, such as the Dupont's Lark (Pérez-Granados et al. 2016). Despite that this study was focused on single species, it could be adapted for multiple-species monitoring programmes by scanning the recordings using species classification algorithms. Advances in computations and automated signal recognition, including machine learning processes allow large datasets to be analysed in a timely manner (Stowell & Plumbley 2014).

Here, we propose a protocol to estimate the correlation function between the VAR and the apparent abundance of monitored species. Our proposed protocol is based on five steps that could be easily adapted to other species or contexts: (1) Estimate the effective distance at which the ARU selected is able to detect the songs of the species selected for monitoring; (2) Identify the period of the day at which singing or calling activity of the monitored species is at a maximum and limit the recording time to this period; (3) Assess the performance of the recognizer to be used after evaluating their true positive rate and recall rate; (4) Estimate the VAR in a number of sites (at least 20-30) and correlate it with estimated abundance of the species within an effective distance from the ARU; and (5) Evaluate the strength of this correlation and the cost and time required for estimating bird abundance using this method. All of these steps should be assessed before using the VAR in any monitoring programme, but once they have been validated, the use

of VAR should be a rapid method to infer bird abundance in new recordings. VAR can be useful for current monitoring programmes, but it may also be estimated retroactively to assess bird abundances and changes in population trends in archived recordings, which could be especially useful for long-term monitoring programs aimed at analysing wildlife population trends. The recent development of open source, low-cost ARUs, together with the increase of citizen-science projects and the establishment of new robust relationships between VAR and species abundance, present a range of new opportunities for wildlife population monitoring.

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CHAPTER IV

Acoustic monitoring of diurnally migrating European Bee-eaters agrees with data derived from citizen science

Gerard Bota, Juan Traba, Francesc Sardà-Palomera,
David Giral & Cristian Pérez-Granados

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Acoustic monitoring of diurnally migrating European Bee-eaters agrees with data derived from citizen science

Gerard Bota¹, Juan Traba^{2,3}, Francesc Sardà-Palomera¹, David Giral¹, Cristian Pérez-Granados^{2,4}

¹ Biodiversity and Animal Conservation Lab. Forest Science and Technology Center of Catalonia (CTFC), Solsona, Catalonia, Spain.

² Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain.

³ Research Center in Biodiversity and Global Change (CIBC-UAM), Universidad Autónoma de Madrid, Madrid, Spain.

⁴ National Institute for Science and Technology in Wetlands (INAU), Federal University of Mato Grosso (UFMT), Computational Bioacoustics Research Unit (CO.BRA), Cuiabá, Mato Grosso, Brazil.

ABSTRACT

The use of Autonomous Recording Units (ARUs) for wildlife monitoring has increased in recent years. Acoustic monitoring has been used for a wide range of research topics, but it has rarely been used for monitoring wildlife migrations. In this work we evaluate the use of acoustic monitoring to characterise the diurnal migration pattern of a bird species, the European Bee-eater *Merops apiaster*. We set up 3–4 acoustic monitoring stations daily from 11 August to 21 September 2017 in north-eastern Spain, during post-breeding migration of the species. We used the Vocal Activity Rate (VAR), defined as the number of calls per unit time, as an index of Bee-eater abundance to describe the daily and seasonal migration pattern of the species. We also assessed the relationship between daily mean VAR estimated by ARUs, with citizen science data uploaded to the platform Ornitho.cat over a large spatial scale. According to mean VAR, intensity of migration increased weekly until the last week of August when it peaked, with species abundance decreasing to the lowest values in the study area by late September. A significantly higher number of calls was detected in the first and last hours of the day. Our results agree with previously published seasonal and daily migration patterns described for the species. VAR was significantly, positively correlated with the percentage of citizen science records of Bee-eater uploaded to complete checklists, used as an independent source to compare migration timing. Overall, our results, validated through citizen science data, show that acoustic monitoring can provide an effective complementary for monitoring the bird migration of vocally active species. The use of ARUs may help to improve our understanding of migratory behaviour and be useful for a wide range of purposes.

Key words: ARUs, automated recognition, citizen science, *Merops apiaster*, migration, Ornitho, Vocal Activity Rate.

INTRODUCTION

Bird migration has attracted the attention of ornithologists for centuries (e.g., White 1788, Irby 1875). Counting birds during migration offers the possibility of monitoring population sizes (Arroyo et al. 2016), estimating population trends (Møller et al. 2008) and evaluating changes in migratory behaviour related to different threats (De Lucas et al. 2004, Lindström et al. 2010) as well as to climate change (Saino et al. 2010). Techniques commonly applied in monitoring bird migration are usually costly in terms of time, human resources and equipment. They may require long-term intensive observation sessions at many sites (e.g., Martín et al. 2016, Miller et al. 2016), trapping sessions carried out by experts to deploy monitoring devices such as geolocators or GPS tracking systems (Bridge et al. 2013, Sperger et al. 2017), several years of ringing sessions for mark-recapture studies (Bairlein 2001) or feather collection for stable isotope analysis of geographic origins (Hobson et al. 2015). In recent years, the analyses of bird observations submitted to citizen science platforms (eBird, Ornitho, Observado, etc.) by volunteer birders have shown to be a useful tool for improving our understanding of bird migration (Hurlbert & Liang 2012, Newson et al. 2016, Schubert et al. 2019). Recent initiatives such as EuroBirdPortal even allow for the observation of bird migration at the European level in near real-time thanks to the integration of data from different citizen science platforms (Gargallo 2017). These platforms have already been used to examine the timing of migration of specific bird species across a large geographic area and this wealth of data provides novel opportunities to unveil patterns of bird migration at large spatial and temporal scales (Sullivan et al. 2009, 2014).

Despite its enormous potential, an important part of citizen science data is subject to certain limitations derived from different sources: differences in detectability between species, different bird-identification skills among observers, unstratified data sampling (which may cause geographic biases in the spatial distribution) and non-homogeneous distribution of the birding community across regions have been described as potential biases associated with this kind of data (Ferrer et al. 2006, Sullivan et al. 2009, Johnston et al. 2018).

The development of non-invasive techniques for monitoring bird migration is desirable, in order to avoid capturing individuals or having to carry out intensive field work. To this end, studies based on horizontally scanning weather radars or vertical wind profilers, among other types, have been used in different regions for monitoring bird migration (Weisshaupt et al. 2018, Horton et al., 2020). This technique can provide information on density, direction, speed and altitude of migrating birds but cannot describe the species involved (Salamon et al. 2016). Among

the non-invasive techniques most commonly deployed for monitoring wildlife in recent years is the use of Autonomous Recording Units (ARUs hereafter; see review in Sugai et al. 2019). This technique is a consistent and suitable alternative to traditional field surveys to estimate parameters such as species presence, abundance or richness (Oppel et al. 2014, Darras et al. 2018, Pérez-Granados et al. 2019a). Acoustic monitoring has already been used to study the migration of different animal groups, such as whales (Burnham & Duffus 2020), bats (Johnson et al. 2011) and birds. Acoustic monitoring of flight calls during the night has proven to be an effective method to provide information on migration routes, timing and relative migration intensity of birds (Larkin et al. 2002, Farnsworth et al. 2004, Farnsworth & Russell 2007, Sanders & Mennill 2014). Some authors have proposed that ARUs could be used to characterize the migration pattern of specific bird species (Salamon et al. 2016) and recent studies have demonstrated their potential to detect the arrival and departure of birds (Oliver et al. 2018, Pérez-Granados & Schuchmann 2020a). As far as we know, such methodology has never been used for monitoring the migration pattern (hours, peak of migration) of a diurnal bird.

In this paper, we evaluate the utility of acoustic monitoring based on ARUs coupled with automated signal recognition as a tool for monitoring the diurnal post-breeding migration of a vocally-active bird species, the European Bee-eater *Merops apiaster* (Bee-eater hereafter). Pérez-Granados et al. (2019a) have recently described a strong and significant relationship between the Vocal Activity Rate index (VAR hereafter; i.e. number of calls detected per time unit) and the number of Bee-eaters flying within a radius of 100 m around ARUs. Therefore, we used the VAR as an index of the abundance of Bee-eaters for describing the daily and seasonal pattern of migration. We also aimed to assess whether the seasonal migration pattern described by using acoustic monitoring was in accordance with the timing of migration obtained with citizen science data, in order to provide a qualitative independent comparison to our acoustic monitoring method. We expected that migration timing obtained thanks to citizen science data would be correlated with that estimated using ARUs.

METHODS

Study species

The European Bee-eater is an insectivorous long-distance migrant bird that breeds in southern Europe, North Africa and western Asia and winters in sub-Saharan Africa (del Hoyo et al. 2001). Western European breeders migrate during the day using a broad front on a western route to non-breeding regions in West Africa (Hahn et al. 2020). Median migration speed during post-

breeding migration is around 159 to 206 km/day until the first sub-Saharan site, depending on the breeding origin (Hahn et al. 2020). Groups are generally stable during migration (Dhanjal-Adams et al. 2018) and this species is known to use both flapping and soaring-gliding flight while migrating (Sapir et al. 2011).

The Bee-eater is a richly coloured bird and is a very vocal species that usually calls while foraging or migrating in flocks (Cramp & Simmons 1985), making it a highly detectable and easily identifiable species for the bird-watching community. The contact call, uttered at low-frequencies (1–3 kHz; Figure S1), is the most frequent vocalisation of the species and it is a repeated, soft but abrupt rolling “prüt”, given in chorus from flying flocks and reaching long distances (Mullarney et al. 1999). Species detectability is a key point in bird monitoring techniques (Sanz-Pérez et al. 2020), and easily detected birds are reported more frequently as well as having less bias than cryptic species in citizen science platforms (Sullivan et al. 2009). We selected the Bee-eater as a study species due to its high detectability, easily identifiable characteristics (factors described as potential biases in citizen science data), because it is vocal while migrating and because of previous studies testing the use of the VAR of the species as an index of Bee-eater abundance (Pérez-Granados et al. 2019a).

Study area

The study area was comprised of a network of nine acoustic monitoring stations placed in a semi-arid, relatively flat agricultural landscape on the eastern edge of the Ebro Valley (41°46'N, 0°46'E) in western Catalonia (Lleida province, NE Spain). Monitoring stations were located within an approximate area of 1000 km² and were separated by 11.1 ± 1.1 km (mean \pm SD).

The study period was from 11 August to 21 September 2017. We consider this period long enough to assess the utility of ARUs for monitoring bird migration and detecting changes in Bee-eater abundance as it matches the main period of the post-breeding migration described for the species in the study area (Muntaner et al. 1983). Acoustic monitoring stations were placed at apiaries (Figure 1), since Bee-eaters usually use beehives as feeding stopovers during migration (Yosef 2004). Previous studies also used apiaries as monitoring stations to study the migration strategy of the species (Yosef et al. 2006). All selected apiaries were composed of 30 to 50 beehives which remained active throughout the study period.

Collection and analysis of acoustic data

During the study period, three to four acoustic monitoring stations were active daily. We placed one ARU per active station, which was systematically and fortnightly shifted between the nine selected apiaries. Each ARU consisted of a USB Voice Recorder SK-001 with processor AC1517D72772-C and one integrated, single-channel microphone (Pérez-Granados et al. 2019b). ARUs were powered by a 12V/8 mAh battery (300-h autonomy), and were started and stopped by an electronic timer. Equipment was protected in easily portable, weatherproof plastic boxes (60×80×160 mm), camouflaged by painting them greenish-brown. ARUs were located on the ground with the microphone in an upward position with no blocking barriers within at least 50 cm (Rempel et al. 2013; see Figure 1). ARUs were programmed to record continuously for 30 minutes at the following local (GMT+2) recording times: 9:15, 11:15, 13:15, 15:15, 17:15 and 19:15. We used a sample rate of 44.1 kHz and 16 bits for all recordings. We estimated that the recorder was able to detect the Bee-eater calls up to 100 m (Pérez-Granados et al. 2019a). Recordings were collected in mp3 format on microSD flash cards (100-h memory) and needed a post-transformation to wav format prior to analysis. Although compression into mp3 format affects the spectral and temporal composition of the signal (Obrist et al. 2010), and its conversion to wav format does not improve recording quality, this should not influence the signal recognition results (Rempel et al. 2005).



Figure 1. One of the locations of beehives with an autonomous recording unit (on the ground in the foreground) used for monitoring European Bee-eater migration in North-eastern Spain (photo Joan Rodriguez, 29 June 2017).

Recordings were analysed automatically using the freely available software Song Scope v. 4.1.5 (Wildlife Acoustics 2011). This software has proven to be one of the most efficient in automated song recognition (Knight et al. 2017). Song Scope is able to create a target signal from the characteristics of the example signals used for training, which is used as a recognizer file to compare when a sound within a recording matches its characteristics (Waddle et al. 2009). The automated signal recognition software and species-specific recognizer were the same as those in Pérez-Granados et al. (2019a), when they assessed the significant relationship between VAR and flying Bee-eaters around recorders. Specifically, the recall of the recognizer, estimated as the proportion of Bee-eater calls detected by the recognizer divided by the total number of Bee-eater calls on sound recordings, was 46.4%. The recall value is highly variable among species and software employed and ours can be considered acceptable when compared with previous studies (see discussion about the recall rate of the recognizer employed in Pérez-Granados et al. 2019a). Moreover, the recognizer built for Pérez-Granados et al. (2019a) was trained using Bee-eater calls recorded in the study area and using the same ARU, so the relationship found in that previous study should be valid for this one. In order to select recordings with different background noise and Bee-eater abundance, we randomly selected 120 30-min recordings (16% of total sample) in which all events reported by the recognizer were visual and/or acoustically checked, always by the same observer. To evaluate automatic signal recognition, we calculated the precision of the recognizer as the proportion of true Bee-eater calls detected by the recognizer divided by the total number of sounds classified as Bee-eater calls by Song Scope (Knight et al. 2017).

Bird citizen science data

We used the online citizen science platform Ornitho.cat (hosted by the Catalan Institute of Ornithology, available at www.ornitho.cat) as a source of quantitative independent data on migration timing of the Bee-eater during the study period and area. We used this platform because Ornitho.cat was the citizen science platform with the highest number of Bee-eater observations during the study period in the area and as it is also the most used platform by local birdwatchers (5,629 registered users on 17 December 2019).

Citizen-science data quality is, among others factors, influenced by the number of observations (La Sorte et al. 2014, Sullivan et al. 2014). In order to maximize the number of observations available for analysis and to cover the entire study period, and given that Bee-eater migration follows a broad front in the region, we conducted the analyses using all uploaded complete checklists (all species observed reported) within the region of Catalonia (31,895 km²) rather than

those uploaded from the study area only (52 complete checklists were available for the study area with no published lists for some of the monitored weeks). We extracted 519 complete checklists with presence of Bee-eater from 11 August to 21 September 2017 in Catalonia (consulted on 10 October 2019). Due to the daily variation in the number of active birders and considering that it may bias the number of Bee-eater observations uploaded, we also extracted all complete checklists per day ($n = 1535$) as a measure of daily birding effort in the region.

Statistical analyses

In order to elucidate whether migration patterns of Bee-eaters depend on the time of day and vary seasonally, we fitted a Generalized Linear Mixed Model (GLMM) with Gaussian distribution error and an identity-link function, using VAR of each recording as response variable and recording time (six categorical levels) and migration week (six categorical levels) as fixed effects. Recording day and acoustic monitoring station were included as random effects. When a fixed effect was found to be significant, a Tukey's post hoc test was performed to assess whether there were differences among levels. We also estimated the Spearman rank correlation between mean VAR of all acoustic stations per day (hereafter mean VAR per day) and corresponding percentage of complete checklists with presence of Bee-eaters in relation to all complete checklists uploaded for the same day. This variable has already been used to define bird migration timing patterns using citizen science data (Sullivan et al. 2009).

All statistical analyses were performed with R v. 3.4.1 (R Core Team 2016) or Statistica v. 10 (Statsoft 2011). We used the packages 'lme4' (Bates et al. 2015) for the GLMMs, 'lmerTest' (Kuznetsova et al. 2017) to calculate the significance of fixed effects and 'multcomp' (Hothorn et al. 2008) for post hoc comparison tests.

RESULTS

We collected 763 30-min recordings (381.5 h of recording) and a total of 223,479 Bee-eater calls were automatically detected. The average number of 30-min recordings per week across stations was 127 ± 35 (\pm SD). The precision of the recognizer was 99.2% (23,537 Bee-eater calls in 23,719 calls automatically detected). We therefore decided to consider all events identified by Song Scope as Bee-eater calls.

VAR differed with week and recording time (Table 1). A smaller VAR was detected during the first two weeks (11 to 24 August) and the last week of the study period (15 to 21 September),

when compared to the central weeks (Table 1, Figure 2). Significantly more Bee-eater calls were detected during the late afternoon (19:15) in relation to the rest of the recording times (Figure 3). The first hours of the day (9:15) and afternoon (17:15) also presented a significantly higher number of calls than midday hours (Figure 3).

Mean VAR per day was significantly and positively correlated with the percentage of complete checklists with presence of Bee-eater ($r_s = 0.65$, $P < 0.001$; Figure 4). Weekly patterns of mean percentage of complete checklists including Bee-eaters uploaded to Ornitho.cat followed a similar pattern to the one described by ARUs (Figure 2). However, the highest peak of VAR observed in the last week of August was not so clear in the migration pattern obtained using citizen science data.

Table 1. Summary table of the results of a Generalized Linear Mixed Model (GLMM) testing the relationship between Vocal Activity Rate of European Bee-eaters (number of calls detected per recording) and the time of the day (Hour) and time of the season (Week). Recording day (Day) and acoustic monitoring station (AMS) were considered as random effects. Total number of recordings: 763.

Fixed effects					
Variable	Sum. Sq.	Sq. Mean	F-value	df	P(>F)
Hour	9,329,214	1,865,843	34.316	5	<0.001
Week	1,186,129	237,226	4.363	5	0.002
Random effects					
Group name	Variance	Std. Dev			
Day (Intercept; $n = 42$)	1686	41.06			
AMS (Intercept; $n = 9$)	17154	130.97			
Residual	54373	233.18			

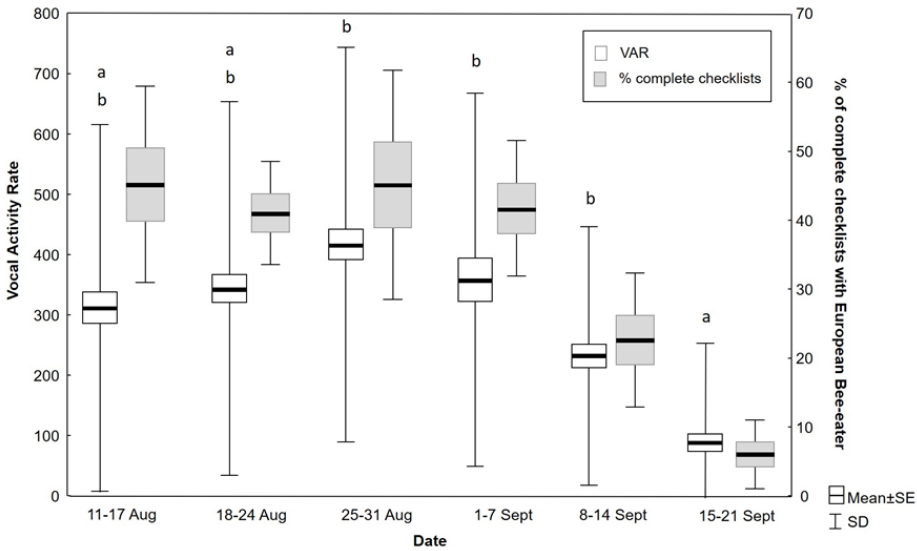


Figure 2. Weekly migration pattern of the European Bee-eater in the study area during the study period. Boxplots showing mean, SE and SD of Vocal Activity Rate (VAR) per week (grey boxes) for all active acoustic monitoring stations and percentage of complete checklists including European Bee-eaters uploaded to Ornitho.cat (empty boxes). Letters on the top show significant differences in hourly VAR between migration weeks from Tukey's post hoc test.

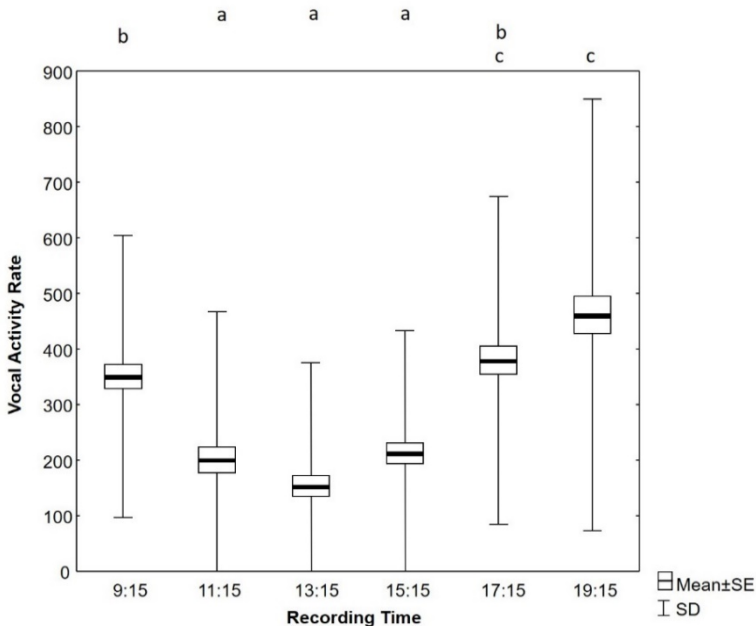


Figure 3. Daily migration pattern for European Bee-eaters in the study area during the study period. Boxplots showing mean, SE and SD of Vocal Activity Rate (VAR) detected during each recording time period. Letters on the top indicate significant differences between recording times from Tukey's post hoc test

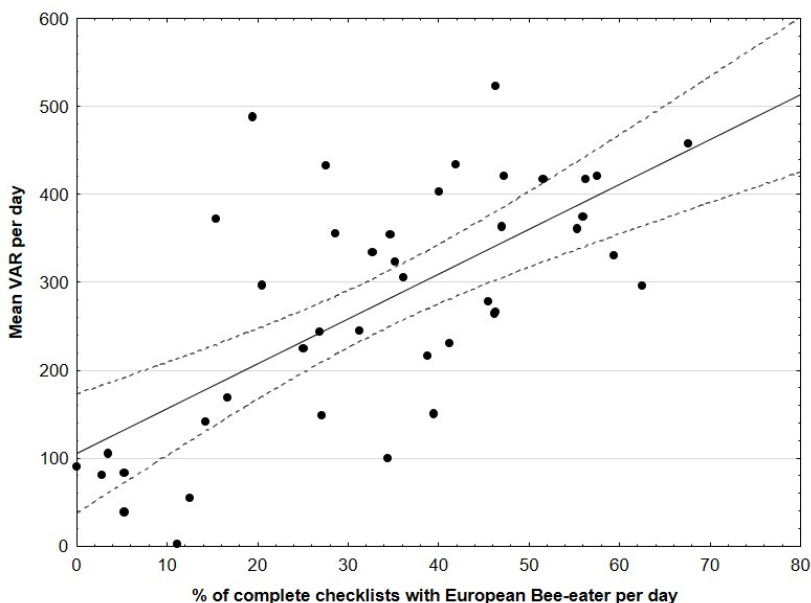


Figure 4. Relationship between mean Vocal Activity Rate (VAR) per day and percentage of complete checklists with presence of European Bee-eaters per day ($r_s = 0.65$, $P < 0.001$). Linear regression line is shown in black, and 95% Confidence Intervals with dashed lines.

DISCUSSION

Our study is the first to use acoustic monitoring to provide accurate information about post-breeding migration timing for a diurnal bird species. The maximum number of calls were detected during the last week of August and afterwards decreased every week until the third week of September, when very low levels were recorded. Our findings on the seasonal pattern of European Bee-eater migration found through the use of ARUs agree with the one described by Muntaner et al. (1983), who also identified the last week of August as the one with the largest number of Bee-eaters migrating over Catalonia. The highest migration peak observed in the last week of August by acoustic monitoring was not so clearly observed in the pattern obtained using citizen science data. Previous studies have stated that Bee-eaters increased their presence in apiaries during and after rain (Glaïim 2014, Moreno-Opo et al. 2018). During the last week of August there were three different rainy days in the study area (Meteocat 2017a). The third week of August accounted for only one rainy day and the first week of September two days (Meteocat 2017a, b). These rainy episodes during migration peak may partly explain the larger presence of Bee-eaters detected by acoustic monitoring around apiaries.

Acoustic monitoring provides robust and continuous information about the daily migration pattern and use of apiaries as stopovers by European Bee-eaters. Daily migration was concentrated primarily in the late hours of the day and secondarily in the early hours. This result is in accordance with those obtained by Moreno-Opo et al. (2018) in western Spain, who found less Bee-eater interactions at apiaries during the middle of the day. Prior studies that focused on the post-breeding migration of the Bee-eater have also found a larger number of birds crossing the Strait of Gibraltar during the first and the last hours of the day (López-Gordo 1975, Tellería 1979). The Bee-eater flight mode is based on flapping but also on soaring-gliding and is highly influenced by atmosphere conditions (Sapir et al. 2010, 2011). The species usually flies higher than 500 m above ground level when using thermals during midday (Yosef et al. 2006). In such cases, birds would not be detected by the recorder which could partly contribute to the lower number of calls detected during the central hours of the day.

According to our prediction, mean VAR per day, as an indicator of bird abundance, was positively correlated with the percentage of complete checklists with presence of Bee-eater coming from a citizen science platform, an independent way to estimate migration timing. Despite differences in spatial scale covered by both methods, our results are in agreement with a previous study that also found positive correlations between data from acoustic monitoring and data from more traditional migration study techniques (Sanders & Mennill 2014). Therefore, we believe that acoustic monitoring data can be a useful tool for monitoring diurnal migration of the European Bee-eater. These results also opened up new perspectives on the use of ARUs for monitoring other diurnal, vocally-active migrating species. Our results are in agreement with prior studies carried out on nocturnal migrants that have proven the functionality of using acoustic monitoring for measuring timing and relative bird migration intensity (Larkin et al. 2002, Farnsworth & Russell 2007, Sanders & Mennill 2014, Salamon et al. 2016).

Acoustic monitoring could be used to assess the timing of migration or the impact of local weather conditions on bird migration (Sapir et al. 2011, La Sorte et al. 2014). This methodology might also be a good cost-benefit alternative for monitoring bird migration in inhospitable or difficult to access, remote areas, such as high mountain passes where bird migration can be bottle-necked in relatively small areas (Komenda-Zehnder et al. 2010, Williams et al. 2011). In addition, acoustic monitoring allows for longer and standardised daily time series of data, probably at a lower cost than human visual counts.

The European Bee-eater is a common and highly vocally active bird species that uses beehives intensively while migrating (Yosef et al. 2006), which enabled us to collect a large amount of data with relatively little effort. We also might expect better performance of passive monitoring acoustic in species like the Bee-eater that concentrate together during migration and perform flocking behaviour. Future studies aiming to monitor other bird species may require a basic knowledge of the migratory strategy of the study species (e.g., preferred habitats, priority migratory pathways and timing) to design monitoring protocols able to collect a satisfactory amount of data for detecting daily and seasonal changes. Furthermore, easy-to-identify species, such as European Bee-eaters, are reported more frequently on citizen science platforms (Sullivan et al. 2009); while lower temporal correlations between VAR and observations uploaded to platforms might be expected for other, less conspicuous, bird species.

The recent development of low-cost ARUs, machine learning processes for detecting bird vocalizations and the possibility of remotely transmitting data (Beason et al. 2018, Hill et al. 2018, Sethi et al. 2018), open the doors for implementing networks of recorders at a relatively low cost. Furthermore, with the rapid development of automated signal recognition software (Knight & Bayne 2018, Stowell et al. 2018), a set of sound recordings coming from acoustic monitoring could be used for describing migration timing and migration intensity for a different number of species at a single time (Pérez-Granados & Schuchmann 2020b). In conclusion, we believe that acoustic monitoring of diurnal flight calls of migrating birds can be an effective, alternative or complementary technique for monitoring migration of vocally-active species, such as different species of cranes, finches, swallows, swifts or wagtails, among other bird groups.

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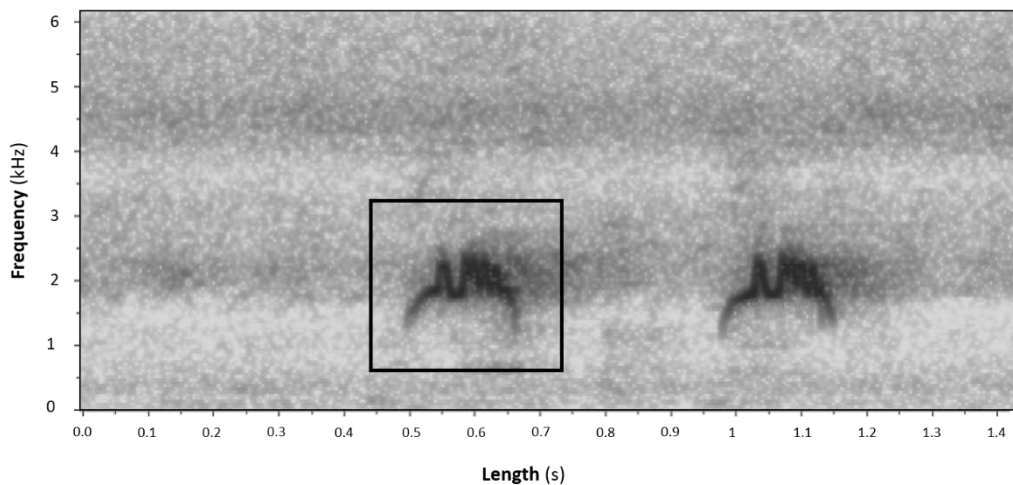
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APPENDIX 1

Supplementary material S1. *Sonogram of a typical European Bee-eater call. The rectangle shows a typical call of the species, which was used for building a species-specific recognizer in the Song Scope software, because of its very particular, recognizable and consistent characteristics.*



CHAPTER V

New technologies for monitoring human-wildlife conflicts: evaluation of passive acoustic monitoring for measuring European Bee-eater predation pressure at beehives

Gerard Bota, Juan Traba, Francesc Sardà-Palomera,
David Giralt & Cristian Pérez-Granados

Ecological indicators (submitted)



New technologies for monitoring human-wildlife conflicts: evaluation of passive acoustic monitoring for measuring European Bee-eater predation pressure at beehives.

Gerard Bota¹, Juan Traba^{2,3}, Francesc Sardà-Palomera¹, David Giralt¹ & Cristian Pérez-Granados^{2,4}

¹ Biodiversity and Animal Conservation Lab. Landscape Dynamics and Biodiversity programme. Forest Sciences and Technology Centre of Catalonia (CTFC), Solsona, Catalonia, Spain.

² Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain.

³ Centro de Investigación en Biodiversidad y Cambio Global (CIBC-UAM), Universidad Autónoma de Madrid, Madrid, Spain.

⁴ Ecology Department, Alicante University, Alicante, Spain.

ABSTRACT

In a context of human-wildlife conflict, it is crucial to develop accurate protocols for the reliable verification of the causative species and its relationship with potential damage claims. One of such conflicts is that occurring between apiarists and bee-eaters. Bee-eaters hunt honeybees at the apiaries and potentially can reduce their foraging activity. Although some methodological approaches had been developed (mainly based on human direct counts) to monitor the predation pressure exerted by bee-eaters on honeybees, they are costly in time and human resources to obtain continuous data over time and in different places. In this work, we aim to assess the utility of passive acoustic monitoring (PAM) as an efficient methodology to measure European bee-eater (*Merops apiaster*) predation pressure at beehives and its impact on honeybees foraging activity. Using Autonomous Recording Units (ARUs) in apiaries, coupled to automated recognition methods for bee-eater calls identification, we found a positive relationship between Vocal Activity Rate (VAR) index and number of bee-eaters hunting attempts on honeybees. So, we considered VAR as a good estimator of bee-eater predation pressure around apiaries. We also found that VAR varied over time, showing a lower predation pressure during midday hours and higher during the post-breeding migratory period. Honeybees flying activity was negatively associated with VAR and this relationship was conditioned by the hour of the day. Our study offers a new application of PAM for the evaluation and quantification of potential damages caused by wildlife, a key element for the resolution or mitigation of human-wildlife conflicts. We focused on the interaction between honeybees and the European bee-eater, but we expect that this technique might be useful also to remotely monitor the predation risk or potential damage pressure caused by other vocally active species to human activities.

Keywords: ARUs, Vocal Activity Rate, apiculture, *Merops apiaster*,

INTRODUCTION

Interactions between wildlife and humans have occurred for millennia (Graham et al. 2005, Redpath et al. 2015), but their frequency has widespread in the last decades due to the exponential increase in human population and the resultant expansion of human distribution range and activities (Sanderson et al. 2002, Anand & Radhakrishna 2017). Negative interactions have usually been termed as human-wildlife conflicts (Graham et al. 2005) and arises due to competition between humans and wildlife for shared and limited resources (Young et al. 2010, Redpath et al. 2015). Human-wildlife conflicts are especially problematic when shared resources have economic value (Manral et al. 2016) and they are exacerbated when the conflicting species are protected or charismatic (Peterson et al. 2010, Athreya et al. 2011). As a result of these conflicts, some wildlife species have been catalogued as pests or human competitors, and historically a great effort has been dedicated to restrain their populations (Thirgood 2000, Yodzis 2001).

One of such human-wildlife conflicts is that occurring between apiarists and bee-eaters (Family Meropidae). On one side there is the apicultural industry, which has a big worldwide economic value producing honey, beeswax and other bee-related products from European honeybees (*Apis mellifera*) (e.g. Lee et al. 2010, Chauzat et al. 2013). In Europe, apiculture sector produced 280.000 tonnes of honey in 2018, making the EU the second largest honey producer after China, and being Spain the first EU country in number of beehives (EU Commission 2019). On the other side there are the bee-eaters, which are generally protected bird species characterized for their ability to hunt flying insects, especially hymenopterans, including honeybees (Fry 2001). Honeybees can constitute an important part of European bee-eater (*Merops apiaster*; hereafter bee-eater) diet depending on the geographical area and season, becoming between one third and one half of the total Hymenoptera predated by bee-eaters (Galeotti & Inglisa 2001, Yosef 2004, Arbeiter et al. 2014, Farinós-Celdrán et al. 2016). Bee-eaters hunt almost entirely flying, either by making short flights from an elevated perch or by hunting in continuous flight, while uttering characteristic contact calls (Fry 2001). Bee-eaters seem to preferably predate on domestic honeybees (Inglisa et al. 1993, Galeotti & Inglisa 2001) due to its high nutritional value, short handling time, and because it is a stable and predictable food source around beehives (Krebs & Avery 1985).

Beekeepers of Mediterranean basin and Africa have pointed bee-eaters as a pest for apiculture, jeopardizing honey production and honeybee colonies vitality (Galeotti & Inglisa 2001, Alfallah et al. 2010, Farinós-Celdrán et al. 2016, Langowska et al., 2018, Floris et al. 2020). The impacts of bee-eaters on honeybees have been relatively well-studied in Europe, and rely on both direct predation, and through the inhibition of honeybees normal flying behaviour due to the high

predation pressure around the apiary (e.g., Fry 1983, Galeotti & Inglisa 2001, Farinós-Celdrán et al. 2016, Moreno-Opo et al. 2018).

Even though the presence of bee-eaters around beehives is unquestionable, their direct negative impact on beehives is controversial. Several authors have pointed out that predation of honeybees by bee-eaters is usually far below the regeneration threshold of a single honeybee colony, and thus their impact on colonies' dynamic might be negligible (Fry 1983, Laplaza & Albero 1997, Alfallah et al. 2010, Farinós-Celdrán et al. 2016). Flight inhibition cause losses in beekeeping economy by reducing beehives' production (Galeotti & Inglisa 2001, Langowska et al. 2018), although this phenomenon seems to be variable in its magnitude between regions (Langowska et al. 2018, Moreno-Opo et al. 2018). While the extent of the impact of bee-eaters predation pressure on honeybees' production is still under discussion, the perception by beekeepers is clearly contrary to the presence of bee-eaters around beehives. Even though it is a protected species in many countries, illegal killing of bee-eaters has been often reported (e.g. Woldheck 1979, Galeotti & Inglisa 2001). In this context, the public administration of some countries, such as Italy or Spain, has economically compensated to beekeepers because of the damage caused by bee-eaters (Galeotti & Inglisa 2001, Villero et al. 2017).

In a context of human-wildlife conflict, it is crucial to develop accurate protocols for the reliable verification by the authority of the causative species and its relationship with damage claims to create public trust in the legitimacy of compensation programs (López-Bao et al. 2017). Verification protocols are essential since self-reporting of wildlife damage may result in overestimates of damage or fraudulent claims (Nyhus et al. 2005). In the case of bee-eaters conflict, a feasible, accurate and easy to use methodology is needed to estimate bee-eater predation pressure at beehives, on which a potential public compensation or mitigation program may be based. Verification is a vexing problem for many human-wildlife compensation programs because evidence from wildlife that can cause harm can be difficult to find and/or quickly disappear (Nyhus et al. 2005). In the case of bee-eaters conflict, this problem is further exacerbated as no direct identifiable trace of potential harm remain.

Some methodological approaches based on direct bird counts or developing bee-eater distribution models together with land apicultural suitability models have been implemented for compensating damage claims (Villero et al. 2017, Moreno-Opo et al. 2018). However, they are costly in time and human resources to obtain continuous data, and/or they are unable to measure the real presence of the species in a specific place and time. Passive acoustic monitoring (PAM), based on the placement of unattended and programmable Autonomous Recording Units (ARUs hereafter), is a recent technique commonly used for terrestrial wildlife monitoring (see review in Sugai et al. 2019). ARUs have been found a suitable alternative to traditional field surveys for detecting birds' species presence or estimating bird species richness or population

densities around recorders (e.g., Darras et al. 2018, Pérez-Granados & Traba 2021). The Vocal Activity Rate index (VAR; number of vocalizations detected per unit time of recording) is the most often applied and validated method for estimating bird abundance using PAM (see a review in Pérez-Granados & Traba 2021). VAR is based on the assumption that number of vocalizations on recordings is associated to number of individuals vocalizing around recorders (Pérez-Granados & Traba 2021). Indeed, the use of ARUs coupled with automated signal recognition software has proven to be able to infer abundance of bee-eaters flying around beehives and to define the migratory pattern of the species (Pérez-Granados et al. 2019a, Bota et al. 2020). These findings suggest that VAR might be also useful to provide a reliable estimation of bee-eater predation pressure at beehives, which might be used as an index for compensating damage claims.

Here, we aim to assess the utility of PAM as an efficient tool to measure bee-eater predation pressure at beehives and to assess its impact on honeybees foraging activity. Our first goal was to assess whether the VAR (number of bee-eater calls per recording) can be a good estimator of bee-eater predation pressure in beehives by assessing the relationship between VAR and the number of bee-eater hunting attempts estimated by human surveyors at different spatial scales. Based on prior findings, we expected that VAR will show a positive relationship with bee-eater predation pressure (Pérez-Granados et al. 2019a).

We also aimed to describe the daily and seasonal patterns of bee-eaters predation pressure at beehives as it may be important to identify when it might be most necessary to apply potential damage prevention measures. Finally, we aimed to evaluate whether the bee-eater predation pressure has an impact reducing the flying activity of honeybees, since it is one of the main mechanisms behind the losses in the beekeeping economy. We expected that bee-eaters will have a negative effect on honeybees activity.

MATERIAL AND METHODS

Study area

The study area comprised a flat farmland area situated on the eastern edge of the Ebro Valley (41°46' N, 0°46' E), in western Catalonia (Lleida province, NE Spain). The study was conducted in 22 different apiaries separated by a minimum distance of 2 km covering around 900 km² (Supplemental Fig. S1). Each apiary comprised between 30-50 beehives and remained in the same place during the study period. Beekeeping activity in the study area is characterized by the exploitation mainly of summer flowering of alfalfa (*Medicago sativa*) fields, and some mass-flowering crops like sunflower (*Helianthus annuus*).

Field work was carried out from 1 July to 21 September 2017, period that corresponds to the end of the breeding period and the post-nuptial migration of the bee-eater in the region (Bota et al. 2020). Average annual rainfall ranges between 300-450 mm and average annual temperature is 14.5 °C, being this region classified as semi-arid (Calvet et al. 2004). Bee-eater abundance in the study area during the breeding period reaches the highest value for the species in Catalonia (Estrada et al 2004).

Passive acoustic monitoring

We selected nine out of the 22 apiaries to be used as acoustic monitoring stations. In each acoustic monitoring station, we placed one ARU at around 4 m of distance from the central beehive on each apiary. Each ARU (model RECoti registered trademark) consisted of an USB Voice Recorder SK-001 with processor AC1517D72772-C and one integrated and single-channel microphone. This model performed as well as commercial costliest device (see more details of the ARU model used in Pérez-Granados et al. 2019b). ARUs were ground-located with microphones in an upward position with no sound blocking barriers by at least 50 cm (Rempel et al. 2013). We used a sample rate of 44.1 kHz and 16 bits in stereo mode for all recordings. The hunting activity of bee-eaters is diurnal (Fry 2001) and varies greatly between daily time and months, with lower presence around beehives during midday hours and higher during the post-breeding migration period (Moreno-Opo et al. 2018, Bota et al. 2020). ARUs were programmed to record for 30 minutes starting at the following local (GMT+2) times: 9:15, 11:15, 13:15, 15:15, 17:15 and 19:15. ARUs were fortnightly moved between the nine selected apiaries along the entire study period. Recordings obtained during moving day were excluded from the analysis since there were no data for the whole day. Each day we simultaneously acoustically monitored 3.2 ± 1.1 stations (Mean \pm SD, range 2-5) during the whole study period.

Recordings were analysed using Song Scope 4.1.5 (Wildlife Acoustics 2011), an efficient automated sound recognition software (Knight et al. 2017). Song Scope is able to create a target signal from the characteristics of the example signals used for training, which is used as a recognizer file to compare when a sound within a recording matches its characteristics (Waddle et al. 2009). We used the same recognizer developed in Pérez-Granados et al. (2019a), which was created using part of the dataset of the current study. The recall rate of the recognizer (estimated as the proportion of bee-eater calls detected by the recognizer divided by the total number of bee-eater calls on sound recordings) was of 46.4% and the precision (events detected by recognizer that were correctly identified as bee-eater calls) was between 97.3-99.2% (mean value of 98.8%, Pérez-Granados et al. 2019a, Bota et al. 2020). Because of the high precision of the recognizer, we therefore decided to consider all events identified by Song Scope as bee-eater calls (see similar approximation in Bota et al. 2020).

Bird data

Number of bee-eater hunting attempts at beehives has been previously used as an indicator of bee-eater predation pressure in apiaries (Moreno-Opo et al. 2018). To validate that acoustic data were reliable and thus a useful indicator of the predation pressure exerted by bee-eaters, we conducted paired sampling (or “double sampling”, see Bart and Earnst 2002, Van Wilgenburg et al. 2017). We performed 35 sampling stations where we simultaneously used an ARU to record the VAR of the bee-eater and performed visual censuses, where a human observer counted the number of bee-eater hunting attempts (successful or not) of bees through pursuit in flight or stalking and capture from perches around beehives. The 35 sampling stations were evenly distributed along the study period. We only counted those bee-eater hunting attempts that occurred within a radius of 20 m around the recorder. Hunting attempts were counted continuously for 30 minutes, simultaneously while the ARU was recording. The average simultaneous bird census with ARU recording per apiary was 3.5 ± 1.1 (Mean \pm SD, range 2-5).

Bird censuses were also carried out using the same methodology in 12 non-acoustically monitored apiaries to assess the relationship between the weekly estimated VAR in the limited number of acoustic monitoring stations ($n=9$) and weekly bee-eater hunting attempts counted in non-acoustically monitored apiaries, located on a larger territorial scale. A total of 134 30-min censuses (11.4 ± 2.6 censuses per week, Mean \pm SD) were performed on selected apiaries along the study period (around one census per apiary and week) (see Supplemental Fig. S1 for location of this apiaries). These censuses included apiaries no used as acoustic monitoring station, and thus data obtained by weekly human censuses were independent of acoustic data.

Honeybees flying activity

To monitor honeybee flying activity, we installed a Melixa system (Melixa S.R.L, Italy, www.melixa.eu/en) at one beehive per apiary (Supplemental Fig. S2). The Melixa system is a commercially available hive remote monitoring system, which allows beekeepers to perform remote and real-time monitoring of the beekeeping activity and researchers to obtain detailed data about hives' dynamics (e.g., Gil-Lebrero et al. 2017, Flores et al. 2019). The vigour, health status and quantity of bees in the beehives monitored using the Melixa system were checked and verified by an expert apiarist at the beginning of the study, to exclude beehives with poor vitality condition.

The foraging activity of honeybees is influenced by several environmental factors such as wind, temperature, humidity, cloud cover, rain or sunlight intensity (Szabo 1980, Burrill & Dietz 1981, Kumar & Singh 2005, Ramírez & Davenport 2013, Moreno-Opo et al. 2018). The Melixa includes a bee counter which counts the number of entries and exits of honeybees per hour (Bee-flow). The system also records hourly data (as an average of measurements every 15 minutes) of environmental temperature (Texternal), temperature inside the hive using a probe (Tinternal),

and the presence of rain events thanks to a specific sensor (Rain) (see Table 1 for variables description). The Melixa system does not disturb the normal flying of access/exit of honeybees from the hive (Supplemental Fig. S2). The entrance is composed by 14 holes of 8 mm of diameter, which corresponds to the bee space and allows movement with no impediment. All data recorded by each Melixa System (bee flow, temperatures, rain, etc.) were automatically transferred hourly via GSM transmission to an online webserver. The same hive was monitored every fortnight in each apiary and Melixa systems were moved between the ten selected apiaries along the entire study period together with the movement of ARUs between apiaries. A total of 256 days of monitoring beehives were obtained (25.6 ± 5.7 monitored days per beehive, Mean \pm SD).

Table 1. Variables considered in the study, including variable name, type of variable (continuous, integer, categorical, count), and description.

Variable name	Type of variable	Description
<i>VAR</i>	Continuous	Total number of bee-eaters calls automatically detected per recognizer by recording (number of calls in 30 min)
<i>BE-attempt</i>	Count	Total number of bee-eaters hunting attempts (successful or not) around beehives (<20 m) in 30 min censuses
<i>Apiary</i>	Categorical	ID of the different apiaries included in the study
<i>Hour</i>	Categorical	Time of starting recording (9:15, 11:15, 13:15, 15:15, 17:15 and 19:15)
<i>Fortnight</i>	Categorical	Corresponding fortnight from 1 st July to 21 st September
<i>Bee-flow</i>	Count	Hourly number of entrances and exits of honeybees in the hive (recorded by MELIXA system). Log transformed
<i>Texternal</i>	Continuous	Hourly environmental temperature (Celsius degrees) outside hive (recorded by MELIXA system)
<i>Tinternal</i>	Continuous	Internal beehive hourly temperature (Celsius degrees) (recorded by MELIXA system)
<i>Rain</i>	Categorical	Presence (1) or absence (0) of rainy events hourly (recorded by MELIXA system)

Statistical analyses

To estimate bee-eater VAR per recording, we used the total number of bee-eater calls automatically detected by the recognizer by recording length (30 min) (Oppel et al. 2014, Pérez-Granados et al. 2019a). To determine the relationship between VAR and the number of bee-eaters' hunting attempts visually counted by human surveyors (BE-attempt), we fitted a linear

regression. We also assessed whether the use of ARUs in a relatively small number of beehives might be useful to infer bee-eater predation pressure at a larger spatial scale. For this purpose, we estimated the Pearson rank correlation between mean VAR detected per week in apiaries monitored by ARUs ($n=9$) and mean number of hunting attempts detected by human surveyors in the rest of non-acoustically monitored apiaries ($n=12$).

To determine if bee-eater predation pressure (using the VAR as a surrogate of predation pressure) significantly varied between hours and fortnights, we fitted a Generalized Linear Mixed Model (GLMM). Preliminary analysis showed overdispersion of the data, so we used a negative binomial distribution error and log-link function to account for overdispersion (Ver Hoef & Boveng, 2007). The GLMM was fitted using VAR index as response variable and Hour (six categorical levels) and Fortnight (six categorical levels) as fixed effects. Acoustic monitoring station (Apiary) was included as random effect (Table 1). When a fixed effect was found to be significant, a Tukey's post hoc test was performed to assess whether there were differences among levels.

We also fitted a GLMM to assess the relationship between flying activity of honeybees (Bee-flow) and bee-eaters' predation pressure (using the VAR as a surrogate). The GLMM (Gaussian distribution error and an identity-link function) was fitted using log transformed hourly Bee-flow as response variable and log transformed VAR as fixed effect. We also included other environmental and temporal factors that may also affect the flying activity of honeybees, and so the variables Hour, Fortnight, environmental temperature (log transformed Texternal), internal temperature of the hive (log transformed Tinternal) and presence of rain events (Rain; categorical, presence/absence) were included as fixed factors (Table 1). Since bee-eater predation pressure around beehives varies depending on daily hour and moment of the season, Hour*VAR and Fortnight*VAR interactions were also included in the model as fixed effects, while Apiary was also included as random effect (Table 1). Model performance was evaluated by plotting standardised residuals versus fixed variables, normal QQ-plots and histogram of residuals. No concrete pattern was found in any case.

All analyses were performed using the software R (v. 3.5.3) and packages "nlme" (Pinheiro et al. 2021), "lme4" (Bates et al. 2015) for GLMM models and "multcomp" (Hothorn et al. 2008) for post-hoc comparison tests.

RESULTS

We collected 1,531 30-mins recordings (765.5 hours of recording) during the study period. Recordings were scanned in about 74 hours and a total of 361,479 acoustic events were identified by Song Scope recognizer and used as bee-eater calls in posterior analyses.

We found a positive significant relationship between VAR recorded by ARUs and number of bee-eaters hunting attempts counted by human surveyors ($F_{1,34} = 72.44$, $R^2 = 0.68$, $P < 0.001$, Figure 1). We also found that mean weekly VAR detected by ARUs in nine acoustically monitored apiaries was strong and positively correlated to mean weekly number of bee-eaters hunting attempts counted by human surveyors in 12 different apiaries at a larger spatial scale (Pearson Rank correlation = 0.763, $P = 0.006$, Figure 2). Despite this overall result, a mismatch between both variables was observed during the end of August and the beginning of September (Figure 2).

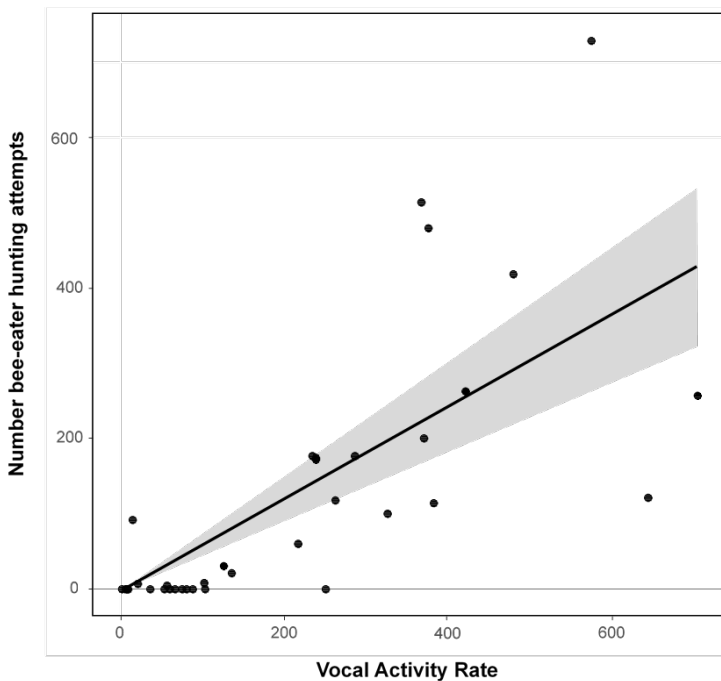


Figure 1. Linear relationship between Vocal Activity Rate (number of calls in 30 min) index and number of European bee-eaters hunting attempts counted by human surveyors ($F_{1,34} = 72.44$, $R^2 = 0.68$, $P < 0.0001$). The observed values (dots), fitted linear regression (black line) and 95% confidence interval (grey area) are depicted.

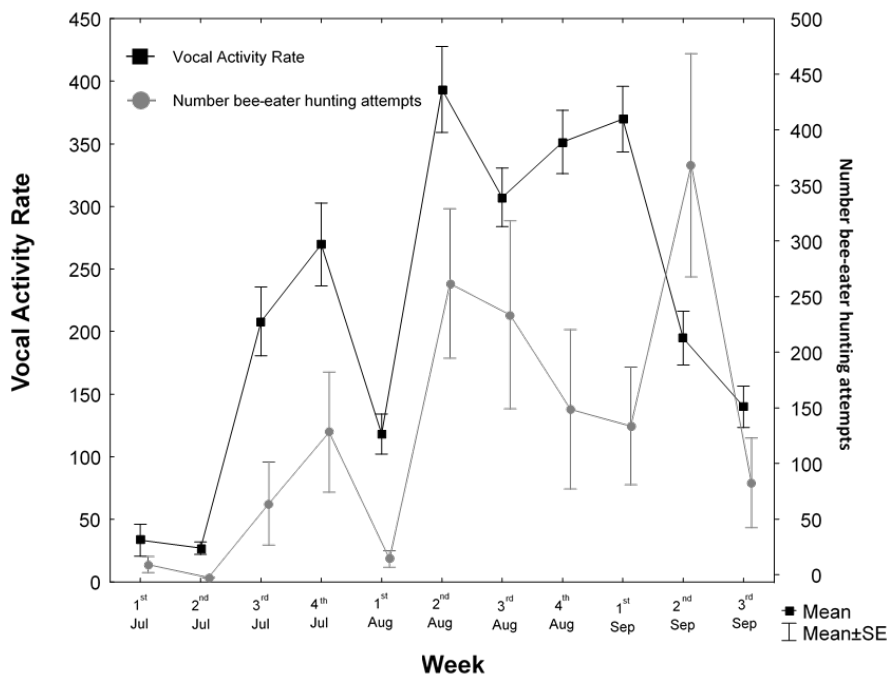


Figure 2. Weekly pattern of Vocal Activity Rate (number of calls in 30 min) and number of European bee-eater hunting attempts at beehives during the monitoring period.

Mean VAR (calls in 30 min) obtained for the entire study period and apiary was 237 ± 289 (Mean \pm SD), with a wide range of average values per apiary (maximum 393 ± 362 ; minimum 104 ± 151 , Mean \pm SD). We found a significantly variable time pattern (daily and fortnightly) in the VAR, a surrogate of bee-eater hunting attempts (Table 2). Specifically, there was a significant higher VAR during early morning (9:15) and afternoon (17:15 and 19:15) in relation to late morning (11:15), midday (13:15) and early afternoon (17:15) (Figure 3). Midday hour (13:15) presented the lowest values of all periods. Similarly, the VAR increased from early July until late August, when the highest VAR was detected, to later decrease until the end of September. Lowest VAR was detected during the 1st fortnight of July (Figure 4).

Honeybee flying activity was negatively associated with VAR, with a lower bee flow during the hours with higher number of bee-eater calls detected (Table 3). The magnitude of the effect was conditioned by the hour, being the impact significantly lower in the central hours of the day and in the afternoon at equal intensity of VAR (Table 3, Figure 5). Honeybee flying activity also decreased as the study period progressed and during rainy events, while it was positively associated with environmental and internal beehive temperature (Table 3).

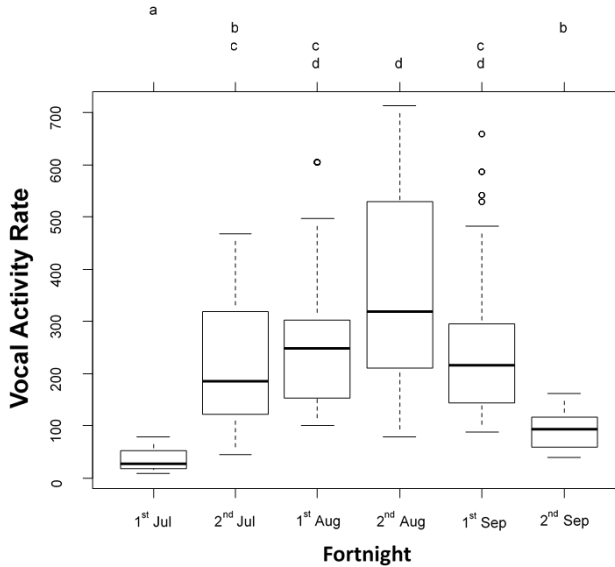


Figure 3. Daily Vocal Activity Rate (number of calls in 30 min) of the European bee-eater. Boxplots show the mean (black horizontal line), twenty-fifth and seventy-fifth percentiles of the data (boxes), and the 95% confidence interval (dashed lines). Vocal Activity Rate (VAR) is shown log transformed. Different letters on the top indicate significant differences between recording times from Tukey's post hoc test.

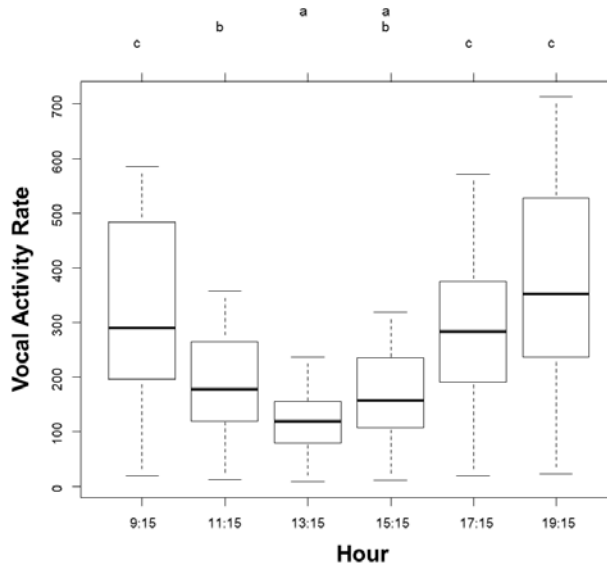


Figure 4. Fortnightly Vocal Activity Rate (number of calls in 30 min) of the European bee-eater. Boxplots show the mean (black horizontal line), twenty-fifth and seventy-fifth percentiles of the data (boxes), and the 95% confidence interval (dashed lines). Vocal Activity Rate (VAR) is shown log transformed. Different letters on the top indicate significant differences between recording times from Tukey's post hoc test.

Table 2. Summary table of the results of a Generalized Linear Mixed Model (GLMM; Negative binomial error and log-link function) testing the relationship between Vocal Activity Rate per recording (VAR; number of bee-eater calls of each 30 min recording) and Hour (six categories) and Fortnight (six categories). Number of European bee-eater calls were monitored through PAM. Acoustic monitoring station (Apiary) was included as random effects. Estimates are expressed as the differences from the intercept, which was estimated using the recordings made at 9:15 and during the first fortnight of July as reference values

Fixed effects					
	Estimate	Std. Error	z-value	P	
(intercept)	3.581	0.219	16.333	<0.0001	**
Hour (11:15)	-0.493	0.128	-3.849	<0.0001	**
Hour (13:15)	-0.906	0.129	-7.021	<0.0001	**
Hour (15:15)	-0.610	0.128	-4.747	<0.0001	**
Hour (17:15)	-0.024	0.127	-0.188	0.8506	
Hour (19:15)	0.197	0.127	1.547	0.1211	
Fortnight (2nd Jul)	1.790	0.200	8.947	<0.0001	**
Fortnight (1st Aug)	2.261	0.152	14.873	<0.0001	**
Fortnight (2nd Aug)	2.372	0.199	11.914	<0.0001	**
Fortnight (1st Sep)	2.295	0.190	12.034	<0.0001	**
Fortnight (2nd Sep)	1.479	0.234	6.312	<0.0001	**

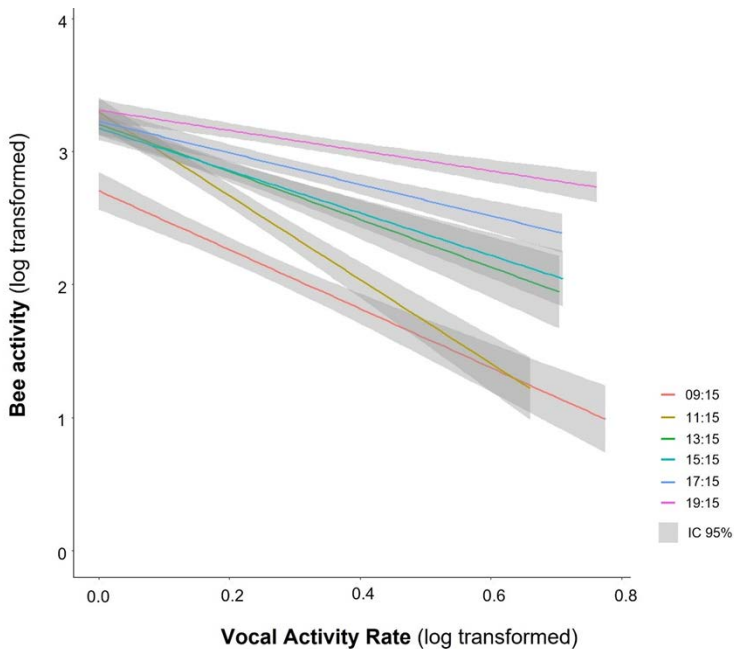


Figure 5. Linear relationship between Vocal Activity Rate (number of calls in 30 min; log transformed) index and Bee activity (hourly number of entrances and exits of honeybees in the hive; log transformed) in different hours. Fitted linear regression (one colour by hour sampled) and 95% confidence interval (grey area) are depicted.

Table 3. Summary table of the results of a Generalized Linear Mixed Model (GLMM; Gaussian distribution error and an identity-link function) testing the relationship between hourly Bee-flow (log transformed) and Vocal Activity Rate per recording (VAR log transformed), Recording time (Hour, 6 categories), External temperature (Texternal log transformed), Internal temperature (Tinternal log transformed), Rain (2 categories), Fortnight (6 categories) and the interactions Hour*VAR and Fortnight*VAR interactions as fixed effects. Apiary was included as random effect. Total number of hourly Bee-flow counts: 1,526. Estimates are expressed as the differences from the intercept, which was estimated using the recordings made at 9:15 and during the first fortnight of July as reference values.

Fixed effects						
	Estimate	Std. Error	Df	t-value	P	
(intercept)	-10.025	1.920	1492	-5.220	<0.0001	**
VAR	-1.488	0.403	1492	-3.685	0.0002	**
Hour (11:15)	0.363	0.058	1492	6.248	<0.0001	**
Hour (13:15)	0.171	0.058	1492	2.919	0.0036	*
Hour (15:15)	0.015	0.063	1492	0.244	0.8072	
Hour (17:15)	-0.008	0.068	1492	-0.129	0.8969	
Hour (19:15)	0.174	0.065	1492	2.684	0.0073	*
Texternal	2.620	0.215	1492	12.142	<0.0001	**
Tinternal	6.186	1.312	1492	4.712	<0.0001	*
Rain	-0.243	0.064	1492	-3.792	0.0002	**
Fortnight (2 nd Jul)	-0.261	0.070	1492	-3.721	0.0002	**
Fortnight (1 st Aug)	-0.303	0.055	1492	-5.426	<0.0001	**
Fortnight (2 nd Aug)	-0.187	0.072	1492	-2.572	0.0102	*
Fortnight (1 st Sep)	-0.519	0.071	1492	-7.283	<0.0001	**
Fortnight (2 nd Sep)	0.056	0.086	1492	0.686	0.5117	
VAR: Hour (11:15)	-0.706	0.202	1492	-3.487	0.0005	**
VAR: Hour (13:15)	0.154	0.225	1492	0.686	0.4926	
VAR: Hour (15:15)	0.451	0.214	1492	2.105	0.0354	*
VAR: Hour (17:15)	1.037	0.191	1492	5.415	<0.0001	**
VAR: Hour (19:15)	1.343	0.177	1492	7.560	<0.0001	**
VAR: Fortnight (2 nd Jul)	0.041	0.405	1492	0.101	0.9194	
VAR: Fortnight (1 st Aug)	-0.707	0.402	1492	-1.758	0.0788	
VAR: Fortnight (2 nd Aug)	-0.431	0.405	1492	-1.062	0.2880	
VAR: Fortnight (1 st Sep)	-0.813	0.417	1492	-1.947	0.0517	
VAR: Fortnight (2 nd Sep)	-0.652	0.517	1492	-1.259	0.2081	

DISCUSSION

Our results have proven that PAM, coupled with automated signal recognition, is an effective and useful tool to provide estimates of bee-eater predation pressure at beehives, and in consequence to assess their impact on bees' foraging activity. The positive significant relationship between VAR and number of bee-eaters hunting attempts counted by human surveyors suggests that VAR could be used as a reliable estimator of bee-eaters abundance around beehives, in agreement with previous studies with that and other bird species (Pérez-Granados et al. 2019a; see review in Pérez-Granados & Traba (2021)). Furthermore, the strong temporal positive relationship between VAR and bee-eaters hunting attempts counted at independent and non-acoustically monitored apiaries suggest that VAR estimated at a local scale may perform as a good estimator of bee-eater predation pressure at a regional scale. Nonetheless, we detected a mismatch between VAR and bee-eaters hunting attempts during late August and early September (Figure 2). This variation could be related to the migratory peak of the species. The defined period coincides with the maximum migratory bee-eater passage in the study area (Bota et al. 2020), so it is likely that the ARUs may have recorded bee-eaters calling in active migration flying above apiaries but non actively hunting.

We found that VAR (bee-eater predation pressure) varied over time. Predation pressure was lower during midday hours and higher during post-breeding migratory period. These results agree with previous studies that showed that the predation pressures of bee-eaters in the hives is not constant over time, but rather is highly concentrated at post-breeding period (especially during August) and in certain hours of the day within this period (Farinós-Celdrán et al. 2016, Moreno-Opo et al. 2018). Despite this general pattern, we detected a high variability among apiaries, thus suggesting variability of predation pressure on beehives at a local scale. PAM allowed us to quantify these variations efficiently and continuously between nearby localities, which would be useful to fine-tune and adjust possible compensations to beekeepers in a more accurate and fairer way.

Honeybees flying activity was negatively associated with VAR (bee-eaters predation pressure). Our results are in agreement with a previous study that also found a significant negative relationship between bee-eaters predation pressure and the number of bees going in and out of the hives (Moreno-Opo et al. 2018), though this study was based on a very limited sample size (around one field census counting bee-eaters per month and apiary). In our case, and thanks to the use of ARUs together with a remote monitoring system located in a hive, we were able to cost-efficiently monitor both the bee-eater predation pressure and the bees flying activity in a continuous way over long periods of time. Our results are similar to those described by Monceau et al. (2018), who found that the Asian hornet (*Vespa velutina*), an invasive honeybee predator, inhibited or reduced the foraging and flying activity of European honeybees when predation

pressure was exerted. Similarly, previous studies stated that honeybees were able to modulate their flying activity at moments when, with the same abundance of Asian hornet, the predator was less effective hunting bees (Monceau et al. 2013). Indeed, honeybees can assess predation risk and communicate this risk to the colony (Goodale & Nieh 2012). In our case, the negative relationship between VAR and bees flying activity was conditioned by the hour. At equal intensity of bee-eaters pressure (VAR), the magnitude of the effect was significantly lower during midday and during the afternoon. This could be related to the natural foraging behaviour of the European honeybee workers since their flying activity peaks during afternoon in summer (Reyes et al. 2019). Indeed, using continuous temporal data from our hive remote monitoring system, we found maximum bees' flight activity between 8 and 9 p.m. Therefore, it is likely that honeybees may be more prone to forage during midday and afternoon hours regardless of bee-eater predation pressure, which may partly explain the lower relationship between VAR and bee flow at these hours. We cannot discard the existence of a certain adaptative behaviour of honeybees to the hunting pressure of bee-eaters, since these two species have been living together for centuries. For example, populations of honeybees, where the Asian hornet is indigenous, have developed defence behaviours, while these behaviours are absent in the areas of recent colonization of this invasive wasp (Requier et al. 2019).

Further research is needed to understand the real impact and ecological mechanisms behind the relation between bee-eater predation pressure, bees flying activity inhibition and final beehive production and vigour parameters. Moreno-Opo et al. (2018) found that higher flying activity of bees did not influence the amount of honey, pollen, and brood produced despite that honey production and vigour hive parameters were negatively related to bee-eater predation pressure. These findings suggest that other factors beyond the presence of bee-eaters may be interacting in the total beehives production results. Protected wildlife usually takes more than its share of the blame (Nyhus et al. 2005) since their damages customarily are more obvious than those produced by more diffuse and complex environmental factors. This is the case of complex interactions between bee predation and environmental factors that affects apiculture production (Potts et al. 2010). If compensation programs for these damages need to be implemented, PAM would allow accurate, fully automated, comparable, and cost-efficient estimations of predation pressure exerted by bee-eaters across different apiaries, and thus, to adjust possible compensations.

The study of the human-wildlife conflicts requires fast and accurate protocols for the reliable estimation of potential competition between humans and protected species, aiming to provide realistic compensation through accuracy assessments of damage verification protocols (López-Bao et al. 2017). PAM based on the use of ARUs have proven to be a suitable and increasing used alternative to traditional field surveys for monitoring wildlife across many research areas (Sugai et al. 2019). Our study offers a new application of ARUs and new tools for the evaluation and

quantification of damages caused by wildlife, a key element for the resolution or mitigation of human-wildlife conflicts. Here, we focused on the interaction between honeybees and the protected European bee-eater, but we expect that this technique might be useful to remotely monitor the predation or damage pressure caused by other vocally active species, such as birds or invasive species damaging agricultural crops (Hu et al. 2009, Gebhardt et al. 2011, Campbell et al. 2017). Similarly, this technique might be also useful for monitoring the predation pressure caused by other conflict species with the beekeeping sector, such as the invasive Asian Hornet (see review about its impact on honeybees' colonies in Laurino et al. 2020), which might be feasible based on their flight buzzing sounds (Gradišek et al. 2017).

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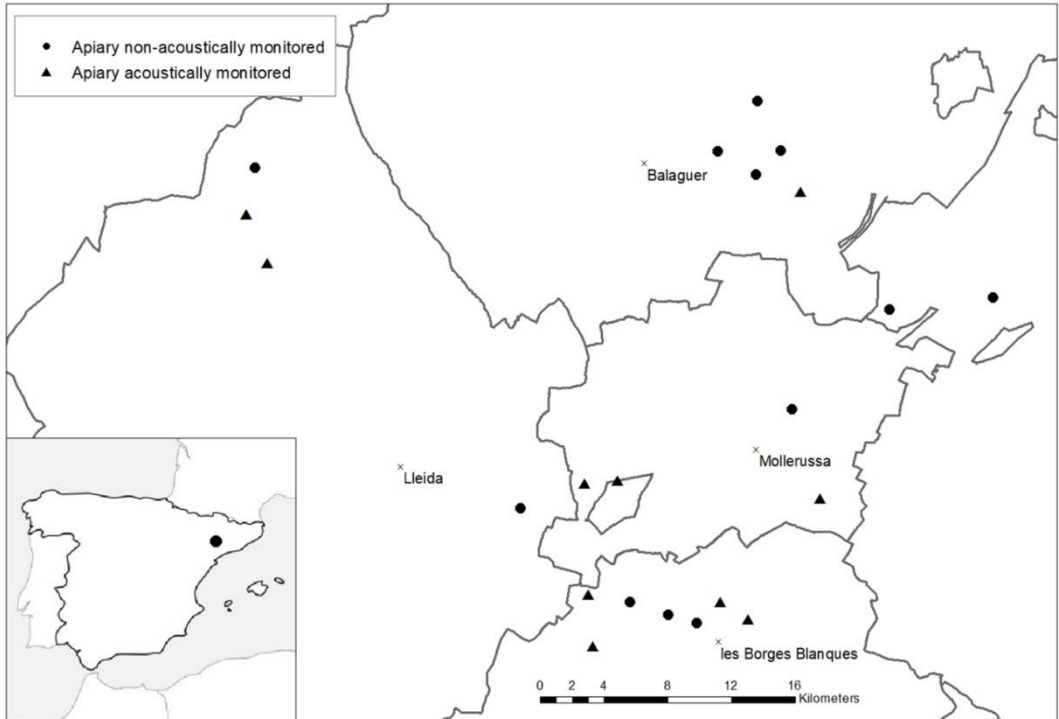
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APPENDIX I

Supplementary material S1. Study area map with the location of the apiaries included in the study, indicating which ones were acoustically monitored (triangles) and which ones non-acoustically monitored (circles).



Supplementary material S2. *Beehive monitored with Melixa system. The bee counting system can be seen at the entrance of the hive.*



GENERAL DISCUSSION

GENERAL DISCUSSION

This thesis has contributed with improvements and novelties to the application of passive acoustic monitoring in the study of birds by, (i) providing technical and methodological basis for the study of an endangered species such as the Dupont's lark, (ii) validating the suitability of using the VAR index as a useful acoustic indicator for estimating bird abundance, and (iii) successfully exploring the potential of PAM in new research fields, such as the study of diurnal bird migration or new tools for the evaluation and quantification of potential damages caused by wildlife. Although our study models have been two bird species, the methodological advancements and conclusions of the present thesis could be useful for improving passive acoustic monitoring for other acoustically active taxa.

PAM relies on technological equipment like ARUs for obtaining sound raw data. Differences between equipment, either due to the use of different recorders with different technical characteristics or technical improvements linked to technological developments, may have different consequences in the precision and quality of the obtained data and it can be a critical point for data interpretation and comparison among studies (Rempel et al. 2013, Darras et al. 2020).

Chapter I revealed large differences between sound recorders in relation to maximum and effective continuum distance for detecting bird song calls, using Dupont's lark as model species. In accordance with the initial hypothesis, detection probability decreased as the distance from the emission source increased, and higher number of vocalisations were detected at the same distance with favourable singing direction (songs uttered towards the recorder). This result highlights the need for assessing the effectiveness of potential selected recorders for acoustic monitoring programmes since its performance greatly influences the area sampled (Llusia et al. 2011, Rempel et al. 2013, Turgeon et al. 2017, Darras et al. 2020). Whenever possible, it is necessary to undertake pilot tests using focal signals played back at varying distances and directions from the recorder (Llusia et al. 2011, Darras et al. 2018, Hagens et al. 2018), to estimate the detection space (or distance) of sensors over the range of monitoring habitats. This information, depending on the monitoring objectives, should be considered to establish the appropriate distance between the sampling sites to avoid pseudoreplication or about the number of ARUs to optimize monitoring efforts (Chapter 2, Sugai et al. 2020). Furthermore, estimation and calibration of detection distance and description of methodological procedures would allow not only a more accurate quantification of the results, such population density or species detection probability, but also will facilitate the comparison among studies, once the different EDR of the ARU employed is considered in the analyses (Llusia et al. 2011, Browning et al. 2017). We are aware, however, that our results are based on one replicate per song type at each distance, and

in a single-species study, and that detection distances would differ between species and habitat type according to their signal characteristics and sound propagation (Llusia et al. 2011, Yip et al. 2017). Therefore, relative differences found between recorders could also change according to the different habitats and species tested.

Long-term monitoring objectives require comparable data sets for estimating changes of measured ecological parameters over time. Given that ARU equipment is in continuous technological evolution and, therefore it is improving its effectiveness and quality, important challenges arise to be able to guarantee the standardization and comparability of the data from PAM in the long term. Microphone degradation with field use can produce decreases in sensitivity and can be a source of variation in bird detectability that will require regular measurement of microphone sensitivity and criteria for microphone replacement to ensure scientifically reproducible results (Turgeon et al. 2017). In this context, having standardized comparison protocols between equipment and record sensitivity as a potential covariate in statistical analyses of acoustic data can be key aspects. We analysed and compared the quality of a small number of sound recorders, many of which have probably already been technologically surpassed since the study was carried out. In this sense, rigorous subsequent comparison between updated and at present widely used ARUs like Audiomoth (Hill et al. 2019), Song Meter Mini and other devices, is needed (but see Toenies & Rich (2021)). In addition, possible technological changes can also occur between versions of the same ARU model by improving and updating components, such as microphones or firmware, that can also affect the equipment performance (Darras et al. 2020, Manzano 2021).

A collateral result of this thesis was the development and registered trademark (RECoti) of a fully functional low-cost ARU based on the excellent results for the Low-Cost Recorder (LCR) obtained in Chapter I (see all technical characteristics and configuration options in Annex I). This ARU model has been successfully used in the following chapters of this thesis (Chapter 2, 3, 4 and 5) as well as in other Dupont's lark published studies (Pérez-Granados & Traba 2019, Pérez-Granados et al. 2018b, 2021), demonstrating the effectiveness and robustness of the equipment. Despite this success and considering that it was created at a time when no low-cost equipment was available in the market, its future use both for the public and for our working group will be relatively reduced. In the very last years, new smaller, cheaper, publicly available and with more functions and capabilities ARUs than self-made ones have appeared (Hill et al. 2018, Beason et al. 2019).

Low-cost ARUs have reduced expenditure of monitoring equipment, which was one of main handicaps for performing PAM during the first years of the development of the technique (Hill et al. 2019). But there are still barriers for the expansion of terrestrial PAM like the need for establishing baselines for standardizing acoustic sampling (Browning et al. 2017, Sugai et al. 2019). Establishing standards for PAM data collection improve the quality of inferences over the broad

scope of PAM research and promote essential standardization for cross-scale research to understand long-term biodiversity trends (Sugai et al. 2020). Based on the need for standardizing acoustic sampling and maximize and optimize acoustic surveys we built the ideas for chapters II and III of the thesis.

Dupont's lark population monitoring requires a lot of human and time resources given its patchy distribution (Suárez 2010), its difficulty to be visually detected, and its nocturnal singing habits with a narrow time-window peak activity (Pérez-Granados et al. 2018b). Recent results highlighted the concerning conservation status of the European Dupont's lark population (restricted to Spain), which is undergoing a 3.9% annual decline rate (Gómez-Catasús et al. 2018), with even worst trends during the last two-three years (Gómez-Catasús 2021). The last national census of the species in Spain, which covered most of the known populations, was carried out during the years 2004-2006 (Suárez 2010) and recent estimations lack of a fully national coverage (Traba et al. 2019). All these considerations suggest the need to update population estimates of the species and to establish common monitoring protocols along the different populations (Pérez-Granados & López-Iborra 2017, Traba et al. 2019).

In **Chapter II**, we developed a species-specific PAM protocol for the detection of Dupont's lark considering relevant ecological aspects, such as population density and vocal behaviour of the species. Besides, we included cost-effective considerations for the sampling design like number of ARUs needed for detecting the species presence under different simulated population scenarios, or optimal recording time schedule. As a relevant result, we defined five logical steps to develop effective other wildlife monitoring protocols using ARUs for detecting species presence, which largely coincide with others proposed subsequently by other authors subsequently (see for instance Sugai et al. 2020).

In relation to Dupont's lark monitoring, Pérez-Granados et al. (2018a), using the same ARU developed in Chapter I and the same acoustic protocol defined in Chapter II, showed the cost-effectiveness of PAM for monitoring unknown Dupont's lark populations in potential habitat patches. These authors were able to detect the species' presence in 100% of the populations where the species was present, according to field surveys, but with a much lower field effort than using traditional censuses methods. Prior research with the species showed that larger populations were more vocally active than smaller ones (Laiolo & Tella 2008). It suggests that a single human observer censusing a small population may provide false negative results due to a low bird vocal activity, while the deployment of ARUs programmed to record during long time periods allows the detection of the species at low bird density. That assumption agrees with the proposed acoustic protocol described in Chapter II, where we proposed the need to leave the recorders for a minimum of two days for detecting the species' presence with 100% certainty, regardless of population density. These results suggest that a combined methodology using

standardised species-specific field censuses for estimating population sizes in already known populations and deploying ARUs following PAM protocols (like the one defined in Chapter 2) together with the use of low-cost ARUs (for monitoring potential or very small populations) may be a cost-efficient method for monitoring large numbers of Dupont's lark sites as it is required in national censuses. Indeed, the ARU developed in Chapter I and the protocol described in Chapter II have already been successfully employed for monitoring hundreds of potential Dupont's lark populations in Castilla-La Mancha and Castilla-León during the last years (Traba et al. 2017, Traba & Garza 2020). In this context, citizen science volunteers may also be motivated to participate in extensive surveys with the opportunity to use state-of-the-art technology, such as ARUs. PAM do not need particular skills in the field so volunteers can obtain data generating positive synergies between citizen science and PAM (Penone et al. 2013, Newson et al. 2015). Beyond the detection of Dupont's lark presence, different acoustic indices based on PAM have also proven to be a good tool for inferring population estimates of the species (Chapter III, but see also Pérez-Granados et al. 2019c & 2021). It opens up new opportunities to have temporal population trends based on standardized acoustic monitoring in certain scarce and/or threatened species (Buxton et al. 2013), like the Dupont's lark. Therefore, the use of common PAM protocols can be a good and complementary step forward to improve the population monitoring at different spatial and temporal scales and even to face new objectives (e.g., population viability prediction using bioacoustics given that populations with lower song repertoires are more prone to extinction (Laiolo et al. 2008)).

Acoustic indices usefulness in monitoring requires rigorous understanding of relationships between indices and ground-truth measures of biodiversity (Browning et al. 2017). VAR index is one of these indices that may answer a key question in any species monitoring program: how many individuals of a certain species are in a monitored site and at a specific moment.

In **Chapter III**, using Dupont's lark and European bee-eater as model species, we found a positive and significant relationship between VAR index and estimated abundance for these two bird species. The strong relationship found suggests that VAR index could be a cheap and rapid method to infer bird abundance around recorders and to evaluate changes over time from recordings obtained with ARUs using omnidirectional microphones. At the time of publication of Chapter 3 (early 2019), this work was among the few to explore and to find a positive and significant relationship between VAR and estimated abundance for a terrestrial bird species (but see Borker et al. 2014, Oppel et al. 2014 for marine species). Despite the promising results obtained in Chapter III, there was still room for improvement including some aspects not considered in this work. For example, we did not include considerations that have been shown to be relevant for using VAR index, such as that the probability of detecting a bird vocalization decreases as the bird is located further to the recorder or that individual (cue rate) and population-level vocal activity may vary according to endogenous and exogenous factors, so direct comparisons in time or space

may be biased if these factors are not considered (Pérez-Granados et al. 2021). Later studies have allowed fine-tuning estimation (considering the coefficient of variation of VAR index as a function of the number of monitoring days) of Dupont's lark abundance using this index and confirmed its usefulness (Pérez-Granados et al. 2019c).

Bird density estimation using VAR index is the only acoustic approach whose effectiveness has been evaluated in a relatively large number of species and under different recording conditions (see review in Pérez-Granados & Traba 2021). It makes of the VAR index as one of the most recommended indices for estimating bird density using PAM. In addition, and for uninitiated people in bioacoustics, the VAR index is one of the indices requiring less effort and expertise for recording interpretation, since it just require counting the number of vocalizations in a sound recording. This process could be even done manually. Moreover, the VAR index also has a very intuitive interpretation of what it means and its relationship with the abundance of species, since a larger number of individuals is related to a larger number of vocalizations. Both facts can facilitate its use in conservation management such as monitoring endangered species by competent administrations. Despite this, it is necessary to continue analysing species-specific sources variation to improve index reliability in abundance estimation.

One of the strengths of the PAM is that it allows, with a relatively low field effort, continuous data monitoring thanks to scheduling options and long autonomy of ARUs (Sugai et al. 2019). The development of acoustic indices, such as VAR tested in Chapter III, opens new opportunities and applications for wildlife population monitoring that go beyond the abundance estimate itself, expanding from ecological and behavioural topics to more applied research into the field of human-wildlife conflicts, which are explored in chapters IV and V. Daily and seasonal post-breeding migration pattern of the European bee-eater described in **Chapter IV** using PAM was corroborated with independent and simultaneous observational data over time, which was essential to validate the PAM methodology for subsequent applications, given the large number of factors, both intrinsic or extrinsic, that may alter the migration pattern of the species (Farnsworth 2005, Salamon et al. 2016). As far as we know, this is the first time to apply PAM for studying a diurnal migratory species. Previous studies have focused on the study of the nocturnal migration of birds exclusively (see for example Farnsworth & Russell 2007, Sanders & Mennill 2014, Salamon et al. 2016), probably due to the difficulties to perform visual counts with traditional methods at night. Although it is possible to carry out visual counts of migrating birds during the day, the PAM continued to offer complementary advantages. On the one hand, this methodology may be an effective, alternative or complementary technique for monitoring migration of vocally-active species, such as different species of cranes, finches, swallows, swifts or wagtails, among other bird groups. Secondly, we found that a limited number of ARUs provided migratory temporal patterns compatible with those observed on a larger scale. Methods to capture multiple taxa information over broad spatial and temporal scales have been a central issue for improving global biodiversity

monitoring in the face of human-driven changes (Schmeller et al. 2017). PAM stations can easily be located close to existing locations where bird migration is usually concentrated and monitored (e.g., migration bottlenecks like sea straits or mountain passes) to provide data from vocal animals (Sugai et al. 2019). This opens up the possibility of creating a network of acoustic PAM stations for bird migration at strategic sites for measuring timing and relative bird migration intensity of bird species over time in wide scales. At these sites, both diurnal and nocturnal migration could be monitored, allowing to study the phenomenon of bird migration using the same methodology in a standardized way (for example with hourly recordings of the same duration) and continuously both at night and day.

Another new potential application of the PAM linked to the use of the VAR index is the possibility to detect the presence and measure the predation pressure of potentially conflictive species with human activities. In a context of increasing human–wildlife conflict and in order to create public trust in the legitimacy of potential compensation schemes, it is crucial to develop accurate protocols for the reliable verification of the causative species and its relationship with potential damage claims (López-Bao et al. 2017, Ravenelle & Nyhus 2017). These protocols require quick verification methods and adequate methodological tools to properly identify and quantify responsible and losses (Nyhus et al. 2003). In the case of bee-eaters and beekeeping conflict, the interactions of the bee-eater with honeybees can be relatively continuous in time (Moreno-Opo et al. 2018) and no direct identifiable trace of potential harm remains, making evaluation of predation pressure by the species difficult by direct observation or expert verifications.

In **Chapter V**, we demonstrated the usefulness of the PAM to evaluate the predation pressure exerted by the European bee-eater to honeybees in different apiaries. First, a significant relationship was established between the VAR index and the number of bee-eaters hunting attempts observed. Once assessed the relationship between VAR and predation pressure of the species, we were able to describe the daily and seasonal pattern of the predation pressure of bee-eaters at beehives, in a continuous way thanks to acoustic monitoring. We also found that honeybee flying activity was negatively correlated with VAR index (surrogate of the bee-eater predation pressure), but the magnitude of the effect was conditioned by the hour. The ability of honeybees to increase their flying activity at equal pressure of bee-eaters during midday and afternoon needs further research. On the one hand, it could be related to the natural foraging behaviour of the European honeybee workers since their flying activity peaks in afternoon during summer (Reyes et al. 2019). On the other hand, honeybees could have developed some adaptation behaviour to bee-eaters hunting pressure since interactions between both species have been taking place in evolutionary time. In the case of the Asian hornet (*Vespa velutina*), a species that operates in a similar way to the bee-eater, the populations of honeybees where this wasp is indigenous have developed defence behaviours, while these behaviours are absent in the areas of recent colonization of this invasive wasp (Requier et al. 2019).

Moreno-Opo et al. (2018) found that the number of bee-eaters interacting and hunting around apiaries was negatively related to honeybee flying activity and to the amount of stored honey, pollen, and brood in the studied hives. Nevertheless, this significant negative relationship was not in a very pronounced way and bee-eaters did not influence beehive survival, one of the main concerns of beekeepers. Inhibition of the flight activity of honeybees due to bee-eaters attacks may be an important mechanism behind production losses, as had been pointed out for other bee-predator species, such as the Asian hornet that act in a similar way than bee-eaters do (Monceau et al. 2018). Despite this, higher flying activity of honeybees did not influence the amount of honey, pollen, and brood found (Moreno-Opo et al. 2018), so other factors beyond the presence of bee-eaters may be interacting in the total beehives production results. The next step should be establishing specific protocols to link predation pressure of bee-eaters measured using PAM with the real damages in the hive production in order to know the ecological mechanisms and other environmental influencing factors behind these potential losses, if ever produced.

Furthermore, preventive measures, like shading roofs for apiaries, were shown as effective alternatives in some contexts for reducing bee-eaters impact (Moreno-Opo et al. 2018). Compensation programs are recommended to link damage payments to conflict prevention measures since full compensation without requiring preventative measures may discourage investment in protection (Rondeau & Bulte 2007). Making compensation conditional on adoption of preventative activities may also encourage farmers to implement approaches to reduce the risk of future conflict (Boitani et al. 2010). Therefore, delving into the potential impact of the bee-eaters on hives production must go hand in hand with the development and improvement of preventive measures. In this context, and beyond the already proposed preventive measures, exploring the possibility of automatic activation of dissuasive measures based on certain thresholds of presence of bee-eaters measured on-site by PAM, may be an important field of technological development.

From our knowledge, this is the first time that the PAM has been used to measure the hunting pressure of a bird species in a context of human-wildlife conflict. In this case, PAM coupled to automated signal recognition programs can provide a reliable method for verifying and quantifying the predation pressure of bee-eaters in hives, continuous in time (necessary since the attacks are not isolated) and comparable between apiaries (if ARUs and protocols for taking common data and analysis are applied). We focused our work on the interaction between honeybees and the European bee-eater, but we expect that PAM might be useful to monitor predation pressure by other conflict species with the beekeeping sector, such as the invasive Asian hornet (Laurino et al. 2020) or in other human-wildlife conflicts caused by other vocally active species, such as birds or invasive species damaging agricultural crops (Hu et al. 2009, Gebhardt et al. 2011, Campbell et al. 2017).

To conclude, throughout this thesis different results have been presented that have brought improvements and novelties to the application of PAM in the study of birds from an applied perspective. The explored approaches and methodologies have made it possible to efficiently address complex questions using other study methods both within (e.g., estimating bee-eater hunting pressure on beehives) and outside (e.g., discovering unknown Dupont's lark populations) the scope of this thesis. In addition, part of the results can be used in other contexts and questions through small adaptations of the methodologies developed. It is evident that: i) PAM is a well-suited methodology for studying and monitoring terrestrial biodiversity, like birds, ii) PAM is here to stay and grow, and iii) this thesis is only a small example of its enormous potential when new questions arise.

CONCLUSIONS

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1. There can be important differences between Autonomous Recording Units (ARUs) models in relation to the distance at which can effectively record bird song vocalizations. Moreover, this distance is highly influenced by external factors, such as bird singing direction. These results highlight the need to evaluate in advance the performance of selected ARUs for Passive Acoustic Monitoring (PAM) in order to optimize the monitoring design to the objectives.
2. Future PAM surveys aiming to detect the presence of the Dupont's lark should leave the recorders in the field from one hour before dawn to dawn and over two consecutive days. The design of specific PAM protocols for bird monitoring, based on the ecology of the target species and the technical characteristics and performance of the ARUS to be used, offers great opportunities for maximizing species detection and minimizes survey efforts.
3. The development of acoustic indicators able to infer bird abundance around recorders is a key aspect to integrate the use of PAM into applied studies of conservation and monitoring programmes of bird species. The Vocal Activity Rate (VAR) index has proven to be a good and feasible indicator to estimate the abundance of the Dupont's lark and the European Bee-eater around recorders.
4. The establishment of new robust relationships between VAR index and species abundance presents a range of new opportunities and applications for wildlife population monitoring that go beyond the abundance estimate itself. For example, it has proven useful in establishing the migratory temporal pattern of the European bee-eater. Acoustic monitoring can provide robust and continuous information about the daily migration pattern of migrating species, allowing for longer and standardised daily time series of data, at a lower cost than human visual counts.
5. PAM, coupled to automated signal recognition programs together with VAR index estimation, can provides a reliable method for verifying and quantifying the predation pressure of European bee-eaters in beehives. This opens up new opportunities to monitor potential beekeepers damage claims, as long as the relationship between predation pressure exerted and economic losses can be established.

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APPENDIX I

Technical characteristics and configuration options of RECoti Autonomous Recording Unit

At the time of the field work (2016 and 2017) of the different chapters, there was no low-cost ARU available in the market. It is not until 2018 when Audiomoth, a low-cost and today widespread used ARU, appeared (Hill et al. 2018). In that context, due to budget limitations to acquire enough ARUs for my objectives, I proposed the design and manufacture of a low-cost ARU called RECoti (Figure 1; Table 1).

For this objective we compared five commercially available sound recorders, ranging from ARUs manufactured by Wildlife Acoustics to available devices, such as low-cost audio recorders (LCRs) and hand sound recorders (see Chapter 1). LCRs are inexpensive sampling audio recorders which have the external shape of a universal serial bus (USB) flash drive, and are composed of a microphone, an analog-to-digital converter, central processing unit with permanent internal or external non-volatile memory, rechargeable battery, and a USB connection. Farina et al. (2014) showed the potential of these devices for use in acoustic monitoring given their high portability, recording quality and low price. I chose a SK-001 LCR, equipped with an AC1517D72772-C microprocessor. The recorded files are saved in mp3 format on a removable micro-SD card to facilitate their subsequent manipulation and processing in field conditions. Despite its characteristics, the LCR tested did not meet the requirements to be an ARU since it had a limited autonomy, it did not have time programming capacity and it was not protected towards cannot be left unattended operated in external conditions.

In order to transform selected LCR to an operative ARU with full capacities, we assembled a digital time programmer, a 12 to 5V DC D-SUN regulator, a signal relay, and a simple circuit to perform the on- off operation. The entire circuit was powered by a 3-cell Lipo battery, protected against over-discharges with a BCP circuit.

As can be seen in the diagram (Figure 2), the on-off operation of the recorder was carried out with a switching circuit provided by the relay. The relay was powered by the digital time programmer, according to the cycle programmed. This programmer allowed me to schedule up to 16 daily recording periods. The power of the recorder was maintained at all times through the regulator, to allow, at the moment of disconnection, the saving of the generated files in the external memory.

The consumption of all the components in the circuit was around 2.8 mAh in standby mode and 20.8 mAh in active mode, which, together with the high capacity of the Lipo batteries, allowed to record about 300 hours continuously (with 8000 mAh batteries and reaching 80% power download). The set was mounted in a watertight box with IP65 degree protection that

allows it to operate in outdoor conditions. After including all components and working-hours for assembling, estimated cost was about 180 Euros per unit that made it an affordable low-cost ARU for our objectives.

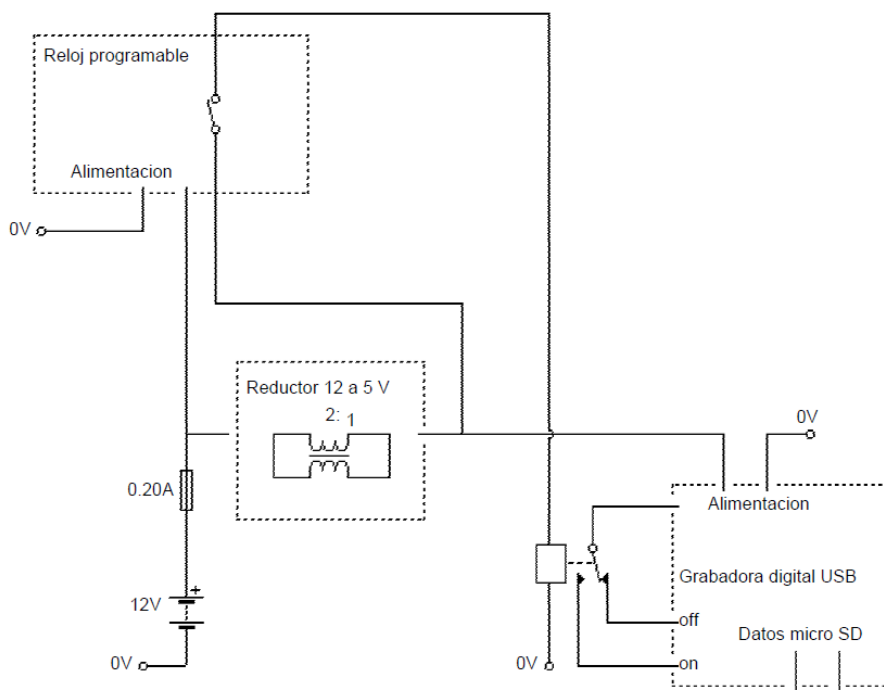
The ARU model created has been registered as a European Union trademark with the name RECoti (see logo at Figure 3) and had been recorded included in the Register of European Union trademark Register.



Figure 1. Photos of the RECoti ARU where it can be seen its interior with the Lipo battery, the motherboard, and the programmer, all protected within the waterproof case.

Table 1. Principal technical characteristics of the RECoti ARU

- SK-001 LCR with AC1517D72772-C processor
- Omnidirectional microphone
- 3-cell LiPo battery, 11.1 V (8000 mAh or 2200 mAh depending on version) including protection circuit (BCP)
- Recording of MP3 files on Micro SD card of 8-16 Gb (1 Mb per minute of recording, approx. 120 hrs autonomy)
- Programmable daily clock with 16 programs, powered by an independent lithium battery (3 years of autonomy)
- Energy consumption: in standby mode (2.8 mA), in recording mode (20.8 mA)
- Dimensions: 171 x 100 x 100mm (not including microphone)
- Weight: 385 g, without batteries (8000 mAh /650 g battery, 2200 mAh / 112g battery)



Grabadora digital temporizada

Figure 2. Electronic diagram of the RECoti ARU.



Figure 3. *RECoti* logo of the registered trademark.

APPENDIX II

COST-EFFECTIVENESS ASSESSMENT
OF FIVE AUDIO RECORDING SYSTEMS
FOR WILDLIFE MONITORING:
DIFFERENCES BETWEEN RECORDING DISTANCES
AND SINGING DIRECTION

EVALUACIÓN DEL COSTE Y EFECTIVIDAD DE CINCO SISTEMAS
DE GRABACIÓN DE AUDIO PARA EL SEGUIMIENTO
DE FAUNA SILVESTRE: DIFERENCIAS ENTRE DISTANCIAS
DE GRABACIÓN Y DIRECCIÓN DEL CANTO

Cristian PÉREZ-GRANADOS^{1, 2 *}, Gerard BOTA³, David GIRALT³,
Josep ALBARRACÍN³ and Juan TRABA^{1, 4}

SUMMARY.— Audio recording systems coupled with automated song recognition are commonly being used for monitoring wildlife. Recorders usually differ in cost and effectiveness, and their performance may vary with source distance, wind speed and acoustic source direction, among other factors. We here assess the cost-effectiveness of five audio systems considering such factors as distance and singing direction. We developed field tests using playback of Dupont's Lark *Chersophilus duponti* songs from nine fixed locations at distances of 1 to 256m, played towards or away from the recorders' position. We selected this species because its very characteristic song should be easily identified by automated signal recognition software. Field tests were carried out during March 2016 in level dwarf-shrub steppe (mean height < 40cm) in NE Spain. We found large differences in effectiveness between recorders. The number of songs detected by an automated signal recognition algorithm significantly decreased with distance and when playback was angled away from the recorder position, a factor never previously tested. Finally, we give the design of a cost-effective Autonomous Recording Unit, based upon the most effective recorder. We recommend researchers working with acoustic recorders to evaluate the performance of several devices before making a selection for long-term monitoring pro-

¹ Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain.

² National Institute for Science and Technology in Wetlands (INAU), Federal University of Mato Grosso (UFMT), Computational Bioacoustics Research Unit (CO.BRA), Cuiabá, Mato Grosso, Brazil.

³ Biodiversity and Animal Conservation Lab. Landscape Dynamics and Biodiversity programme. Forest Science and Technology Center of Catalonia (CTFC), Solsona, Catalonia, Spain.

⁴ Centro de Investigación en Biodiversidad y Cambio Global (CIBC-UAM), Universidad Autónoma de Madrid, Madrid, Spain.

* Corresponding author: cristian.perez@ua.es

grammes, and to consider such factors such as singing direction in their analyses. —Pérez-Granados, C., Bota, G., Giralt, D., Albarracín, J. & Traba, J. (2019). Cost-effectiveness assessment of five audio recording systems for wildlife monitoring: differences between recording distances and singing direction. *Ardeola*, 66: 311-325.

Key words: acoustic monitoring, automated signal recognition, Autonomous Recording Unit, *Chersophilus duponti*, classification software, playback.

RESUMEN.—Los sistemas de grabación de sonido acoplados al reconocimiento automático de cantos mediante programas informáticos se están usando cada vez más comúnmente para el seguimiento de fauna. Los grabadores habitualmente difieren en su coste y efectividad, y su desempeño puede variar con la distancia y dirección a la fuente de sonido, la velocidad del viento y otros factores. Aquí evaluamos la relación entre el coste y la eficacia de cinco sistemas de grabación de audio factores como la distancia y la dirección a la fuente de sonido. Desarrollamos pruebas de campo usando grabaciones del canto de la alondra ricotí *Chersophilus duponti* desde nueve localizaciones fijas a distancias entre 1 a 256 m, reproducidas hacia la posición del grabador o en sentido opuesto. Seleccionamos esta especie porque su canto es muy característico y puede identificarse fácilmente mediante los programas de reconocimiento de señales acústicas. Las pruebas de campo se hicieron durante marzo de 2016 en estepas de caméfitos (altura media < 40 cm) en el noreste de España. Encontramos grandes diferencias entre grabadores. El número de cantos detectados por el algoritmo de reconocimiento automático de señales acústicas decreció significativamente con la distancia y cuando la emisión del reproductor se desvió de la dirección del grabador, un factor que no se había evaluado antes. Finalmente, proporcionamos el diseño de un grabador automático rentable en términos de efectividad y coste, de acuerdo con nuestros resultados. Recomendamos a los investigadores que trabajan con grabadores de sonido que evalúen el desempeño de distintos aparatos antes de seleccionar uno para programas de seguimiento a largo plazo, y que se considere la dirección del canto entre los factores a tener en cuenta en los análisis. —Pérez-Granados, C., Bota, G., Giralt, D., Albarracín, J. y Traba, J. (2019). Evaluación del coste y efectividad de cinco sistemas de grabación de audio para el seguimiento de fauna silvestre: diferencias entre distancias de grabación y dirección del canto. *Ardeola*, 66: 311-325.

Palabras clave: *Chersophilus duponti*, grabador automático, reconocimiento automatizado de señales, reproducción, seguimiento acústico, software de clasificación.

BACKGROUND

In recent years, researchers have used Autonomous Recording Units (hereafter ARUs) as a tool to monitor a wide range of taxa based on the sounds they produce. Such taxa include mammals (Heinicke *et al.*, 2015), amphibians (Acevedo & Villanueva-Rivera, 2006; Shearin *et al.*, 2012), insects (Brandes, 2005) and birds, the last of these being the most commonly surveyed group (e.g. Digby *et al.*, 2013; Ganchev *et al.*, 2015, Alquezar & Machado, 2015). This methodology requires the placement of one or several ARUs in the field to record sounds during the time

of interest (e.g. dawn, Gil *et al.*, 2014; Zwart *et al.*, 2014) or for 24-hour periods (Jahn *et al.*, 2017) followed by interpretation of the recordings. Several studies have demonstrated that ARUs are able to offer a suitable alternative to traditional field survey methods for detecting species presence and for describing habitat occupancy and community composition of animals that produce sounds (see review for birds in Leach *et al.*, 2016).

Field surveys performed by observers are subject to intrinsic biases, notably those due to interpersonal variation in the ability to detect and identify songs, resulting from differences in observer age, experience and

hearing acuity (Cyr, 1981; Kepler & Scott, 1981). Observer presence may also influence vocal activity and natural behaviour or may provoke non-natural displacements of the studied species (Acevedo & Villanueva-Rivera, 2006). Moreover, field surveys are usually short and time-restricted and thus prone to temporal biases due to weather conditions, moon phase, daily vocal activity variation, etc. (Bibby *et al.*, 2000; Catchpole & Slater, 2008, Pérez-Granados & López-Iborra, 2017). The use of ARUs offers an efficient alternative independent of many of the biases of field surveys, since the technique is non-invasive, consistent over time and creates a permanent and archivable record of surveys (Acevedo & Villanueva-Rivera, 2006; Brandes, 2008). Recordings can be re-examined by experienced observers (Rempel *et al.*, 2005), re-analysed using song identification programs (de Oliveira *et al.*, 2015) and can provide useful information for future studies and comparisons (Alquezar & Machado, 2015).

In the last decade, the use of ARUs for monitoring biodiversity has increased in popularity. However, some shortcomings associated with their use should not be overlooked. Species with low sound output can be missed or can remain unidentified (Acevedo & Villanueva-Rivera, 2006; Alquezar & Machado, 2015). Moreover, ARUs usually have a lower sensitivity than a human listener (Hutto & Stutzman, 2009; Yip *et al.*, 2017) that decreases with distance (Jahn *et al.*, 2017). Sound recordings obtained by ARUs provide little or no ability to determine sound direction in three dimensions and to distinguish between individuals, which makes it difficult to estimate wildlife abundance (but see Drake *et al.*, 2016 and Pérez-Granados *et al.*, 2019). Furthermore ARUs require a large amount of expert time for the analysis of recordings (Hutto & Stutzman, 2009; Digby *et al.*, 2013), although automated signal recognition and classification softwares for processing large data sets promptly have

favoured their use (Heinicke *et al.*, 2015; de Oliveira *et al.*, 2015).

ARUs differ in price, size, weight, sound sensitivity, signal-to-noise ratio, quality and the directionality of microphones, among other factors (Venier *et al.*, 2012; Fristrup & Mennitt, 2012; Rempel *et al.*, 2013; Turgeon *et al.*, 2017). Differences between ARUs may cause bias in, for example, estimated population size or trend estimates (Rempel *et al.*, 2013), and so it is essential to test variation in effectiveness between different ARUs. Only Rempel *et al.* (2013) have empirically tested the differences between six different recording systems. They found differences in their sensitivity (response to different frequencies) and in the signal-to-noise ratio, as well as in the number of bird species detected by a listener after song analyses, irrespective of cost (Rempel *et al.*, 2013). Yip *et al.* (2017) also evaluated the detection distances of four different ARUs but offer no comparisons or data regarding the differences found between them. Likewise, there are no studies assessing the effectiveness of different recording systems using automated signal recognition software.

Animals use directionality of sound to avoid the risk of being detected by unintended listeners (Larsen & Dabelsteen, 1990) and also to focus the sound intensity in the direction of intended listeners. The direction of sound propagation and the head and body position are important factors determining sound transmission efficiency in birds (Titze & Palaparathi, 2018). The sensitivity of ARUs is therefore expected to vary with the direction in which sound is being propagated, a prediction so far untested.

The main goal of this study was to evaluate the performance of five different recording systems, ranging from low- to high-cost multipurpose digital recorders with realistic field tests, aiming to elucidate the effect of playback distance and singing direction their effectiveness. We used automated song de-

tection software to estimate the number of signals automatically detected at different distances both with playback broadcasted directly towards recorders (favourable singing direction, hereafter) and also in the opposite direction (unfavourable singing direction, hereafter), to determine an index of their effectiveness. We predicted that more vocalisations would be detected at closer source locations compared to more distant ones. Also there would be greater detection distances and a higher number of vocalisations detected at the same distance with favourable than with unfavourable singing directions. For each recorder, we also calculated the total amount of software processing time and we made a cost-effectiveness assessment to identify the most adequate recorder to be used for long-term monitoring studies. Playback consisted of a series of Dupont's Lark songs and were carried in dwarf-shrub steppe, a typical habitat for the species (Seoane *et al.*, 2006, Pérez-Granados & López-Iborra, 2017). We selected this species because its song consists of a discrete number of song types (Pérez-Granados *et al.*, 2016) that end with a very species specific and common sequence (see Supplementary Material, appendix 1), that should be easily identified in the spectrogram. Our ultimate goal was to build our own weatherproof and programmable ARU, equipped with long-life batteries, for use in further field studies (see Pérez-Granados *et al.*, 2018a; 2018b).

METHODS

Field tests

Field tests were carried out on the sunny and cloudless morning of 24 March 2016 in the Timoneda d'Alfés (Lleida, Catalonia, North-eastern Spain, 45.50°N, 77.76°W). Tests started at 9:28 a.m and finished at 10:10 a.m (local time, GMT +1). The Timoneda d'Alfés

is a continuous and homogeneous dwarf-shrub steppe (mean height < 40cm) of 1km² dominated by *Thymus vulgaris*, *Sideritis scordioides* and *Helianthemum* spp. We used five sets of recording equipment ranging in cost from 35 to 1,100 euros (2016 prices; Table 1). These were the Wildlife Acoustics Song Meter™ SM2 with pencil microphones (SM2 hereafter), Olympus DM650 16-Bit PCM Stereo Recording (Olympus), Sony ICD-P320 with compact 6-mm element microphones (Sony), Sytech Digital recorder SY-1707 with compact 25-mm element microphones (Sytech), and a Mini USB Voice Recorder SK-001 with AC1517D72772-C processor and integrated microphones (USB, Table 1). All recorders remained available in March 2019, unless SM2 has been updated by SM4. Recorders were attached to a 50cm-tall wooden stick and remained in the same position and above natural vegetation throughout the tests. Recorders were located with microphones in an up-position, separated by 1m in order to minimise any blockage of sound (Rempel *et al.*, 2013). All recordings were made at 44 kHz and 16 bits and using a bit rate of 1411 kbps. Recordings for SM2 and Sytech were made in wav format while recordings for the other units were made in mp3 format and needed post-transformation to wav format for data analyses in Song Scope (see below). Although compression into mp3 format affects the spectral and temporal composition of the signal (Obrist *et al.*, 2010), and its conversion to wav format does not improve recording quality, this should not influence the signal recognition results (Rempel *et al.*, 2005). We broadcast a digital and standardised recording (hereafter playback) of Dupont's Lark songs to aid in interpreting the results and facilitate automatic signal recognition by the classification software. The playback lasted for 70 seconds and comprised 13 Dupont's Lark songs recorded at different sites. Playback equipment consisted of a digital player

(CAPADI, ref. MR102) with a coupled tweeter (CAPADI, ref. AB105). Playback volume and height above the ground (1m) were constant throughout the tests. The intensity of the playback, measured as Leq (Equivalent Continuous Sound level) at 2m, was 76.3 dB similar to a normal singing volume of the study species (authors' own data). The recording equipments had omnidirectional microphones and recorded simultaneously. Likewise, device locations did not vary throughout the field tests and were under the same environmental conditions in order to avoid biases.

Distance and singing direction response

The playback was broadcasted from nine fixed locations, 32m apart and from 1m to

256m away. We carried out field tests broadcasting the playback both directly towards and away from the recorders, to estimate recorder performance under the most favourable field conditions – a bird singing towards the recorders, and under the most unfavourable conditions – a bird singing directly away from the recorders. We only broadcast the playback once at each distance and singing direction. Wind speed during field tests was very low (< 2m/s) and so was disregarded in our analyses.

Sound analyses

Field recordings were analysed by the same observer (CPG) and using the same laptop (Intel(R) Celeron(R) 2.16 GHz, 4096 MB RAM), automated signal recognition soft-

TABLE 1

Names and specifications of the five recorders used in the study. The approximate cost per unit (€) at the time of purchase is also shown. Costs and specifications for the USB recorder are shown for the customised recorder (Supplementary Material, appendix 6). *At maximum recording quality and storage capacity.

[Nombre y especificaciones de los cinco grabadores empleados en el estudio. También se muestra el precio aproximado por unidad (€) a la hora de la compra. El coste y especificaciones para el grabador USB se refieren al grabador creado (Material Suplementario, apéndice 6). *A máxima calidad de grabación y capacidad de almacenamiento.]

Abbreviation	Digital recorder	Manufacturer	Storage Capacity (GB)	Recording time* (h)	Programmable	Weatherproof	Cost (Euros)
SM2	Wildlife Acoustic Song Meter™ SM2	Wildlife Acoustics	4 × 32	185	Yes	Yes	1,100
Olympus	Olympus DM650 16-Bit PCM Stereo	Olympus	16	107	Yes	No	160
Sony	Sony ICD-P320	Sony	2	7	Partially	No	60
Sytech	Digital recorder Sytech SY-1707	Sytech	8	48	No	No	35
USB	Mini USB Voice Recorder SK-001	QFRR009	16	260	Yes	Yes	180

ware and headphones. Automated song detection was performed using Song Scope 4.1.5 (Wildlife Acoustics, 2011), with the help of the spectrogram visualisation tool of the software. Song Scope is able to create a target signal from the characteristics of the set of signals used for training, and uses it as a recogniser file to compare when a recorded sound matches the target signal (Waddle *et al.*, 2009). We built viable and customised recognisers for each recorder (Towsey *et al.*, 2012), always using the same settings after following the software recommendations (see Supplementary Material, appendix 2). We used a specific recogniser for each recorder after comparing the number of songs detected under favourable singing conditions per recording by both a recorder-specific and a neutral recogniser (the latter using those calls broadcast at playbacks, see Supplementary Material, appendix 3). In our analyses, we only considered the final sequence of the Dupont's Lark song as a target signal, since this is easily identified in the spectrogram and should be easily detected by automated classification software (see Supplementary Material, appendix 1).

For building recognisers, we aimed to adjust sample rate, frequency range and minimum frequency to help isolate the target signal, and remove all lower and higher-amplitude events, which are not likely to be part of the target signal (Waddle *et al.*, 2009). In this way, we annotated selected songs in a clean spectrogram (Digby *et al.*, 2013; de Oliveira *et al.*, 2015). We chose and annotated 12 Dupont's Lark songs as models to create one specific recogniser for each recorder (Waddle *et al.*, 2009). We selected six songs from the tests performed under favourable singing directions and another six performed under unfavourable singing directions. In both cases, three songs were selected at 1m, two at 32m and one at 64m. We selected songs from different distances and singing directions to create a more accurate recogniser,

able to detect the target signal under imperfect recording conditions. All recognisers were created using at least ten of the 12 selected songs, and in the cases in which a particular song could not be used (e.g. bad spectrogram quality), another song from the same distance and singing direction was selected to minimise biases between recognisers. The most important setting to be considered when building a recogniser is the *cross-training* value, which is a measure of how well the recogniser is expected to perform (Wildlife Acoustics, 2011). A low score (e.g. < 50%) may indicate that the generated recogniser may not accurately find the target signal within a recording (Wildlife Acoustics, 2011). Recordings were scanned with their own recognisers using algorithm 2.0 in Song Scope (Waddle *et al.*, 2009). Recogniser scanning reported a series of events identified as a target signal by the recogniser. All events were visually and/or acoustically checked, and a true positive was considered when the software correctly matched a Dupont's Lark song, while a false positive was noted when a non-Dupont's Lark song was recognised (Wolfgang & Haines, 2016).

For each distance and singing direction we estimated the total number of events detected, the number of true positives and the number of false positives. For each recorder and singing direction we calculated: (1) the maximum detection distance at which at least one Dupont's Lark song was detected; and (2) the effective continuum distance, as the distance at which songs were detected in all shorter distance intervals. We also estimated: (3) detection rate: the percentage of Dupont's Lark songs detected in relation to the total number of songs played, (4) success rate: the percentage of Dupont's Lark songs correctly classified relative to the total number of events recognised, and (5) the time needed by automated signal recognition software to complete recording analyses for each recording system.

Cost-effectiveness assessment

To objectively identify the most effective recorder to be used as an ARU, we proposed a methodology based on scoring each device according to its detection and success rate at each distance and singing direction. Three points were awarded when detection or success rate was > 50%, two points when detection or success rate varied between 50% and 25%, one point when detection or success rate was < 25% and zero points when no songs were detected or correctly classified. We also estimated the number of units of each ARU needed –according to their continuum distance under unfavourable singing directions, limiting distance– to monitor simultaneously a potential habitat patch of 1km²: this number was multiplied by their estimated price to estimate the total cost.

Statistical analyses

We used GLMM with family binomial to test the probability of a song being detected, using type of recorder (five levels), distance (nine levels) and singing direction (two levels) as fixed variables, and detection success (detected/undetected) as the dependent variable. As the playback employed 13 different Dupont's Lark songs, all of them being broadcast at all distances and directions, we included song type (13 levels) as a random factor. If a fixed effect was significant, a Tukey's *post hoc* test was performed to test for differences between levels. Model performance was evaluated by plotting standardised residuals versus fixed variables, normal QQ-plots and histogram of residuals. No concrete pattern was found in any case. We tested the difference in the number of songs detected at each specific distance between the tests performed with favourable and unfavourable singing directions using Mann-Whitney U tests. Data analyses were

conducted in R 3.4.1 (R Core Team 2016). We used packages “lme4” (Bates *et al.*, 2015) for logistic GLMM and “multcomp” (Hothorn, 2008) for post-hoc comparison tests.

RESULTS

Recording analyses

Cross-training values for recognisers created for each recorder were similar and ranged between 72.2 and 78.1 (Table 2). Software processing time differed among devices. Sytech recordings were the fastest to be analysed (226 sec.) while Sony were the most time-consuming (910 sec., Table 2).

Recorder effectiveness

According to GLMM, there were significant differences among recorders in the total number of songs automatically detected (Table 3). SM2 and USB detected a significantly higher number of songs than the rest of recorders (c.22%), while Olympus detected the lowest (c.8%) (Table 2). SM2 and USB did not differ significantly in the Tukey *post-hoc* test (Figure 1).

The maximum detection distance differed greatly among recorders and was strongly affected by singing direction (Table 2). For example, the maximum detection distance in tests performed with favourable singing direction ranged between 256m (SM2, Sony, USB) and 128m (Olympus). Under unfavourable singing direction these values decreased and varied between 128m for the best case (Olympus and USB) and 64m for the worst (Sytech, Table 2). We also detected large differences between recorders and singing direction in relation to effective continuum distance (Table 2). When singing direction was favourable, this value was relatively high and varied between 256m (USB)

TABLE 2

Cross-training of each recogniser and time needed to perform sound-analyses in lab for each recorder. Total number of detections, total number of true positives (% of songs recognised in respect to total songs broadcast by playback is shown between brackets), total number of false positives, maximum detection distance and effective continuum distance per recorder are shown separately for tests performed with favourable and unfavourable singing directions.

[‘Cross-training’ de cada reconocedor y tiempo empleado para realizar los análisis de sonido en laboratorio para cada grabador. El número total de detecciones, número de verdaderos positivos (% de cantos reconocidos respecto al total de cantos emitidos por el ‘playback’ se muestran entre paréntesis), número total de falsos positivos, máxima distancia de detección y distancia continua efectiva se muestran por separado para los tests desarrollados bajo condiciones favorables y desfavorables de dirección del canto.]

		Favourable singing direction					
	Cross-training	Time (s)	Total events	True positives	False positives	Max. distance	Effect. distance
SM2	78.1 ± 5.8	451	46	34 (29.1%)	12	256	192
Olympus	73.9 ± 14.6	235	20	12 (10.3%)	8	128	64
Sony	75.6 ± 8.1	910	67	18 (15.4%)	49	256	192
Sytech	75.8 ± 12.1	226	22	19 (16.2%)	3	224	224
USB	72.2 ± 13.4	525	62	35 (29.9%)	27	256	256
TOTAL			217	118 (20.2%)	106		
		Unfavourable singing direction					
	Cross-training	Time (s)	Total events	True positives	False positives	Max. distance	Effect. distance
SM2	78.1 ± 5.8	451	30	17 (14.5%)	13	96	96
Olympus	73.9 ± 14.6	235	10	5 (4.3%)	5	128	1
Sony	75.6 ± 8.1	910	48	11 (9.4%)	37	96	96
Sytech	75.8 ± 12.1	226	10	9 (7.7%)	1	64	64
USB	72.2 ± 13.4	525	33	18 (15.4%)	15	128	128
TOTAL			131	60 (10.3%)	72		

and 64m (Olympus), but it decreased under unfavourable singing directions up to 128m (USB) and 1m (Olympus, Table 2).

The GLMM showed that number of songs detected decreased significantly with distance (Figure 2 and Table 4). Distances greater than 128m showed similar detection success,

attending to Tukey *post-hoc* tests (see Supplementary Material, appendix 4). Tests performed under favourable singing directions detected a significantly greater number of songs when compared to those carried out with unfavourable singing direction (Figure 2). More specifically, tests performed under

TABLE 3

Estimates of a general linear mixed model (GLMM) testing the effect of recorder type, distance to playback source and singing direction on the probability of detecting a Dupont's Lark song. Summary table of type-II partitioning of variances performed for each factor is also shown.

[Resultados de un modelo lineal mixto general (GLMM) que testa el efecto del tipo de grabador, distancia al altavoz y dirección de emisión en la probabilidad de detectar un canto de alondra ricotí. También se muestra una tabla resumen de la partición de la varianzas (tipo-II) para cada factor.]

Response variable	Estimate	Std. Error	Z value	Pr(> z)	
(Intercept)	-0.469	0.381	-1,231	0.218	
Distance 32	-0.970	0.289	-3,356	< 0.001	
Distance-64	-1.767	0.317	-5,574	< 0.001	
Distance-96	-2.376	0.353	-6,731	< 0.001	
Distance-128	-2.545	0.367	-6,935	< 0.001	
Distance-160	-3.739	0.515	-7,260	< 0.001	
Distance-192	-3.366	0.456	-7,382	< 0.001	
Distance-224	-4.710	0.750	-6,280	< 0.001	
Distance-256	-3.980	0.561	-7,094	< 0.001	
Recorder-SM2	1.714	0.344	4,983	< 0.001	
Recorder-Sony	0.763	0.362	2,108	0.035	
Recorder-Sytech	0.710	0.368	1,929	0.051	
Recorder-USB	1.822	0.343	5,312	< 0.001	
Singing direction-Unfavourable	-1.081	0.203	-5,325	< 0.001	
Fixed effect	df	Sum Sq	Mean Sq	F	P
Distance	8	113.8	14.2	14.2	< 0.001
Recorder	4	35.7	8.9	8.9	< 0.001
Singing direction	1	28.3	28.3	28.3	< 0.001

favourable singing directions detected significantly more songs at four of the considered distances (64, 128, 160 and 192m). Conversely, the number of songs detected for the remaining distances was unrelated to singing direction (Supplementary Material, appendix 5).

Cost-effectiveness assessment

There were large differences in the total scores obtained per recorder. USB and SM2 had the highest scores, while the other three recorders compared poorly (Table 4), and thus were excluded from the cost-effectiveness

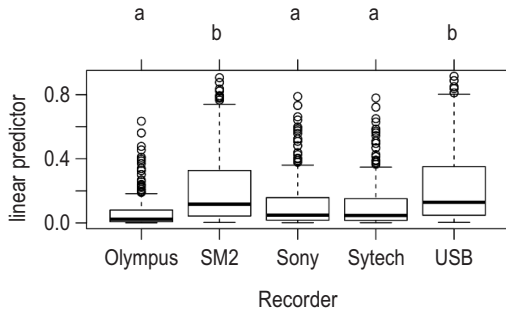


FIG. 1.—Results of the Tukey *post-hoc* test for the factor ‘Recorder’. Detection significantly varied among recorders, SM2 and USB detecting significantly more songs than the other recorders, and with no differences between them. Different letters mean significant differences in the detection success after Tukey test.

[Resultados del test *post-hoc* de Tukey para el factor Grabador. La detección varió significativamente entre los grabadores, SM2 y USB detectaron significativamente más cantos que el resto de grabadores, sin diferencia entre ellos. Diferentes letras muestran diferencias significativas en el éxito de detección tras el test de Tukey.]

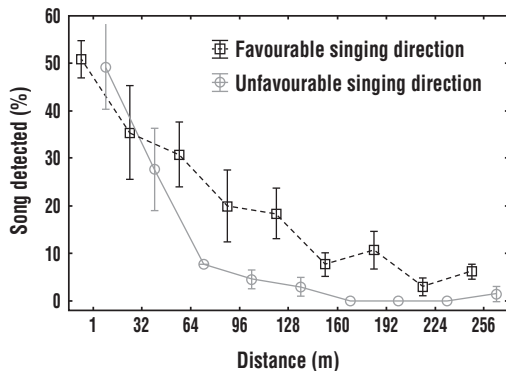


FIG. 2.—Mean percentage of songs detected (\pm SE) as a function of playback distance. Results are shown separately for tests performed with favourable and unfavourable singing directions. [Porcentaje medio de cantos detectados (\pm SE) en función de la distancia del reclamo. Los resultados se muestran por separado para los test desarrollados con direcciones de canto favorables y desfavorables.]

assessment. According to their effective continuum distance under an unfavourable singing direction (limiting distance for monitoring studies) 16 USB or 25 SM2 recorders would be needed for monitoring a potential habitat patch of 1km². This would imply a total cost of 2,880 euros using USB and 27,500 euros using SM2.

DISCUSSION

Our results revealed large differences among different recorders. In general, and in agreement with our predictions and previous studies, recorder performance decreased with increasing distance from sound source (e.g. Rempel *et al.*, 2013; Yip *et al.*, 2017). Maximum and effective detection distance differed greatly among devices, and that difference may be greater than 100m in some instances. This result highlights the need for assessing the effectiveness of selected recorders before considering an effective distance for monitoring programmes. We have also found that the probability of detecting songs differed with singing direction, as expected. More vocalisations were detected at intermediate-long distances (64–192m) in tests performed at favourable than with unfavourable singing directions. This could be because singing direction had no effect on recording quality at short distances, while at distances greater than 192m, songs were uttered too far and therefore only occasionally detected even with favourable singing direction. To our knowledge, this is the first study showing the strong influence of singing direction on detection distance by recorder, which highlights the need to include this factor in further research and to consider it when estimating effective detection radius of acoustic recorders. We are aware, however, that our results are based on one replicate per song type at each distance, and in a single-species study, and that detection distances would differ between

TABLE 4

Number of songs detected per recorder and distance. Percentage of songs recognised in respect to total songs broadcasted by playback is between brackets. Results are shown separately for tests performed with favourable and unfavourable singing directions. The total Score obtained per recorder is also shown. A score was given at each distance and singing direction according to detection and success rate: 3 points when detection or success rate was > 50%, 2 points when detection or success rate was between 50% and 25%, 1 point when detection or success rate was < 25% and 0 points when no songs were detected or correctly classified.

[Número de cantos detectados para cada grabador y distancia. El número total de cantos reconocidos respecto al total de cantos emitidos por el 'playback' está entre paréntesis. Los resultados se muestran por separado para los tests desarrollados bajo condiciones favorables y desfavorables de dirección del canto. También se muestra la puntuación total obtenida para cada grabador. A cada distancia y dirección del canto se le asignó una puntuación según su tasa de detección y éxito: 3 puntos cuando la tasa de detección o éxito fue superior al 50%, 2 puntos cuando la tasa de detección o éxito osciló entre 50% y 25%, 1 punto cuando la tasa de detección o éxito fue inferior al 25% y 0 puntos cuando ningún canto fue detectado o clasificado correctamente.]

Recorder	TOTAL SCORE	Singing direction	Distance									
			1m	32m	64m	96m	128m	160m	192m	224m	256m	
SM2	56	Favourable	7 (53.8%)	6 (46.2%)	6 (46.2%)	6 (46.2%)	5 (38.5%)	1 (7.7%)	2 (15.4%)	0 (0%)	1 (7.7%)	
		Unfavourable	9 (69.2%)	6 (46.2%)	1 (7.7%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
Olympus	31	Favourable	7 (53.8%)	2 (15.4%)	2 (15.4%)	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
		Unfavourable	3 (23.1%)	0 (0%)	1 (7.7%)	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
Sony	35	Favourable	6 (46.2%)	3 (23.1%)	2 (15.4%)	2 (15.4%)	2 (15.4%)	1 (7.7%)	1 (7.7%)	0 (0%)	1 (7.7%)	
		Unfavourable	6 (46.2%)	3 (23.1%)	1 (7.7%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
Sytech	47	Favourable	5 (38.5%)	3 (23.1%)	4 (30.8%)	2 (15.4%)	2 (15.4%)	1 (7.7%)	1 (7.7%)	1 (7.7%)	0 (0%)	
		Unfavourable	5 (38.5%)	3 (23.1%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	
USB	57	Favourable	8 (61.5%)	9 (69.2%)	6 (46.2%)	3 (23.1%)	2 (15.4%)	2 (15.4%)	3 (23.1%)	1 (7.7%)	1 (7.7%)	
		Unfavourable	9 (69.2%)	6 (46.2%)	1 (7.7%)	1 (7.7%)	1 (7.7%)	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)	

species and habitat type according to their signal characteristics and sound propagation (Oppel *et al.*, 2014; Yip *et al.*, 2017). Therefore relative differences found between recorders could also change according to the different habitats and species tested.

The cost-effectiveness assessment also showed large differences between recorders and revealed that a medium priced unit (USB) performed as well as the costliest device (SM2). Rempel *et al.* (2013) also detected similar performance between the cheapest (Zoom H2) and the most expensive unit (IR-C1). Our cost-effectiveness assessment suggests that a USB recorder could be a good choice for long-term studies, where costs are a limiting factor and sound quality recording (USB does not record on wav format) may not be a priority. However, cost performance should not be the only parameter used to select a recording system for research and monitoring studies (Rempel *et al.*, 2013). According to our results, USB was also the device with the largest effective continuum distance, thereby confirming its suitability for long-term studies, given the lower number of devices, and thus shorter period for analysis, needed, to cover certain areas.

We are aware that we based our cost-effectiveness assessment on success and detection rate across distance. This variable is quite important to avoid biases when estimating animal abundance or site occupancy and to estimate how many devices are needed to survey a habitat patch (Pérez-Granados *et al.*, 2018a). A low detection distance can be a handicap when covering large areas (Efford *et al.*, 2009) but, depending on the objectives and/or the limitations of each study, other factors, such as recording time, battery life, available memory, programmability and portability should also be considered. After selecting the USB as the most effective recorder to build our own recorder, we needed to make a significant number of alterations to turn it into a functional ARU

(see procedure and final recorder in Supplementary Material, appendix 6). However, it is not available for purchase by the general public. Moreover, in the last few years, new ARUs that are smaller, cheaper, available to the general public and with more functions and capabilities than self-adapted USB, have appeared (Hill *et al.*, 2018; Beason *et al.*, 2018).

ARUs are a useful tool for monitoring wildlife and there are many recorders with very different configurations (ability to make different recordings in each channel, GPS location, automatic modification of starting time according to sunrise, sample rate selection, etc.). Researchers will probably base their selection on cost, expert opinion or previous experience, among other factors, but our study suggests that this may not be the only approach. Instead, we also recommend that the field performance of a set of different recorders should be evaluated before choosing one for a monitoring program, since effectiveness may greatly differ among them.

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SUPPLEMENTARY ELECTRONIC MATERIAL

Additional supporting information may be found in the on-line version of this article. See volume 66(2) on www.ardeola.org

Appendix 1: Sonogram of a typical Dupont's Lark song.

Appendix 2: Settings used for recogniser creation in Song Scope.

Appendix 3: Total number and percentage of true positives of songs recognised in respect to total songs uttered by playback under favourable singing direction.

Appendix 4: Results of the Tukey *post-hoc* test for the factor *Distance*.

Appendix 5: Comparison between the total number of songs detected at each distance by all ARUs with favourable and unfavourable singing directions.

Appendix 6: Pictures where the recorder (black USB), the outdoor microphone and the long-life battery can be seen at top.

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A cost-effective protocol for monitoring birds using autonomous recording units: a case study with a night-time singing passerine

Cristian Pérez-Granados ^{1a}, Gerard Bota ^{2b}, David Giralt ^{2b} and Juan Traba ^a

^aTerrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain; ^bBiodiversity and Animal Conservation Lab, Landscape dynamics and biodiversity Programme, Forest Sciences Center of Catalonia (CTFC), Catalonia, Spain

ABSTRACT

Capsule: We describe an effective monitoring protocol for detecting wildlife presence using autonomous recording units (ARUs) under different density scenarios.

Aims: To describe an effective protocol for monitoring a night-time singing passerine, the Dupont's Lark *Chersophilus duponti*, using ARUs.

Methods: We estimate, using both simulations and field-collected data, the number of devices needed to reliably detect the species under different density scenarios and to assess recording time and the number of working days needed to ensure species detection. We placed between four and six ARUs in three Dupont's Lark populations with different bird densities. Devices were programmed to record for 90 minutes per day for four consecutive days. ARUs were deployed between April and June of 2017.

Results: We found large differences in the number of recorders needed to detect species presence under different density scenarios, with more ARUs required in less dense populations. The number of ARUs needed to be differed between estimates obtained by simulations and with field data. This could be related to movements of the monitored species while they were singing. According to our results, the monitoring period for detecting the Dupont's Lark could be as little as one hour of recording (from one hour before dawn to dawn) and two monitoring days, the minimum monitoring time needed to detect the species in all populations surveyed, regardless of density scenarios.

Conclusion: Our results cannot be directly extrapolated to other singing species since singing behaviour and characteristics greatly differ between species. We describe five logical steps to develop effective wildlife monitoring protocols using ARUs for detecting species presence, which may be helpful for future studies and with different species.

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Due to the global decline in biodiversity, there is an urgent need for rapid and effective monitoring programs to assess the conservation status of species and to maximize the effectiveness of conservation effort (Brandes 2008, Potamitis *et al.* 2014). The minimum knowledge needed for species conservation is whether target species are present within an area, and how they are distributed (Li *et al.* 2010). Furthermore, well-designed monitoring programs with standardized equipment and protocols can provide highly repeatable and reliable data to estimate population trends (Pereira & Cooper 2006, Buxton *et al.* 2013). In this context, the use of acoustic Autonomous Recording Units (ARUs hereafter) as a tool for monitoring a wide range of taxa have rapidly increased in the last few years (e.g. Alquezar & Machado 2015, Heinicke *et al.* 2015, Hedley *et al.* 2017, Van Wilgenburgh *et al.* 2017). ARUs function on their own while offering a suitable and efficient alternative

for wildlife monitoring (see review for birds in Leach *et al.* 2016). Moreover, monitoring programs based on ARUs are not subject to many of the biases of traditional field monitoring (detection differences between observers, temporal bias, etc.), since this is a non-invasive technique that creates a permanent, repeatable and archivable record of surveys (Acevedo & Villanueva-Rivera 2006, Brandes 2008), which can be checked by different observers.

The use of ARUs has some disadvantages that must not be overlooked and may even rule out their use for monitoring purposes. Recorders usually have lower sensitivity than a human listener (Hutto & Stutzman 2009, Yip *et al.* 2017), which may be relevant for monitoring rare species or those with reduced vocal activity, and they have costs related to device acquisition. However, the main obstacle that has hampered the widespread use of ARUs is the large amount of expert time needed to analyse recordings

(Hutto & Stutzman 2009, Digby *et al.* 2013). In recent years, significant progress has been made in audio signal processing and automated signal recognition, making it possible to process large data sets in a timely manner (see review in Knight *et al.* 2017), and therefore facilitating the use of ARUs as an automated non-invasive monitoring technique (Buxton *et al.* 2013).

The number of ARUs deployed per site for wildlife monitoring depends on the radius within which it is possible to effectively detect distant songs of monitored species. This radius can differ greatly among recorders, habitats and species (Digby *et al.* 2013, Bota *et al.* 2017, Yip *et al.* 2017). Likewise, monitoring time using ARUs, including recording time per day and number of days of monitoring, greatly differs between model species and study objectives. Monitoring periods in previous studies have varied between a few minutes to several hours per day (e.g. 10 min in a single day, Celis-Murillo *et al.* 2012; 6 h per day during seven consecutive months, Heinicke *et al.* 2015), making data analysis difficult even with the help of automated signal recognition software. The monitoring protocol can even differ between studies with a similar purpose, causing bias and making comparisons difficult. For example, Holmes *et al.* (2015) monitored bird presence by obtaining eight recordings each day (recording length varied from 15 to 75 minutes) during a mean number of ten work-days, while Goyette *et al.* (2011) monitored bird presence with a single recording but at least 48 h of continuous recording. The development of effective and standardized monitoring protocols will lead to greater time and economic efficiency and, when possible, to a greater degree of standardization in data collection (Celis-Murillo *et al.* 2009, Venier *et al.* 2012). This may allow the collection of repeatable and robust data that is comparable over time and between sites, which may be used as an effective technique for monitoring habitat quality (Lin *et al.* 2017) and changes in species presence or abundance (Buxton & Jones 2012, Buxton *et al.* 2013).

Although a protocol for the use of ARUs should be a pre-requisite for any monitoring program, we found no study assessing the recording time and number of ARUs needed for monitoring species presence within a patch. In this paper, we aim to describe an effective protocol for detecting the presence of a night-time singing passerine using ARUs. We chose the Dupont's Lark *Chersophilus duponti* as a study model. Traditional field surveys developed for monitoring the Dupont's Lark have always been based on auditory contacts due to its mainly nocturnal singing and evasive day-time behaviour (Pérez-Granados & López-Iborra 2017). Moreover, European Dupont's Lark populations are

patchily distributed over a large number of remote sites throughout peninsular Spain at different densities (Suárez 2010), which makes a spatially homogenized and well-distributed sampling effort difficult. The species is classified as 'Near Threatened' in the IUCN Red List (BirdLife International 2017) and as 'Vulnerable' in the European Red List of Birds (BirdLife International 2015). Therefore, we consider acoustic monitoring with ARUs as especially well suited to improving monitoring programs of this species. This is evidenced by the fact that some managers have begun to suggest the use of ARUs as an aid for monitoring the Dupont's Lark (authors' own data).

The main objectives of this paper were to (1) test and describe the procedure of an effective monitoring protocol using ARUs and automated song analyses for a night-time singing passerine, the Dupont's Lark, under different plausible density scenarios; (2) determine the minimum number of recorders needed to detect species presence using virtual and field data under different density scenarios; and (3) estimate the minimum recording length per night and number of nights of monitoring needed to detect species presence under three different bird density scenarios. While our study was focused on the Dupont's Lark, we describe a series of steps to develop an effective monitoring protocol that may be useful for different taxa.

Methods

Simulations under different density scenarios

We mathematically estimated the minimum number of ARUs needed to reliably detect the presence of the Dupont's Lark within a virtual 100 ha habitat patch. For the experiment, we considered an effective continuum distance of 128 m, which is the effective continuum distance estimated for our ARU for the studied species for opposite singing direction (see Field Recording section for how continuum distance was estimated). By using a virtual space, we sequentially added virtual ARU devices one by one in up to 16 locations (constrained to not overlap in their effective continuum distance). In this way, the entire virtual area would eventually be covered. We simulated the presence of singing individuals under four different plausible bird density scenarios (0.1, 0.25, 1, and 4 males/10 ha) and built accumulation curves of detectability rate, as a function of the number of ARUs placed. We assumed that each virtual male was a singing male. We considered 0.1 males/10 ha as an example of habitat patches occupied with a much

reduced presence of the species. The density scenarios of 0.25 and 1 males/10 ha correspond to the average Dupont's Lark density estimated in Spain (0.27 males/10 ha, Suárez 2010), and to that found in core areas, such as the Layna moorland (1 males/10 ha, Garza *et al.* 2005), respectively. We used the density of 4 males/10 ha as an example of the highest densities estimated for the species in specific habitat patches (Suárez 2010).

Study area

The study area comprised three Dupont's Lark populations located in northeast Spain (Figure 1). The Timoneda d'Alfés (hereafter Alfés) population was located in Lleida (Catalonia, 41.30°N, 0.37°E, Figure 1), occupying a low shrub-steppe dominated by Thyme *Thymus vulgaris*, Shepherd's Tea *Sideritis scordioides* and rushrose (*Helianthemum* spp.). The Barcones (41.17°N, 2.49°W) and Alcubilla de las Peñas (hereafter Alcubilla; 41.15°N, 2.31°W) populations were located in Soria (Castile and León, Figure 1), both on natural steppes dominated by small shrubs, such as thyme (*Thymus* spp.), broom (*Genista* spp.) and lavender (*Lavandula* spp.). We selected these sites because they had a similar patch size (range 70–100 ha) but different Dupont's Lark densities during 2017 (see below).

Bird data

The abundance of Dupont's Lark males at each site was estimated by a mapping method, based on four visits following line transects with a 500 m maximum detection band on each side of the observer (Pérez-Granados & López-Iborra 2017). Censuses were performed from 10 April to 20 June 2017 and only males were considered since females are presumed to be vocally inactive in this species. The distance of singing males from the observer was estimated acoustically and its location was recorded by a global positioning system (GPS) handset. The location of singing males detected in each visit were mapped (using ArcGIS 9.3, ESRI 2008) and a territory was defined when at least two registrations of a singing male (Bibby *et al.* 2000) were within a radius of 100 m, following the proposed methodology for counting Dupont's Lark (Pérez-Granados & López-Iborra 2017). We assumed a probability of detection equal to 1, since Dupont's Lark songs may be heard from up to 1 km away (Laiolo *et al.* 2007), and the entire habitat patches were covered during the censuses. Therefore, density (males/10 ha) in each population was calculated by dividing the total number of males estimated by patch

size of suitable habitat. Censuses were carried out by walking at a constant speed (1–3 km) on dry and windless days. Census time was from 60 minutes before dawn to dawn, the maximum singing activity period for the species (Pérez-Granados & López-Iborra 2017). In Alcubilla, we estimated a mean density of 0.29 males/10 ha (2 males in 70 ha), 0.9 males/10 ha in Alfés (9 males in 100 ha) and 3.62 males/10 ha in Barcones (29 males in 80 ha). Therefore, we used these as study models of Dupont's Lark populations with low (Alcubilla), medium (Alfés) and high density (Barcones).

Field recording using autonomous recording units

We custom designed our ARUs after performing field tests and a cost-effectiveness assessment for five acoustic recorders (Traba *et al.* 2017). In the assessment, we broadcasted Dupont's Lark songs from nine fixed locations from 1 to 256 m, either towards or against the recorder positions aiming to estimate parameters such as maximum detection distance or effective continuum distance (distance at which at least one Dupont's Lark song was automatically detected in all distance intervals, with no distance lag; own data). ARUs consisted of a USB Voice Recorder SK-001 with processor AC1517D72772-C and one integrated and single-channel electret microphone. Recorders were powered by 12 V/1.8 mAh Lipo batteries (minimum 15-hour autonomy recording continuously) and were also connected to a digital timer to program recorder activation and registration at selected times. Recordings were collected on 4 Gb microSD memory cards capable of storing 60 hours of continuous data. Equipment was protected in easily portable and weatherproof plastic boxes (60 × 80 × 160 mm) made cryptic by painting them greenish-brown with spots.

We placed six ARUs in each monitored population during the breeding season of 2017 (April–May). However, only four units worked properly in the Alfés population due to technical issues. Locations of recorders were selected to cover as much of the patch area as possible but were constrained to not overlap in their radius of effective continuum distance. ARUs were ground-located with omnidirectional microphones in an upward position and horizontally separated by at least 50 cm from natural vegetation to minimize sound blockage (Rempel *et al.* 2013). ARUs were left in each population for four consecutive days, and a digital timer was programmed to record for 90 minutes, from one hour before dawn to 30 minutes after dawn (Pérez-Granados & López-Iborra 2017). Recording time covers the maximum singing activity period of the species. Recordings were split into 30-minute length files for

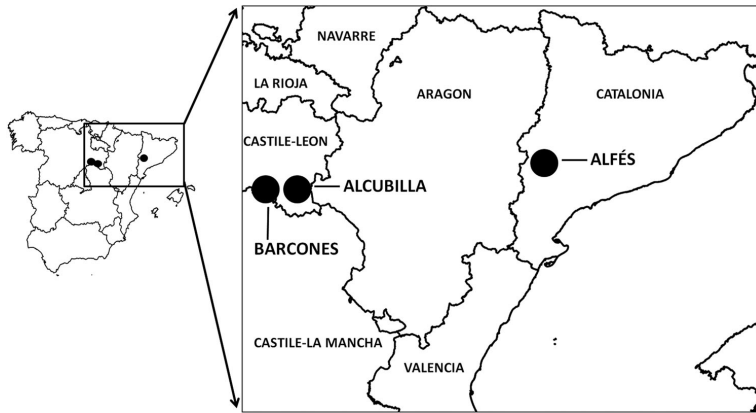


Figure 1. Location in northeast Spain of the three Dupont's Lark populations surveyed during the breeding period of 2017.

analytical purposes. Daily times of sunrise at the geographic location of the study areas were obtained from the Spanish Ministry of Development (<http://www.fomento.es>). We used a sample rate of 44.1 kHz and 16 bits in stereo mode for all recordings. Recordings were obtained in mp3 format and needed a post-transformation to wav format for analysis in Song Scope (see below).

Recording analyses

Automatic song recognition of Dupont's Lark presence was performed using Song Scope 4.1.5 (Wildlife Acoustics 2011), which is one of the most efficient programs available for automatic song recognition (Knight *et al.* 2017). We created a target signal for Dupont's Lark songs with the help of the spectrogram visualization tool in the software, which was used as a recognizer (Waddle *et al.* 2009). To build the recognizer, we used the final sequence of the species' song; the 'whee-ur-wheeee' song described by Cramp (1988), which is easily identified in a spectrogram and

thus, can be easily detected by an automated classification software (Figure 2). To build the recognizer, we adjusted sample rate and frequency ranges aiming to isolate the target signal and removed all lower and higher amplitude events (Waddle *et al.* 2009, Towsey *et al.* 2012). Song Scope output reported a number of events that matched the target signal. These events were visually and/or acoustically checked by the same researcher, to confirm Dupont's Lark presence when needed.

Statistical analyses

We mathematically assessed the minimum number of ARUs needed to reliably detect the presence of the Dupont's Lark under the four different density scenarios. We considered that the presence of the species was reliably detected when we reached 90% probability of detection. We estimated the number of times an individual was detected through 1000 randomizations using random locations for individuals and fixed locations for ARUs. We considered detection as the event when a virtual male fell within the effective radius of an ARU (128 m). Males were created to be static (i.e. not moving within a buffer), so these results should be considered as a minimum approximation to detection probability. The probability of detecting species presence for each simulated density scenario was evaluated through logistic regression using presence/absence of the species as a dependent variable and number of ARUs as a predictor variable. Data analyses were conducted in R 3.4.1 (R Core Team 2016). We also created accumulation curves of detectability rate with field data collected in the three populations surveyed as a function of the number of ARUs deployed.

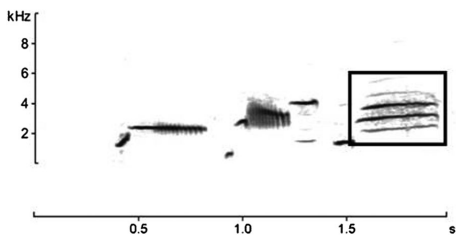


Figure 2. Sonogram of a typical Dupont's Lark song. Rectangle shows the final song of the species, which was used for building the recognizer.

Results

Simulations under different density scenarios

The number of ARUs needed to detect species presence in the considered virtual habitat patch (100 ha) differed greatly between the four density scenarios (Figure 3). In the lowest density scenario (0.1 males/10 ha), 16 units were needed to ensure the detection of the species, while in those scenarios with a mean density of 0.25 and 1 males/10 ha, nine and four ARUs were sufficient to detect species presence with 90% of confidence, respectively (Figure 3). Only one ARU was required to reach a similar confidence percentage when density increased to 4 males/10 ha, respectively (Figure 3).

Estimation of ARUs needed to detect the species under real situations

We found large differences in the number of ARUs needed to accurately detect the presence of the species between the three monitored populations. In the high density population, the species was detected in all locations where ARUs were placed (Figure 4). However, two and four devices were needed to reliably detect Dupont's Lark presence in the medium and low-density populations, respectively (Figure 4).

Recording time and number of nights

Dupont's Lark presence was detected in 39 out of 64 ARUs/night. In 37 of the cases (95%), the species was detected during the first night after ARUs were deployed. However, twice (5%) the species was not detected until the second night of monitoring, both cases in the low-density population. Therefore, the species was always detected within the first two nights of monitoring in all studied populations.

In relation to recording time, the species was always detected in the first hour of recording ($N=39$), from one hour before dawn to dawn, with no new detections in the third recording phase (from dawn to 30 minutes after dawn). The best time for detecting the species was the period between one hour and 30 minutes before dawn, when the species was detected in 95% of the cases with known presence ($N=37$), followed by the period between 30 minutes before dawn to dawn (82%, $N=32$) and lastly the 30 minutes after dawn (46%, $N=18$).

Discussion

In this paper, we describe for the first time a protocol for songbird monitoring using ARUs, in which we estimate

the number of devices, the number of nights and recording time needed to ensure species detection. Our results suggest that this validation is needed and useful before starting a monitoring program with automatic acoustic recorders, since the probability of detection of the monitored species may vary greatly depending on bird singing characteristics, habitat and the number of ARUs deployed, recording time and the number of nights of monitoring. Moreover, we present a test of the number of ARUs needed to detect species presence based on simulations and field data. Our simulations showed that the number of devices needed to detect species presence depends on bird density ranging from 1 ARU per 100 ha under a high bird density scenario (4 males/10 ha) to 16 ARUs per 100 ha under the lowest density scenario (0.1 males/10 ha). We also found large differences in the number of ARUs required to detect species presence in the three surveyed populations according to mean bird density, with an increasing number of devices needed under lower bird density scenarios. The estimated number of ARUs needed to detect the monitored species in the highest density scenarios by both simulations and field data were in agreement, and indicate that Dupont's Lark or a hypothetical species may be properly detected with a low number of devices and effort. However, estimates of the number of ARUs needed to reliably detect species presence in simulations performed with 0.25 and 1 male/10 ha were around two times greater than those obtained in the field in populations with similar densities. This contradiction highlights the need for field-testing results obtained by simulations under controlled situations. We are aware that much of the variation found can be explained by the fact that we considered virtual static individuals in simulations, while Dupont's Lark tends to move while singing (Pérez-Granados & López-Iborra 2017, David Serrano pers. com.), which may increase the probability of individual detection.

Our results also highlight that a period of one hour of recording, from one hour before dawn to dawn, and two monitoring nights is sufficient time for detecting the presence in all surveyed Dupont's Lark populations, regardless of population density. However, singing activity and singing behaviour differ greatly between species, which suggests a necessity to adequately estimate the maximum period of singing activity for monitored species before planning any monitoring programme, so as to increase the effectiveness and reduce time and cost of data analyses. For example, the first 30 minutes after dawn, when many of the European passerines are most vocally active, was the worst time for detecting the Dupont's Lark during the monitoring period, due to its mainly nocturnal singing

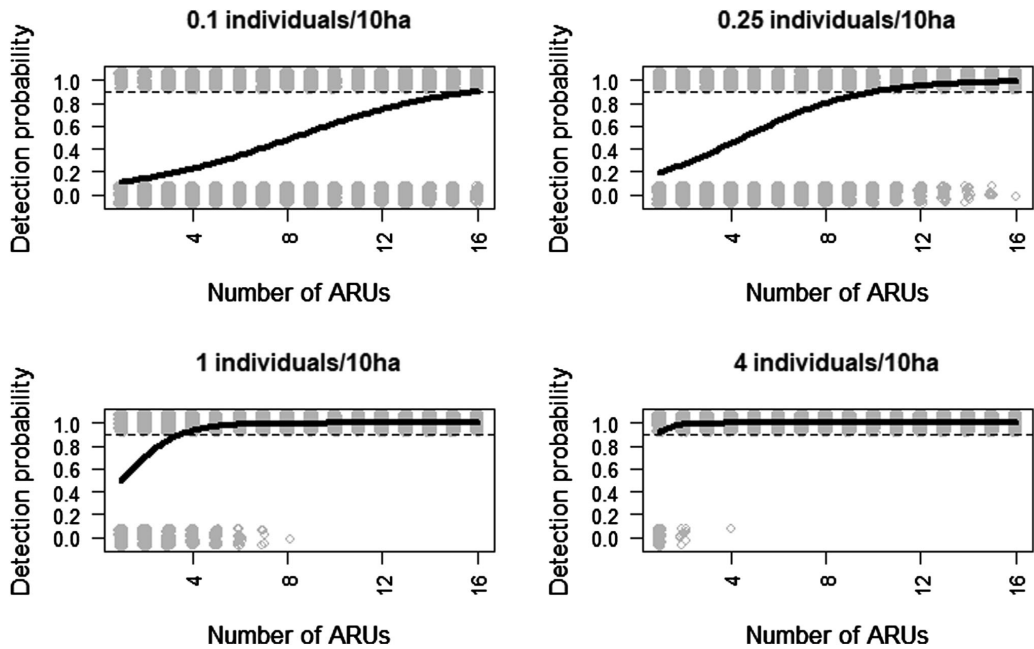


Figure 3. Estimates of the probability of detecting the presence of the Dupont's Lark under four density scenarios in a 100 ha patch, given a different number of autonomous recording units (ARUs) deployed. Estimates were obtained through logistic regression using presence/absence as a dependent variable and the number of ARUs as a predictor variable. Grey points show the estimates (presence/absence) derived from 1000 randomizations using different random locations per individuals and regular locations for ARUs. The dotted line marks 90% probability to detect the species.

behaviour (Laiolo *et al.* 2007, Pérez-Granados & López-Iborra 2017). We also found differences in the number of nights of monitoring needed to reliably detect Dupont's Lark as a function of bird density. The species was always detected during the first night of monitoring in

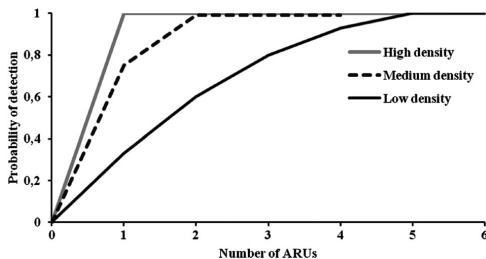


Figure 4. The probability of detection of Dupont's Lark as a function of the number of autonomous recording units (ARUs) deployed in three populations surveyed during the 2017 breeding period in Spain. Populations had different densities and curves were built for each density scenario separately. In the population of medium density only four ARUs were used and thus the curve is shorter.

the high and medium density populations, but it was not detected until the second night in the lowest density population. This could be related to different singing behaviour between populations according to mean density, since Dupont's Lark song rate and song diversity are positively related to population size (Laiolo & Tella 2005, 2007, Laiolo *et al.* 2008). Likewise, the number of monitoring days needed to detect species presence also differs between species according to their singing behaviour. During spring 2017, we used the same ARU for detecting the presence of Western Capercaillies *Tetrao urogallus* in singing leks, and in some leks, the species was not detected until the seventh day of monitoring. In other leks, however, males were detected during the first day of monitoring (D. Guixé pers. comm.). Our results show that field tests to determine recording time and a number of nights needed for effective monitoring must be conducted before starting any monitoring programme using ARUs with other species, since they seem to differ greatly between considered species. Our results may be used as a source of comparison and protocol.

The use of ARUs for wildlife monitoring has rapidly increased in recent years (e.g. Heinicke *et al.* 2015, Hedley *et al.* 2017, Van Wilgenburgh *et al.* 2017), and with the rapidly decreasing costs of advanced technology (Hill *et al.* 2018), their use has already become an important tool in applied field studies (see review for birds in Shonfield & Bayne 2017). Therefore, the description of effective monitoring protocols can be useful to future studies and to improve monitoring programmes. We are aware that our study involved only a single model species, and surveys were carried out only during the breeding season, when detection probability is at its maximum. Therefore, we propose a minimum protocol for detecting the presence of a songbird species using ARUs. Our protocol is based on five steps that could be easily adapted to other species or contexts: (1) Estimate the effective distance at which the ARU is able to detect the species selected for monitoring; (2) estimate the number of nights/days of monitoring required to detect species presence with high probability; (3) assess the number and cost of devices needed for species detection under real field conditions; (4) limit the recording time to the period when singing activity is at a maximum; and (5) evaluate the cost and time required for data analyses. All recommendations should be assessed before beginning any monitoring programme, to reduce costs, keep the amount of recording time at a minimum and maximize monitoring effectiveness.

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ORCID

Cristian Pérez-Granados  <http://orcid.org/0000-0003-3247-4182>

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Short communication

Vocal activity rate index: a useful method to infer terrestrial bird abundance with acoustic monitoring

CRISTIAN PÉREZ-GRANADOS,^{1,2*†} GERARD BOTA,^{3†} DAVID GIRALT,³ ADRIÁN BARRERO,¹ JULIA GÓMEZ-CATASÚS¹ DANIEL BUSTILLO-DE LA ROSA¹ & JUAN TRABA^{1,4}

¹Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain

²National Institute for Science and Technology in Wetlands (INAU), Computational Bioacoustics Research Unit (CO.BRA), Federal University of Mato Grosso (UFMT), Cuiabá, Mato Grosso, Brazil

³Biodiversity and Animal Conservation Lab, Forest Science and Technology Center of Catalonia (CTFC), Solsona, Catalonia, Spain

⁴Centro de Investigación en Biodiversidad y Cambio Global (CIBC-UAM), Universidad Autónoma de Madrid, Madrid, Spain

Autonomous recording units have been widely used in a large number of bird studies in recent years, but challenges remain in estimating abundance based on acoustic monitoring. We tested whether vocal activity rate index (VAR; the number of songs per unit time for a species), recorded using autonomous recording units, was related to population abundance in two terrestrial bird species, the European Bee-eater *Merops apiaster* and the Dupont's Lark *Chersophilus duponti*. We took recordings at sites where censuses were also carried out to estimate local populations around recorders. We found a positive and significant relationship for the two monitored species. Although our results are not conclusive, the strong and significant relationship found for both monitored species suggests that VAR may be used to infer bird abundance around recorders in terrestrial species. We describe five logical steps for using the VAR with autonomous recording units in other species to guide future studies.

*Corresponding author.
Email: cristian.perez@ua.es
Twitter: @Gerard_Bota

†Equal contributors.

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The use of autonomous recording units (hereafter ARUs) for monitoring wildlife has increased widely in recent years (reviewed by Sugai *et al.* 2019). ARUs are a suitable alternative to traditional field surveys for detecting species presence, and for estimating species richness or population densities (Darras *et al.* 2018). ARUs have several advantages over traditional field surveys. The devices can be deployed and retrieved at any time, making fieldwork more flexible and avoiding disturbances to vocal activity, as in human presence surveys (Venier *et al.* 2012). Researchers can cover large spatial and temporal scales simultaneously, eliminating temporal differences between samples, at a relatively low cost (Alquezar & Machado 2015) and they can operate unattended in remote locations or areas with limited visibility (e.g. rain forest) to monitor cryptic species (e.g. nocturnal animals) and regardless of weather conditions (Pérez-Granados *et al.* 2018a). In addition, recordings can be automatically scanned, avoiding biases due to researcher ability, leading to a greater degree of standardization in data collection (Venier *et al.* 2012) and ARUs create a permanent and archivable record of surveys that can provide useful information for future studies and comparison (Alquezar & Machado 2015).

Despite these advantages, there are some obstacles that have hampered the widespread use of ARUs, such as extra costs for acquiring devices. The recent development of open source, low-cost detectors may overcome these cost barriers (Hill *et al.* 2018). However, the time needed for post-recording analyses and the difficult-to-estimate abundances remain key obstacles to their use (Knight *et al.* 2017). Advances in computation and automated signal recognition software have led to the development of species-specific recognizers, including machine-learning processes, which allow researchers to manage the large volumes of acoustic data recorded (reviewed by Knight *et al.* 2017).

Commercial ARUs typically only include one, or at best two, microphones (Hill *et al.* 2018). Therefore, sound recordings obtained with ARUs provide little or no ability to determine sound direction in three dimensions, hindering the assessment of animal abundance due to the difficulty in mapping individual locations. For this reason, ARUs have been widely used to document community composition or species presence/absence but scarcely used for monitoring programmes, especially at large spatial scales, due to the difficulty of inferring densities for monitored species (Dawson & Efford 2009). To extend acoustic monitoring beyond activity monitoring, there is a need to estimate the relationship between

acoustic activity and population density (Desjonquères *et al.* 2018). Different methods have been described and tested to infer population densities from sound recordings (e.g. Dawson & Efford 2009, Hedley *et al.* 2017). Some authors have used an array of microphones to locate precise sounds, 'capturing' time in each microphone, allowing the generation of an 'observed distance', which can be analysed by conventional distance sampling methods (e.g. Dawson & Efford 2009). Other authors have used complex microphone systems to estimate sound direction with high precision (Hedley *et al.* 2017). However, the measurement of sound location, even with the use of multiple or complex microphones, is still imprecise and does not directly lead to an estimate of density (Hedley *et al.* 2017). Sebastián-González *et al.* (2018) have evaluated a new method to estimate bird population densities using omnidirectional ARUs. This method requires measuring cue rate from the target species, environmental conditions and an estimate of the distance of the individual to the recorder based on the power of the received sound (Sebastián-González *et al.* 2018). That study represents an effective method for estimating animal density, but a large number of parameters are needed to develop the method, so this approach is still resource-intensive. Moreover, its implementation is hampered in nocturnal and elusive animal species due to difficulties in obtaining quality recordings at precise distances.

An alternative, rapid and cheap method described to infer population densities using ARUs with omnidirectional microphones is the use of the vocal activity rate index (hereafter VAR), defined as the number of songs per time unit for the target species (Oppel *et al.* 2014). VAR has been used as an indicator of species activity and is expected to increase with population density (density-dependent; Farnsworth *et al.* 2004, Oppel *et al.* 2014). Previous studies have confirmed the existence of a positive relationship between VAR and abundance in different taxa, such as mammals or anurans (Nelson & Graves 2004, Barlow & Taylor 2005). In birds, VAR has been used to assess migration intensity (e.g. Farnsworth *et al.* 2004). Furthermore, the index has been successfully used to assess changes and estimate population size in seabirds (Buxton *et al.* 2013, Borker *et al.* 2014, Oppel *et al.* 2014). However, Zwart *et al.* (2014) found no relationship between the amount of vocalization recorded per ARU, and the abundance of the European Nightjar *Caprimulgus europaeus* in the only known study focused on VAR of breeding terrestrial birds.

In this study, we examined whether VAR, estimated by acoustic monitoring, can be used as an alternative method to infer abundance of two terrestrial bird species, European Bee-eater *Merops apiaster* and Dupont's Lark *Chersophilus duponti*, which have very different singing behaviour, diurnal activity patterns and habitat selection. We deployed ARUs in areas of known

estimated population size for each species to elucidate whether there is a significant relationship between vocal activity rate and the number of individuals around recorders. We predicted that vocal activity rate would increase with population size.

METHODS

Study species and singing behaviour

The European Bee-eater, a summer migrant, is a very vocal species that mostly calls at low frequencies (1–3 kHz) during daytime when foraging or migrating and can be heard at long distances. The contact call (Appendix: Fig. S1) is the most frequent vocalization of the species, a short, pleasant rolling sound ('pruuk') uttered for both sexes. It is simply structured but with some subtle differences between calls and individuals (Valera 2016). The Dupont's Lark is a resident, territorial passerine whose song (see Appendix: Fig. S1) ranges from low (< 2 kHz) to high frequencies (> 6 kHz). Songs are largely shared and repeated between neighbouring males before dawn, when they usually engage in countersinging disputes. Females are presumed to be vocally inactive in this species (Pérez-Granados *et al.* 2018b). The Dupont's Lark song usually ends with a common sequence, 'whee-ur-wheeee' (Pérez-Granados *et al.* 2018b). The use of ARUs has been assessed previously for monitoring the presence of Dupont's Lark (Pérez-Granados *et al.* 2018a,c).

Study area

The study area comprised sites located in both central and northeastern Spain. The European Bee-eater recordings were made in 10 different apiaries (composed of 30–50 beehives each) separated by 2–20 km located in farmland areas dominated by irrigated orchards, Alfalfa *Medicago sativa* and Maize *Zea mays* in western Catalonia (41°31'N, 0°55'W, northeastern Spain). The study area surveyed for the Dupont's Lark comprised 24 breeding sites located in central Spain (40°37'N, 3°09'W, Guadalajara and Soria provinces) and three sites in western Catalonia (41°32'N, 0°39'E, northeastern Spain). The habitat patches monitored for Dupont's Lark were flat areas dominated by low scrubs or shrubs, such as thyme (*Thymus* spp.), broom (*Genista* spp.) and lavender (*Lavandula* spp.).

Acoustic recording

We used an ARU consisting of a USB Voice Recorder SK-001 with an AC1517D72772-C processor and one integrated omnidirectional microphone. ARUs were built after a cost-effectiveness comparison of five

different recording devices (Pérez-Granados *et al.* 2019). Recorders were powered by a 12 V/8.0 mAh battery (> 300 h-autonomy), and were also connected to a digital timer that allowed us to programme the ARUs to record at selected times and days. Recordings always used a sample rate of 44.1 kHz and 16 bits in stereo mode, and were stored on microSD memory cards capable of storing 520 h of continuous data. Equipment was protected in easily portable and waterproof boxes (171 × 100 × 100 mm) made cryptic by painting them greenish-brown with spots (Pérez-Granados *et al.* 2019). These ARUs have previously shown reliability and effectiveness for bird song recording in similar conditions (Pérez-Granados *et al.* 2018a,c).

Microphones were located in an upward position and horizontally separated by at least 50 cm from natural vegetation in order to minimize any sound blocking (see Supporting Information Appendix: Fig. S2). To reduce variation in vocal activity, we limited recording analyses to time periods when singing activity was expected to be highest for each studied species (Oppel *et al.* 2014). In the case of the European Bee-eater, an ARU was located on each apiary from 15 July to 15 September 2017, a period of maximum presence of the species around apiaries in the study area (Bota *et al.* 2018). ARUs were programmed to record during the daytime, when individuals were calling while foraging or migrating in the vicinity of the beehives. Each recording was limited to 30 min and was taken every 2 h from 09:15 to 19:15 h. Although ARUs were active during two consecutive months, we analysed a subsample of recordings from which we gathered information about European Bee-eater abundance during the recording schedule (see below). Dupont's Lark recordings were carried out by placing one ARU per site. ARUs were programmed to record continuously for 30 min starting 1 h before sunrise, which is the highest singing activity period for the species. This allowed sufficient time to detect the presence of the species in 95% of cases (Pérez-Granados *et al.* 2018c). Each site was monitored for 1 day between 10 April and 15 May 2017.

Acoustic data analyses

Recordings were automatically scanned using SONG SCOPE 4.1.5 (Wildlife Acoustics 2011), which creates a target signal from the feature characteristics of the example songs used for training. This target signal can then be used as a recognizer file for comparison to determine when a sound within a recording matches these characteristics (Wildlife Acoustics 2011). We built customized species-specific recognizers for each studied species after training by setting parameters (e.g. adjusting sample rate, frequency range, minimum frequency) to isolate targeted signals and reduce the inclusion of

false-positives, such as background noise or other uninteresting songs incorrectly identified as a target signal. Recordings of each species were always scanned with their own species-specific recognizer and using algorithm 2.0 in SONG SCOPE. We selected results with a score > 40% and quality above 20. The results of automated song recognition were visual and/or acoustically checked by the same observer (C.P.G.) to separate false-positives from true-positives and calculate the true-positive rate, estimated as the number of true-positives found within all events detected (Knight *et al.* 2017). Therefore, posterior analyses about vocal activity rate were only based on true-positives (i.e. correct detections made by the recognizer). Recordings were not checked to find targeted songs not automatically detected by recognizers, as this might be more time-consuming than identifying songs by manually scanning spectrograms and our aim was to propose a rapid method to infer relative bird abundances.

We calculated the recall for each recognizer after randomly checking 12 European Bee-eater and nine Dupont's Lark recordings. Recall is an index that represents the proportion of species calls that were automatically detected and is a typical metric for assessing recognizer performance (Knight *et al.* 2017). Recall was determined for each recognizer by dividing the number of true-positives by the total number of calls uttered by monitored species during the recording (Knight *et al.* 2017). To estimate the total number of calls uttered by monitored species per recording, an experienced observer (C.P.G.) checked the selected recordings visually and acoustically and annotated the total number of calls of monitored species within them.

Bird data censuses

Methods for estimating bird abundances around ARUs differed among studied species according to counting method and previous tests to assess the distance at which our ARU was able to record vocalizations of the monitored species (Pérez-Granados *et al.* 2018c, 2019).

We conducted 35 visual censuses of European Bee-eaters in the apiaries coinciding with the ARU's recording schedule (mean number of censuses per apiary = 3.5). European Bee-eaters closer than 100 m to each apiary (and ARU) were counted at 10-min intervals within each 30-min recording. We chose that distance because we have estimated that it was the distance at which our ARU was able to detect the European Bee-eater calls (G. Bota unpubl. data). We used the mean number of European Bee-eaters counted every 10 min during the 30-min counting period as bird abundance because we considered that mean values are well suited for accounting for normal variations in abundance between intervals.

Dupont's Lark censuses were carried out in the 4 days after recordings in order to avoid modifying natural singing behaviour while recording. In each of the 27 monitored sites, we performed a census following the line transect method with a 500-m maximum detection band on each side of the observer, within which we assumed a probability of detection equal to 1 for singing males (Pérez-Granados & López-Iborra 2017). The distance of singing males from the observer was estimated acoustically and its location was recorded by GPS. Censuses were carried out by walking at a constant speed (1–3 km/h) on dry and windless days. Census time was from 60 min before dawn to dawn, a period during which the detection probability of the species remains broadly constant (Pérez-Granados *et al.* 2018b). According to field tests, we considered a 200-m buffer around each ARU to estimate the number of Dupont's Lark males potentially recorded at each ARU, as the probability of detecting the species beyond that distance under favourable singing conditions is always lower than 15% (Pérez-Granados *et al.* 2019). Therefore, we used the total number of Dupont's Lark males detected by surveyors within the 200-m buffer around recorders as an index of abundance.

Statistical analyses

VAR was considered the total number of songs per minute for each species (Garamszegi *et al.* 2007, Pérez-Granados *et al.* 2016). To estimate VAR, we divided the total number of true-positives automatically detected per recognizer by recording length (Oppel *et al.* 2014, Zwart *et al.* 2014). We fitted a linear and logarithmic regression for each of the studied species to estimate the most appropriate relationship between vocal activity rate and abundance (Borker *et al.* 2014). We used linear regressions because we expected that VAR would increase linearly with abundance (Farnsworth *et al.* 2004), but also tested logarithmic regressions because signal recognition software may become overloaded under large bird density scenarios (Oppel *et al.* 2014).

RESULTS

In 2017, we estimated VAR and bird abundance for 62 different recordings (35 recordings and 17.5 h of recording for the European Bee-eater and 27 recordings and 13.5 h of recording for the Dupont's Lark). Actual bird abundance extracted from bird censuses around ARUs differed greatly among studied species and sites, and ranged from zero to eight males for the Dupont's Lark and from zero to 36 individuals for the European Bee-eater.

ARUs detected the species in all cases where at least one individual was censused by human surveyors. The true-positive rate differed greatly between studied species. The true-positive rate for the European Bee-eater

was 97.34 (7049 calls in 7241 events detected) and 38.18 for the Dupont's Lark (4407 calls in 11 542 events detected). Recall rate for the European Bee-eater recognizer was 46.4% (2112 calls detected of the 4555 calls annotated in the validation dataset) and 63.0% for the Dupont's Lark (1177 calls detected of the 1868 calls annotated). We estimated a mean number of 12 and 10 min for scanning and checking 1-h recordings of European Bee-eater and Dupont's Lark, respectively.

We found a positive and significant relationship between VAR and estimated bird abundance for both monitored species. For the European Bee-eater, the linear regression (parameter estimate \pm se = 0.031 ± 0.004 , $F_{1,34} = 72.54$, $P < 0.001$, $R^2 = 0.68$) was found to be a better fitting model than the logarithmic model ($F_{1,34} = 37.31$, $P < 0.0001$, $R^2 = 0.52$), according to R^2 . For Dupont's Lark, logarithmic regression (parameter estimate \pm se = 1.876 ± 0.163 , $F_{1,26} = 132.70$, $P < 0.001$, $R^2 = 0.84$) was better fitting than the linear model ($F_{1,26} = 73.48$, $P < 0.0001$, $R^2 = 0.74$; Fig. 1).

DISCUSSION

Our study is the first to explore and to find a positive and significant relationship between VAR and estimated abundance for terrestrial bird species. The strong relationship found for monitored species suggests that VAR could be a cheap and rapid method to infer an abundance estimate relative to other sampled sites and to evaluate changes over time from recordings obtained with ARUs using omnidirectional microphones. Our study was focused on terrestrial birds, but VAR has potential wider applications and could be used, based on a previous assessment, for other acoustically active taxa.

Despite significant relationships found between VAR and estimated bird abundance, we are aware that our results are not fully conclusive in that VAR may not be useful to estimate an accurate absolute abundance for bird species, and that some variation in VAR remains unexplained. Several factors could be responsible for such variation, such as declining detection probabilities with distance of individuals to ARUs. For example, we monitored a site with four Dupont's Lark males within the effective detection radius but with a very low vocal activity rate (0.4 songs/min). However, three of the four males were located further than 160 m away from the ARU, a distance at which detectability for Dupont's Lark can decline by up to 20% (Pérez-Granados *et al.* 2019), which may have influenced our results.

As for censuses performed by human observers, VAR results depend on a large number of factors not accounted for in our study, such as breeding seasonality, number of conspecifics, mating success, weather conditions and time of year (Catchpole & Slater 2008), which

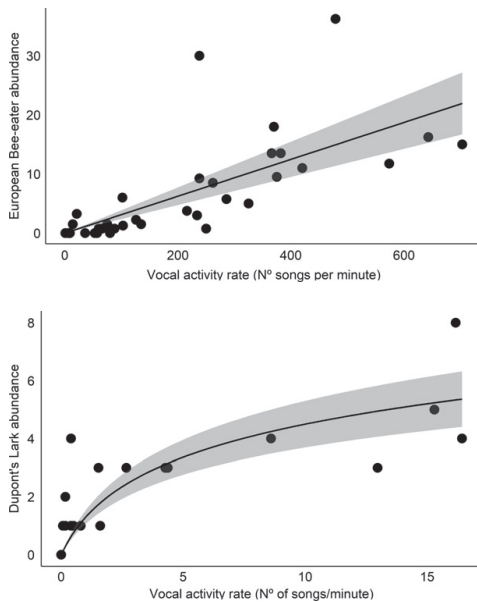


Figure 1. Relationship between vocal activity rate (number of songs per minute) index and number of European Bee-eaters (top) and Dupont's Larks (bottom). The observed values (black dots), fitted regression (black line) and 95% confidence interval (grey surface) are depicted.

may compromise the utility of VAR to infer bird abundance from sound recordings. We encourage researchers to record over several monitoring days in order to obtain an averaged VAR per site (Buxton *et al.* 2013, Oppel *et al.* 2014), which may help to control the variability in bird singing activity. The relationship between VAR and censused bird abundance can be also affected by observers' ability to detect individuals, as in traditional field censuses. Further research should focus on this topic, including imperfect detection and associated errors when estimating the relationship between VAR and estimated bird abundance.

The creation of a good recognizer is also a key aspect to approximate bird abundances from sound recordings. In this case, recognizers successfully accomplished objectives, despite variations in recall and true-positive rate between monitored species. Differences in true-positive rate have no influence on estimated relationships, as automated species identifications were verified to remove false-positives prior to analyses. However, a small true-positive rate can preclude the use of recognizers for automated species recognition at large spatial and temporal scales due to the large amount of effort and

expertise required to remove false-positives. Differences in true-positive rate between monitored species may be related to specific conditions under which recordings were taken. In the European Bee-eater, recordings were made on summer days, outside most species' breeding seasons, when general bird singing activity is low in temperate zones. Conversely, Dupont's Lark recordings were taken during dawn choruses in the breeding season. A large number of birds, including several lark species such as the Eurasian Lark *Alauda arvensis* and the Greater Short-toed Lark *Calandrella brachydactyla*, share the dawn chorus with the Dupont's Lark (C. Pérez-Granados pers. obs.), which may partly explain the low true positive rate found in this species.

Recall rate may have a great impact or even preclude the use of VAR to infer bird abundances if they are too low. Recall of recognizers is highly variable among species, but in our case they can be considered high when compared with previous studies (e.g. Digby *et al.* 2013, Shonfield *et al.* 2018, but see Potamitis *et al.* 2014 and De Oliveira *et al.* 2015). The Dupont's Lark recognizer had a higher recall rate than that for the European Bee-eater, which may partly explain the poorer relationship found between VAR and European Bee-eater abundance. In both cases, a large number of calls from long distances may not be well recognized, but plausibly fairly well detected by the researcher on the spectrogram viewer or when hearing the recordings. For the European Bee-eater, numerous calls can be made in a continuous manner for a large of group of birds. In those cases, a visual check seems to be more effective for counting calls than automated scanning due to song overlap.

Little is known about the species, study conditions and vocalizations (contact calls, territorial calls, display calls, etc.) for which the VAR may be useful to infer abundances. Previous studies (Borker *et al.* 2014, Oppel *et al.* 2014) found a strong and significant relationship for seabird species with high VAR while breeding in their colonies. However, Zwart *et al.* (2014) did not find any relationship for the European Nightjar, also a nocturnal species, but one that vocalizes during short periods while breeding, and which usually sings while flying and foraging. In agreement with those and our results, we believe that candidate species for which VAR might be useful can be identified based on their behaviour and ecology. Candidate species mainly include those living and singing or calling in groups, such as seabirds (Buxton *et al.* 2013, Oppel *et al.* 2014) or the European Bee-eater, which suggests that the VAR might also be useful for monitoring other colonial, vocally active species. VAR can also be used to infer the abundance of territorial birds, such as passerines, at least in those species whose singing behaviour does not vary with population density, such as the Dupont's Lark (Pérez-Granados *et al.* 2016). Although this study was focused on single

species, it could be adapted for multiple-species monitoring programmes by scanning the recordings using species classification algorithms. Advances in computations and automated signal recognition, including machine-learning processes, allow large datasets to be analysed in a timely manner (Stowell & Plumbley 2014).

Here, we propose a protocol to estimate the correlation function between the VAR and the apparent abundance of monitored species. Our proposed protocol is based on five steps that could be easily adapted to other species or contexts:

- 1 Estimate the effective distance at which the ARU selected is able to detect the songs of the species selected for monitoring.
- 2 Identify the period of the day at which singing or calling activity of the monitored species is at a maximum and limit the recording time to this period.
- 3 Assess the performance of the recognizer to be used after evaluating their true-positive rate and recall rate.
- 4 Estimate the VAR in a number of sites (at least 20–30) and correlate it with estimated abundance of the species within an effective distance from the ARU.
- 5 Evaluate the strength of this correlation and the cost and time required for estimating bird abundance using this method.

All of these steps should be assessed before using the VAR in any monitoring programme, but once they have been validated, the use of VAR should be a rapid method to infer bird abundance in new recordings. VAR can be useful for current monitoring programmes, but it may also be estimated retroactively to assess bird abundances and changes in population trends in archived recordings, which could be especially useful for long-term monitoring programmes aimed at analysing wildlife population trends. The recent development of open source, low-cost ARUs, together with the increase of citizen-science projects and the establishment of new robust relationships between VAR and species abundance, present a range of new opportunities for wildlife population monitoring.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Appendix: Figure S1. Sonogram of a typical European Bee-eater call (top) and Dupont's Lark (bottom) call as visualized in Song Scope.

Appendix: Figure S2. Pictures showing location of autonomous recording units used for monitoring European Bee-eater (top) and Dupont's Lark (bottom) in Spain.

Acoustic monitoring of diurnally migrating European Bee-eaters agrees with data derived from citizen science

Gerard Bota^{1,*}, Juan Traba^{2,3}, Francesc Sardà-Palomera¹,
David Giral¹ & Cristian Pérez-Granados^{2,4}



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The use of Autonomous Recording Units (ARUs) for wildlife monitoring has increased in recent years. Acoustic monitoring has been used for a wide range of research topics, but it has rarely been used for monitoring wildlife migrations. In this work we evaluate the use of acoustic monitoring to characterise the diurnal migration pattern of a bird species, the European Bee-eater *Merops apiaster*. We set up 3–4 acoustic monitoring stations daily from 11 August to 21 September 2017 in north-eastern Spain, during post-breeding migration of the species. We used the Vocal Activity Rate (VAR), defined as the number of calls per unit time, as an index of Bee-eater abundance to describe the daily and seasonal migration pattern of the species. We also assessed the relationship between daily mean VAR estimated by ARUs, with citizen science data uploaded to the platform Ornitho.cat over a large spatial scale. According to mean VAR, intensity of migration increased weekly until the last week of August when it peaked, with species abundance decreasing to the lowest values in the study area by late September. A significantly higher number of calls was detected in the first and last hours of the day. Our results agree with previously published seasonal and daily migration patterns described for the species. VAR was significantly, positively correlated with the percentage of citizen science records of Bee-eater uploaded to complete checklists, used as an independent source to compare migration timing. Overall, our results, validated through citizen science data, show that acoustic monitoring can effectively provide complementary data for monitoring the bird migration of vocally active species. The use of ARUs may help to improve our understanding of migratory behaviour and be useful for a wide range of purposes.

Key words: ARUs, automated recognition, citizen science, *Merops apiaster*, migration, Ornitho, Vocal Activity Rate

¹Landscape Dynamics and Biodiversity programme, Forest Science and Technology Center of Catalonia (CTFC), Solsona, Catalonia, Spain;

²Terrestrial Ecology Group (TEG-UAM), Department of Ecology, Universidad Autónoma de Madrid, Madrid, Spain;

³Centro de Investigación en Biodiversidad y Cambio Global, Universidad Autónoma de Madrid (CIBC-UAM), Madrid, Spain;

⁴National Institute for Science and Technology in Wetlands (INAU), Federal University of Mato Grosso (UFMT), Computational Bioacoustics Research Unit (CO.BRA), Cuiabá, Mato Grosso, Brazil;

*corresponding author (gerard.bota@ctfc.cat)

Bird migration has attracted the attention of ornithologists for centuries (e.g. White 1788, Irby 1875). Counting birds during migration offers the possibility of monitoring population sizes (Arroyo *et al.* 2016), estimating population trends (Møller *et al.* 2008) and eval-

uating changes in migratory behaviour related to different threats (De Lucas *et al.* 2004, Lindström *et al.* 2010) as well as to climate change (Saino *et al.* 2010). Techniques commonly applied in monitoring bird migration are usually costly in terms of time, human

resources and equipment. They may require long-term intensive observation sessions at many sites (e.g. Martín *et al.* 2016, Miller *et al.* 2016), trapping sessions carried out by experts to deploy monitoring devices such as geolocators or GPS tracking systems (Bridge *et al.* 2013, Sperger *et al.* 2017), several years of ringing sessions for mark-recapture studies (Bairlein 2001) or feather collection for stable isotope analysis of geographic origins (Hobson *et al.* 2015). In recent years, the analyses of bird observations submitted to citizen science platforms (e.g. eBird, Ornitho, Observation.org) by volunteer birders have shown to be a useful tool for improving our understanding of bird migration (Hurlbert & Liang 2012, Newson *et al.* 2016, Schubert *et al.* 2019). Recent initiatives such as EuroBirdPortal even allow for the observation of bird migration at the European level in near real-time thanks to the integration of data from different citizen science platforms (Gargallo 2017). These platforms have already been used to examine the timing of migration of specific bird species across a large geographic area and this wealth of data provides novel opportunities to unveil patterns of bird migration at large spatial and temporal scales (Sullivan *et al.* 2009, 2014).

Despite its enormous potential, an important part of citizen science data is subject to certain limitations derived from different sources: differences in detectability between species, different bird-identification skills among observers, unstratified data sampling (which may cause geographic biases in the spatial distribution) and non-homogeneous distribution of the birding community across regions have been described as potential biases associated with this kind of data (Ferrer *et al.* 2006, Sullivan *et al.* 2009, Johnston *et al.* 2018).

The development of non-invasive techniques for monitoring bird migration is desirable, in order to avoid capturing individuals or having to carry out intensive field work. To this end, studies based on horizontally scanning weather radars or vertical wind profilers, among other types, have been used in different regions for monitoring bird migration (Weisshaupt *et al.* 2018, Horton *et al.*, 2020). This technique can provide information on density, direction, speed and altitude of migrating birds but cannot describe the species involved (Salamon *et al.* 2016). Among the non-invasive techniques most commonly deployed for monitoring wildlife in recent years is the use of Autonomous Recording Units (ARUs hereafter; see review in Sugai *et al.* 2019). This technique is a consistent and suitable alternative to traditional field surveys to estimate parameters such as species presence, abun-

dance or richness (Oppel *et al.* 2014, Darras *et al.* 2018, Pérez-Granados *et al.* 2019a). Acoustic monitoring has already been used to study the migration of different animal groups, such as whales (Burnham & Duffus 2020), bats (Johnson *et al.* 2011) and birds. Acoustic monitoring of flight calls during the night has proven to be an effective method to provide information on migration routes, timing and relative migration intensity of birds (Larkin *et al.* 2002, Farnsworth *et al.* 2004, Farnsworth & Russell 2007, Sanders & Mennill 2014). Some authors have proposed that ARUs could be used to characterize the migration pattern of specific bird species (Salamon *et al.* 2016) and recent studies have demonstrated their potential to detect the arrival and departure of birds (Oliver *et al.* 2018, Pérez-Granados & Schuchmann 2020a). As far as we know, such methodology has never been used for monitoring the migration pattern (hours, peak of migration) of a diurnal bird.

In this paper, we evaluate the utility of acoustic monitoring based on ARUs coupled with automated signal recognition as a tool for monitoring the diurnal post-breeding migration of a vocally-active bird species, the European Bee-eater *Merops apiaster* (Bee-eater hereafter). Pérez-Granados *et al.* (2019a) have recently described a strong and significant relationship between the Vocal Activity Rate index (VAR hereafter; i.e. number of calls detected per time unit) and the number of Bee-eaters flying within a radius of 100 m around ARUs. Therefore, we used the VAR as an index of the abundance of Bee-eaters for describing the daily and seasonal pattern of migration. We also aimed to assess whether the seasonal migration pattern described by using acoustic monitoring was in accordance with the timing of migration obtained with citizen science data, in order to provide a qualitative independent comparison to our acoustic monitoring method. We expected that migration timing obtained thanks to citizen science data would be correlated with that estimated using ARUs.

METHODS

Study species

The European Bee-eater is an insectivorous long-distance migrant bird that breeds in southern Europe, North Africa and western Asia and winters in sub-Saharan Africa (del Hoyo *et al.* 2001). Western European breeders migrate during the day using a broad front on a western route mainly to non-breeding regions in West Africa (Hahn *et al.* 2020). Median



A male European Bee-eater presenting a honey bee to his mate (photo Roger Guillen, 29 May 2018, Bellmunt-Almenara, Catalonia).

migration speed during post-breeding migration is around 159 to 206 km/day until the first sub-Saharan site, depending on the breeding origin (Hahn *et al.* 2020). Groups are generally stable during migration (Dhanjal-Adams *et al.* 2018) and this species is known to use both flapping and soaring-gliding flight while migrating (Sapir *et al.* 2011).

The Bee-eater is a richly coloured bird and is a very vocal species that usually calls while foraging or migrating in flocks (Cramp & Simmons 1985), making it a highly detectable and easily identifiable species for the bird-watching community. The contact call, uttered at low-frequencies (1–3 kHz; Figure S1), is the most frequent vocalisation of the species and it is a repeated, soft but abrupt rolling “prüt”, given in chorus from flying flocks and reaching long distances (Mullarney *et al.* 1999). Species detectability is a key point in bird monitoring techniques (Sanz-Pérez *et al.* 2020), and easily detected birds are reported more frequently as well as having less bias than cryptic species in citizen science platforms (Sullivan *et al.* 2009). We selected the Bee-eater as a study species due to its high detectability, easily identifiable characteristics (factors described as potential biases in citizen science data),

because it is vocal while migrating and because of previous studies testing the use of the VAR of the species as an index of Bee-eater abundance (Pérez-Granados *et al.* 2019a).

Study area

The study area was comprised of a network of nine acoustic monitoring stations placed in a semi-arid, relatively flat agricultural landscape on the eastern edge of the Ebro Valley (41°46'N, 0°46'E) in western Catalonia (Lleida province, NE Spain). Monitoring stations were located within an approximate area of 1000 km² and were separated by 11.1 ± 1.1 km (mean \pm SD).

The study period was from 11 August to 21 September 2017. We consider this period long enough to assess the utility of ARUs for monitoring bird migration and detecting changes in Bee-eater abundance as it matches the main period of the post-breeding migration described for the species in the study area (Muntaner *et al.* 1983). Acoustic monitoring stations were placed at apiaries (Figure 1), since Bee-eaters usually use beehives as feeding stopovers during migration (Yosef 2004). Previous studies also used apiaries as monitoring stations to study the migration strategy of

the species (Yosef *et al.* 2006). All selected apiaries were composed of 30 to 50 beehives which remained active throughout the study period.

Collection and analysis of acoustic data

During the study period, three to four acoustic monitoring stations were active daily. We placed one ARU per active station, which was systematically and fortnightly shifted between the nine selected apiaries. Each ARU consisted of a USB Voice Recorder SK-001 with processor AC1517D72772-C and one integrated, single-channel microphone (Pérez-Granados *et al.* 2019b). ARUs were powered by a 12V/8 mAh battery (300-h autonomy), and were started and stopped by an electronic timer. Equipment was protected in easily portable, weatherproof plastic boxes (60×80×160 mm), camouflaged by painting them greenish-brown. ARUs were located on the ground with the microphone in an upward position with no blocking barriers within at least 50 cm (Rempel *et al.* 2013; see Figure 1). ARUs were programmed to record continuously for 30 minutes at the following local (GMT+2) recording times: 9:15, 11:15, 13:15, 15:15, 17:15 and 19:15. We used a sample rate of 44.1 kHz and 16 bits for all recordings. We estimated that the recorder was able to detect the Bee-eater calls up to 100 m (Pérez-Granados *et al.* 2019a). Recordings were collected in mp3 format

on microSD flash cards (100-h memory) and needed a post-transformation to wav format prior to analysis. Although compression into mp3 format affects the spectral and temporal composition of the signal (Obrist *et al.* 2010), and its conversion to wav format does not improve recording quality, this should not influence the signal recognition results (Rempel *et al.* 2005).

Recordings were analysed automatically using the freely available software Song Scope v. 4.1.5 (Wildlife Acoustics 2011). This software has proven to be one of the most efficient in automated song recognition (Knight *et al.* 2017). Song Scope is able to create a target signal from the characteristics of the example signals used for training, which is used as a recognizer file to compare when a sound within a recording matches its characteristics (Waddle *et al.* 2009). The automated signal recognition software and species-specific recognizer were the same as those in Pérez-Granados *et al.* (2019a), when they assessed the significant relationship between VAR and flying Bee-eaters around recorders. Specifically, the recall of the recognizer, estimated as the proportion of Bee-eater calls detected by the recognizer divided by the total number of Bee-eater calls on sound recordings, was 46.4%. The recall value is highly variable among species and software employed and ours can be considered acceptable when compared with previous studies (see discussion



Figure 1. One of the locations of beehives with an autonomous recording unit (on the ground in the foreground) used for monitoring European Bee-eater migration in North-eastern Spain (photo Joan Rodriguez, 29 June 2017).

about the recall rate of the recognizer employed in Pérez-Granados *et al.* 2019a). Moreover, the recognizer built for Pérez-Granados *et al.* (2019a) was trained using Bee-eater calls recorded in the study area and using the same ARU, so the relationship found in that previous study should be valid for this one. In order to select recordings with different background noise and Bee-eater abundance, we randomly selected 120 30-min recordings (16% of total sample) in which all events reported by the recognizer were visual and/or acoustically checked, always by the same observer. To evaluate automatic signal recognition, we calculated the precision of the recognizer as the proportion of true Bee-eater calls detected by the recognizer divided by the total number of sounds classified as Bee-eater calls by Song Scope (Knight *et al.* 2017).

Bird citizen science data

We used the online citizen science platform Ornitho.cat (hosted by the Catalan Institute of Ornithology, available at www.ornitho.cat) as a source of quantitative independent data on migration timing of the Bee-eater during the study period and area. We used this platform because Ornitho.cat was the citizen science platform with the highest number of Bee-eater observations during the study period in the area and as it is also the most used platform by local bird-watchers (5629 registered users on 17 December 2019).

Citizen-science data quality is, among others factors, influenced by the number of observations (La Sorte *et al.* 2014, Sullivan *et al.* 2014). In order to maximize the number of observations available for analysis and to cover the entire study period, and given that Bee-eater migration follows a broad front in the region, we conducted the analyses using all uploaded complete checklists (all species observed reported) within the region of Catalonia (31,895 km²) rather than those uploaded from the study area only (52 complete checklists were available for the study area with no published lists for some of the monitored weeks). We extracted 519 complete checklists with presence of Bee-eater from 11 August to 21 September 2017 in Catalonia (consulted on 10 October 2019). Due to the daily variation in the number of active birders and considering that it may bias the number of Bee-eater observations uploaded, we also extracted all complete checklists per day ($n = 1535$) as a measure of daily birding effort in the region.

Statistical analyses

In order to elucidate whether migration patterns of Bee-eaters depend on the time of day and vary season-

ally, we fitted a Generalized Linear Mixed Model (GLMM) with Gaussian distribution error and an identity-link function, using VAR of each recording as response variable and recording time (six categorical levels) and migration week (six categorical levels) as fixed effects. Recording day and acoustic monitoring station were included as random effects. When a fixed effect was found to be significant, a Tukey's post hoc test was performed to assess whether there were differences among levels. We also estimated the Spearman rank correlation between mean VAR of all acoustic stations per day (hereafter mean VAR per day) and corresponding percentage of complete checklists with presence of Bee-eaters in relation to all complete checklists uploaded for the same day. This variable has already been used to define bird migration timing patterns using citizen science data (Sullivan *et al.* 2009).

All statistical analyses were performed with R v. 3.4.1 (R Core Team 2016) or Statistica v. 10 (Statsoft 2011). We used the packages 'lme4' (Bates *et al.* 2015) for the GLMMs, 'lmerTest' (Kuznetsova *et al.* 2017) to calculate the significance of fixed effects and 'multcomp' (Hothorn *et al.* 2008) for post hoc comparison tests.

RESULTS

We collected 763 30-min recordings (381.5 h of recording) and a total of 223,479 Bee-eater calls were automatically detected. The average number of 30-min recordings per week across stations was 127 ± 35 (\pm SD). The precision of the recognizer was 99.2% (23,537 Bee-eater calls in 23,719 calls automatically detected). We therefore decided to consider all events identified by Song Scope as Bee-eater calls.

VAR differed with week and recording time (Table 1). A smaller VAR was detected during the first two weeks (11 to 24 August) and the last week of the study period (15 to 21 September), when compared to the central weeks (Table 1, Figure 2). Significantly more Bee-eater calls were detected during the late afternoon (19:15) in relation to the rest of the recording times (Figure 3). The first hours of the day (9:15) and afternoon (17:15) also presented a significantly higher number of calls than midday hours (Figure 3).

Mean VAR per day was significantly and positively correlated with the percentage of complete checklists with presence of Bee-eater ($r_s = 0.65$, $P < 0.001$; Figure 4). Weekly patterns of mean percentage of complete checklists including Bee-eaters uploaded to

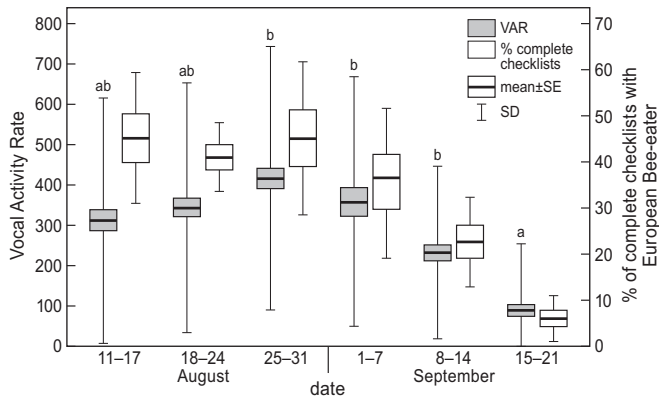


Figure 2. Weekly migration pattern of the European Bee-eater in the study area during the study period. Boxplots showing mean, SE and SD of Vocal Activity Rate (VAR) per week (grey boxes) for all active acoustic monitoring stations and percentage of complete checklists including European Bee-eaters uploaded to Ornitho.cat (empty boxes). Letters on the top show significant differences in hourly VAR between migration weeks from Tukey's post hoc test.

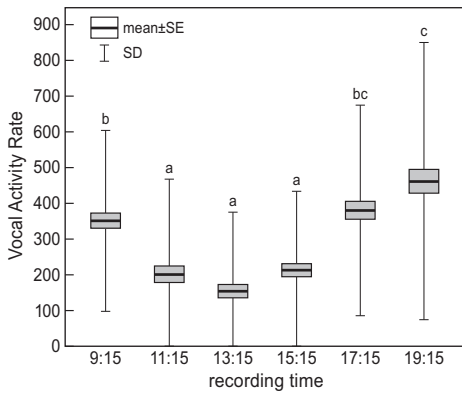


Figure 3. Daily migration pattern for European Bee-eaters in the study area during the study period. Boxplots showing mean, SE and SD of Vocal Activity Rate (VAR) detected during each recording time period. Letters on the top indicate significant differences between recording times from Tukey's post hoc test.

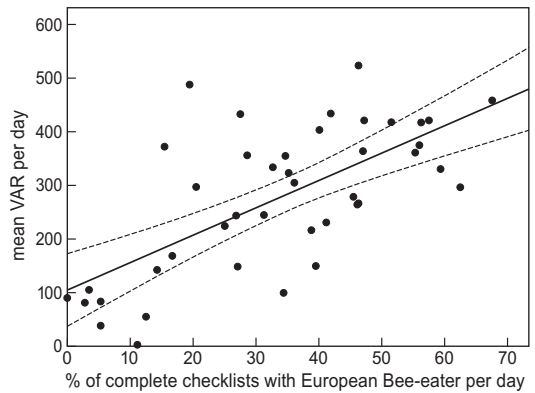


Figure 4. Relationship between mean Vocal Activity Rate (VAR) per day and percentage of complete checklists with presence of European Bee-eaters per day ($r_s = 0.65$, $P < 0.001$). Linear regression line is shown in black, and 95% Confidence Intervals with dashed lines.

Ornitho.cat followed a similar pattern to the one described by ARUs (Figure 2). However, the highest peak of VAR observed in the last week of August was not so clear in the migration pattern obtained using citizen science data.

DISCUSSION

Our study is the first to use acoustic monitoring to provide accurate information about post-breeding migration timing for a diurnal bird species. The maximum number of calls were detected during the

last week of August and afterwards decreased every week until the third week of September, when very low levels were recorded. Our findings on the seasonal pattern of European Bee-eater migration found through the use of ARUs agree with the one described by Muntaner *et al.* (1983), who also identified the last week of August as the one with the largest number of Bee-eaters migrating over Catalonia. The highest migration peak observed in the last week of August by acoustic monitoring was not so clearly observed in the pattern obtained using citizen science data. Previous studies have stated that Bee-eaters increased their presence in apiaries during and after rain (Glaiim 2014,

Table 1. Summary table of the results of a Generalized Linear Mixed Model (GLMM) testing the relationship between Vocal Activity Rate of European Bee-eaters (number of calls detected per recording) and the time of the day (Hour) and time of the season (Week). Recording day (Day) and acoustic monitoring station (AMS) were considered as random effects. Total number of recordings: 763.

<i>Fixed effects</i>					
Variable	Sum. Sq.	Sq. Mean	F-value	df	P (>F)
Hour	9,329,214	1,865,843	34.316	5	<0.001
Week	1,186,129	237,226	4.363	5	0.002
<i>Random effects</i>					
Group name	Variance	Std. Dev			
Day (Intercept; n = 42)	1686	41.06			
AMS (Intercept; n = 9)	17154	130.97			
Residual	54373	233.18			

Moreno-Opo *et al.* 2018). During the last week of August there were three different rainy days in the study area (Meteocat 2017a). The third week of August accounted for only one rainy day and the first week of September two days (Meteocat 2017a, b). These rainy episodes during migration peak may partly explain the larger presence of Bee-eaters detected by acoustic monitoring around apiaries.

Acoustic monitoring provides robust and continuous information about the daily migration pattern and use of apiaries as stopovers by European Bee-eaters. Daily migration was concentrated primarily in the late hours of the day and secondarily in the early hours. This result is in accordance with those obtained by Moreno-Opo *et al.* (2018) in western Spain, who found less Bee-eater interactions at apiaries during the middle of the day. Prior studies that focused on the post-breeding migration of the Bee-eater have also found a larger number of birds crossing the Strait of Gibraltar during the first and the last hours of the day (López-Gordo 1975, Tellería 1979). The Bee-eater flight mode is based on flapping but also on soaring-gliding and is highly influenced by atmosphere conditions (Sapir *et al.* 2010, 2011). The species usually flies higher than 500 m above ground level when using thermals during midday (Yosef *et al.* 2006). In such cases, birds would not be detected by the recorder which could partly contribute to the lower number of calls detected during the central hours of the day.

According to our prediction, mean VAR per day, as an indicator of bird abundance, was positively corre-

lated with the percentage of complete checklists with presence of Bee-eater coming from a citizen science platform, an independent way to estimate migration timing. Despite differences in spatial scale covered by both methods, our results are in agreement with a previous study that also found positive correlations between data from acoustic monitoring and data from more traditional migration study techniques (Sanders & Mennill 2014). Therefore, we believe that acoustic monitoring data can be a useful tool for monitoring diurnal migration of the European Bee-eater. These results also opened up new perspectives on the use of ARUs for monitoring other diurnal, vocally-active migrating species. Our results are in agreement with prior studies carried out on nocturnal migrants that have proven the functionality of using acoustic monitoring for measuring timing and relative bird migration intensity (Larkin *et al.* 2002, Farnsworth & Russell 2007, Sanders & Mennill 2014, Salamon *et al.* 2016).

Acoustic monitoring could be used to assess the timing of migration or the impact of local weather conditions on bird migration (Sapir *et al.* 2011, La Sorte *et al.* 2014). This methodology might also be a good cost-benefit alternative for monitoring bird migration in inhospitable or difficult to access, remote areas, such as high mountain passes where bird migration can be bottle-necked in relatively small areas (Komenda-Zehnder *et al.* 2010, Williams *et al.* 2011). In addition, acoustic monitoring allows for longer and standardised daily time series of data, probably at a lower cost than human visual counts.

The European Bee-eater is a common and highly vocally active bird species that uses beehives intensively while migrating (Yosef *et al.* 2006), which enabled us to collect a large amount of data with relatively little effort. We also might expect better performance of passive monitoring acoustic in species like the Bee-eater that concentrate together during migration and perform flocking behaviour. Future studies aiming to monitor other bird species may require a basic knowledge of the migratory strategy of the study species (e.g. preferred habitats, priority migratory pathways and timing) to design monitoring protocols able to collect a satisfactory amount of data for detecting daily and seasonal changes. Furthermore, easy-to-identify species, such as European Bee-eaters, are reported more frequently on citizen science platforms (Sullivan *et al.* 2009); while lower temporal correlations between VAR and observations uploaded to platforms might be expected for other, less conspicuous, bird species.

The recent development of low-cost ARUs, machine learning processes for detecting bird vocalizations and

the possibility of remotely transmitting data (Beason *et al.* 2018, Hill *et al.* 2018, Sethi *et al.* 2018), open the doors for implementing networks of recorders at a relatively low cost. Furthermore, with the rapid development of automated signal recognition software (Knight & Bayne 2018, Stowell *et al.* 2018), a set of sound recordings coming from acoustic monitoring could be used for describing migration timing and migration intensity for a different number of species at a single time (Pérez-Granados & Schuchmann 2020b). In conclusion, we believe that acoustic monitoring of diurnal flight calls of migrating birds can be an effective alternative or complementary technique for monitoring migration of vocally-active species, such as different species of cranes, finches, swallows, swifts or wagtails, among other bird groups.

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SAMENVATTING

In de afgelopen jaren is het gebruik van automatisch werkende opnameapparatuur om dieren in het wild te monitoren sterk toegenomen. Akoestische monitoring wordt voor de beantwoording van veel onderzoeksvragen gebruikt, maar zelden voor het monitoren van de trekintensiteit van vogels. Wij hebben gebruikmakend van 3–4 meetstations met behulp van deze techniek het dagelijkse trekpatroon van Bijeneters *Merops apiaster* onderzocht. Het onderzoek vond plaats tussen 11 augustus en 21 september 2017 (dus na het broedseizoen van de vogels) in het noordoosten van Spanje. Het aantal roepjes per tijdseenheid werd gebruikt als maat voor de treksterkte. De resultaten werden vergeleken met gegevens van waarnemers afkomstig uit een groot gebied, die waren geüpload naar het platform Ornitho.cat. (citizen science project). Volgens de door ons verkregen gegevens nam de intensiteit van de trek vanaf het begin tot de laatste week van augustus toe, waarna de intensiteit afnam en eind september het laagste niveau bereikte. Er werd in het eerste en laatste uur van de dag een significant groter aantal roepjes gedetecteerd dan gedurende de rest van de dag. Onze resultaten komen overeen met eerder gepubliceerde trekpatronen van de soort en vertonen een positieve correlatie met de resultaten van het citizen science project. Het onderzoek laat zien dat automatische registratie van geluiden een effectief alternatief of complementair kan zijn om de trek van vogelssoorten te monitoren die vocaal actief zijn.

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