



Original investigation

Evaluation of methods to monitor wild mammals on Mediterranean farmland

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ABSTRACT

Managers have, over the years, searched methods with which to monitor wildlife populations that will have the best cost-performance balance for each case scenario. Monitoring techniques are of particular importance when attempting to attain either population density estimates or species richness values, since they enable management decisions to be made. It is, therefore, imperative to assess the capability of the methods employed to detect a wide range of species as a means to evaluate their cost-efficiency. Seventeen mammal species on Mediterranean farmland were sampled using three monitoring methods: camera trapping, spotlight counts and indirect indices based on track and faeces counts. The method able to detect the greatest number of species with the best cost-performance balance was the indirect index. There were no significant differences among seasons when applying the same sampling methodology. Studies analysing methods with which to monitor wildlife populations are in great demand in Europe, where attempts are gradually being made to standardise protocols, reduce field effort and costs, and maximise data potential and data gathering over the years. Our manuscript contributes towards finding a cost-effective method and a standardised field protocol to be applied in Mediterranean habitats.

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Introduction

All wildlife management strategies require knowledge and monitoring of the population size in order to assess the effects of management actions on the population dynamics (Yoccoz et al., 2001), including game management (Delibes-Mateos et al. 2008), define conservation criteria for threatened species (Garrote et al. 2011) and/or understand the epidemiology of relevant pathogens (Acevedo et al. 2007, Gortázar et al. 2006). It is not usually feasible to perform a complete census of a wildlife population, and managers (in a broad sense) rely instead on estimations of population abundance (e.g. Engeman et al. 2013). Monitoring population abundance is, therefore, mandatory for every management plan in order to establish comparisons among populations and/or assess the dynamics of the population over the years (Battersby and Greenwood 2003, Valente et al. 2014). It should, however, be borne

in mind that the management actions applied to a given species also have effects on other species, and the design of population monitoring programs should, therefore, ideally be sufficiently comprehensive to obtain information for all species potentially related to the target one (Pollock et al. 2002).

The choice of the method should be made by bearing certain significant aspects in mind, such as the logistical and financial resources available, the ecology of the studied species, the periodicity of the sampling and the management questions that need to be addressed (Mayle et al. 1999, Pollock et al. 2002, Valente et al. 2014, Wilson and Delahay 2001). The methods used to estimate population abundance can be broadly classified as direct and indirect. When using direct methods, the observer has direct contact with the animal being monitored (Focardi et al. 2002, Lyra-Jorge et al. 2008), while indirect methods are based on presence signs such as foot tracks and, more commonly, pellet groups (Lyra-Jorge et al. 2008, Valente et al. 2014, Virgós and Casanovas 1999). Direct methods, such as spotlight counts, have the main advantage of providing information about the sex-ratio, age structure and behaviour of the population being studied (Mayle et al. 1999). However, indirect methods are preferred when working in forested areas, with elusive species, or with populations at low densities (Marques et al.

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2001, Tellería 1986). One of these two types of methods is camera trapping, which has the advantages of both classes of methods – we can see the animals but this does not require the presence of an observer (Garrote et al. 2011, Rovero and Marshall 2009).

The methods most commonly used to monitor wildlife species in general, and wild mammals in particular, are (e.g. www.aphaea.eu/cards/species/): spotlight counts, indirect indices and camera trapping. The selection of the best methodology with which to monitor wildlife on extensive Mediterranean farmland is not straightforward, although farmland represents an important percentage of the world's surface (Foley et al. 2005, Ramankutty et al. 2008) and constitutes one of the habitats with the greatest biodiversity in Europe (Altieri and Nicholls et al., 1999, Krebs et al. 1999), thus making it essential to correctly define and standardise population monitoring protocols. Despite the fact that most of the methods have been comparatively assessed for target species (Rovero and Marshall 2009, Wilson and Delahay 2001), it is now necessary to evaluate the capacity of each method to detect the whole community of species in order to evaluate their cost-performance balance. Past studies, such as Acevedo et al. (2008) and Putman et al. (2011)

dealt with ungulate populations reviewing and comparing different methods taking into account several aspects of the methods, as costs, difficulty, suitability for low/high densities, among others, which can be very useful when designing a monitoring project. However, no studies were carried out on a broad set of species and on farmlands. In this context, the aim of this study was, therefore, to evaluate simultaneously the three methods most frequently used to monitor wildlife population abundance (track and faeces sampling, spotlight counts and camera trapping) in a Mediterranean farmland habitat, in order to assess their performance as regards detecting the whole wild mammal community and to analyse the variations in species detectability throughout three different seasons: winter, spring and summer.

Material and Methods

Study Area

The study area (UTM 10x10 km - 30TWN91) is located in Navarra, a region in the north east of the Iberian Peninsula (Fig. 1).

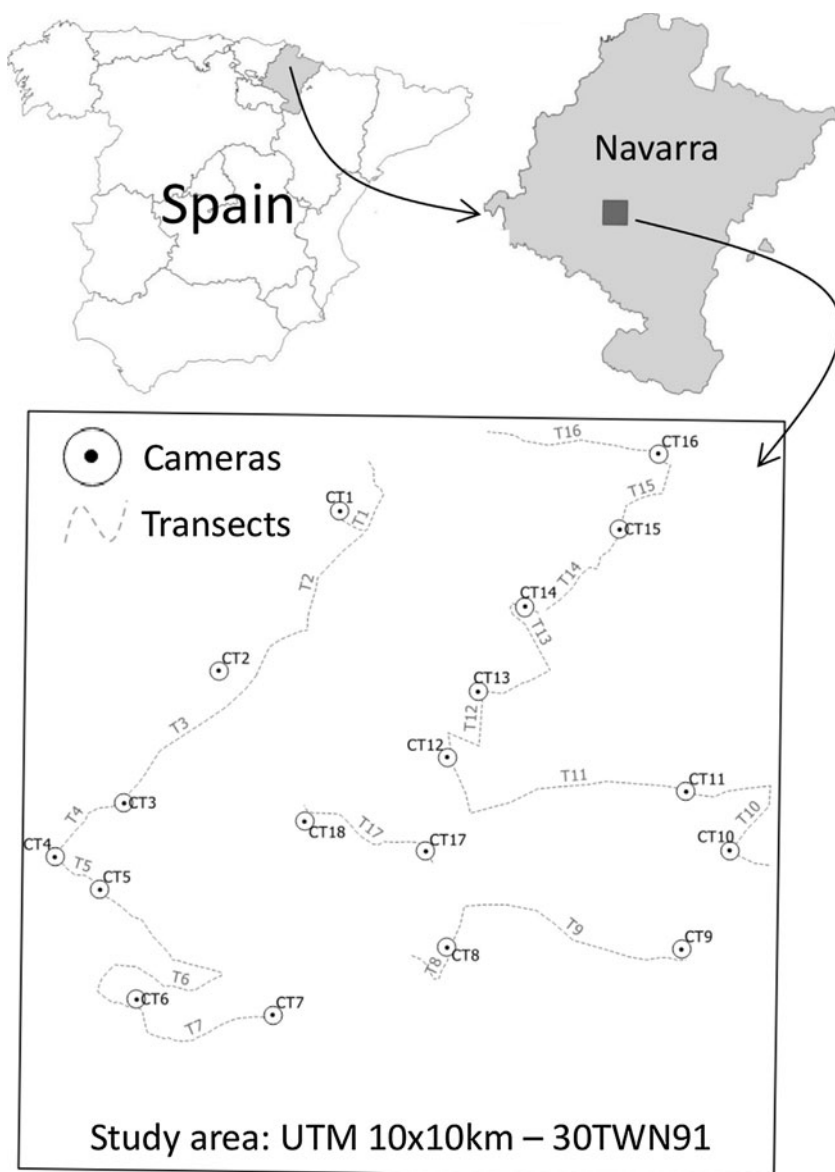


Fig. 1. Location of the study area in the Iberian Peninsula and in the Navarra region. The locations of the camera traps (coded from CT1 to CT17) and the transects (coded from T1 to T17) are also shown.

The climate is Meso-Mediterranean with a mean annual temperature of 12 °C and a mean precipitation of 400–500 mm. Natural vegetation is represented by a small riparian gallery forest dominated by *Fraxinus* spp., *Populus alba*, *Rubus* spp., *Rosa* spp., *Crataegus monogyna* and small patches of *Quercus ilex*, *Quercus coccifera*, *Juniperus oxycedrus* and *Rosmarinus officinalis* dispersed in an agricultural matrix comprising of cereal crops, vineyards and olive trees.

The medium-size mammal community in the study area is composed of 17 species: the Iberian hare (*Lepus granatensis*), the European rabbit (*Oryctolagus cuniculus*), roe deer (*Capreolus capreolus*), wild boar (*Sus scrofa*), the beech marten (*Martes foina*), the least weasel (*Mustela nivalis*), the European polecat (*Mustela putorius*), the European mink (*Mustela lutreola*), the European otter (*Lutra lutra*), the European badger (*Meles meles*), the common genet (*Genetta genetta*), the wildcat (*Felis silvestris*), the red fox (*Vulpes vulpes*), the European beaver (*Castor fiber*), the European hedgehog (*Erinaceus europaeus*) (de Navarra, 2013, Palomo et al. 2007), and two domestic animals – the domestic cat (*Felis domesticus*) and dog (*Canis lupus familiaris*).

Sampling Design

Fieldwork was compliant with the cereal cycle. Sampling was conducted in January (2014), just after the cereal was sown (which comprised the winter sampling), in May (2014), just before the harvest (which comprised the spring sampling) and in September (2014), after the harvest and before sowing (which comprised the summer sampling). The sampling was carried out using indirect index (tracks and faeces counting), spotlight counts and camera trapping. The detectability was assumed to be constant to record presence/absence data, and the same transects/camera locations were repeated between seasons to avoid detectability-related bias in the estimates.

Spotlight counts

The spotlight counts were conducted in a vehicle equipped with two moveable spotlights which drove at a constant speed (20–30 km/h). All the transects ($n = 17$; see Fig. 1) were performed by the same three observers, 2 trained biologists using the spotlights (H. Bintanel and D. Villanúa) and a naturalist like driver. Seventeen consecutive tracks (mean length: 2.2 km) were designed in order to obtain a total sampling effort of 37.6 km in the sampling area. The counts were carried out in each of the seasons studied (winter, spring and summer) and the performance of the transects started one hour after sunset. The total effort for sampling with spotlight counts was 24 hours (4 hours for each of the two technicians in three seasons).

Indirect index: track and faeces sampling

The same 17 transects that were designed for the spotlight counts were used to perform the indirect methodology through the use of track and faeces sampling. The characteristics of the habitats provided similar good visibility in a 2 m-wide strip. The transects were performed on foot by the same observers and all faeces or track contacts were registered and georeferenced. If the species could not clearly be identified using one particular presence sign, it was photographed to allow a later classification, as suggested by Blanco (1998) and Olsen (2013). The total effort for sampling with indirect index was 48 hours (4 field trips of 4 hours each in the three seasons).

Camera trapping

Eighteen photo trapping cameras (Leafriver IR-5 Infrared) were left in the field for 7 consecutive nights in each sampling season (winter, spring and summer), resulting in a total effort of

378 cameras-days. The cameras were generally located along the transects designed for the other monitoring methods and along natural mammals' travel routes (see Fig. 1). Blind set cameras were installed 2–3 m perpendicular to the travel route in order to obtain a broadside photograph of the animal as it moved by. Cameras were deployed on the tree nearest to the computer-generated point, 30–50 cm above the ground level, with the angle of view parallel to the overall slope and unobstructed by vegetation. The total effort for sampling with camera trapping was 84 hours (36 h to leave the equipment, 36 h to remove it and 12 h to analyse the images).

Statistical Analysis

Data were analysed at the level of season and method, and a total of 9 samples (3 methods and 3 seasons combined analyses) were obtained. The species richness was obtained and analysed in R (R Core Team, 2014, www.r-project.org, accessed 20 Mar 2017) through the use of the “vegan” package (Oksanen et al. 2013). Mean species richness and standard deviation were compared at transect/camera level by means of t-student tests among seasons and methods in order to search for significant differences among them (p -value < 0.05). A total of 15 t-student tests were performed, all of them paired to compare the richness values and standard deviation between methods (in the three seasons – 9 comparisons) and within methods (the same method in different seasons – 6 comparisons) using the means for the cameras or transects (17 and 18 respectively) achieved with a certain monitoring method in a selected season. Species accumulation curves (i.e. a plot showing the cumulative number of species recorded in a particular area as a function of the sampling effort) were built for each of the 9 samples. Species accumulation curves were built using two different approaches: 1) applying the “exact” method followed by the “lomolino” model, which extracts the exact species richness for each site (i.e. camera for camera trapping or transect for both spotlights counts and indirect index), and 2) applying the “random” method followed by the “Arrhenius” model in order to perform permutations (100 permutations in each case). The last approach was carried out to increase data potential and to ensure that bias owing to sampling design would not lead us to attain the wrong insights from the results obtained when using only the “exact” method. However, it is worth noting that the accumulation curves (obtained with either one of the methods) represent the average number of species under all possible permutations (Ugland et al. 2003). The asymptotes values calculated – i.e. the threshold from which an increment of the sampling effort does not add much more information to the survey – can only be achieved when using the “exact” method, i.e., the software only gives this value when applying this type of analysis. We also estimated the Shannon diversity index – based on the number of species present and their abundance – which is used to measure and characterise species diversity in a community.

Results

The results showed few visible differences (between methods and seasons) when applying the “exact” method (Fig. 2) and the “random” method. The success regarding observation of animals/signs of presence for each method is shown in Table 1, where we can see highest percentages of success for the indirect index method. The mean Shannon index values (along with the confidence intervals at 95%, standard deviation values and asymptote values) are shown in Table 2, with an asymptote peak of ~32 species for the indirect index method in winter.

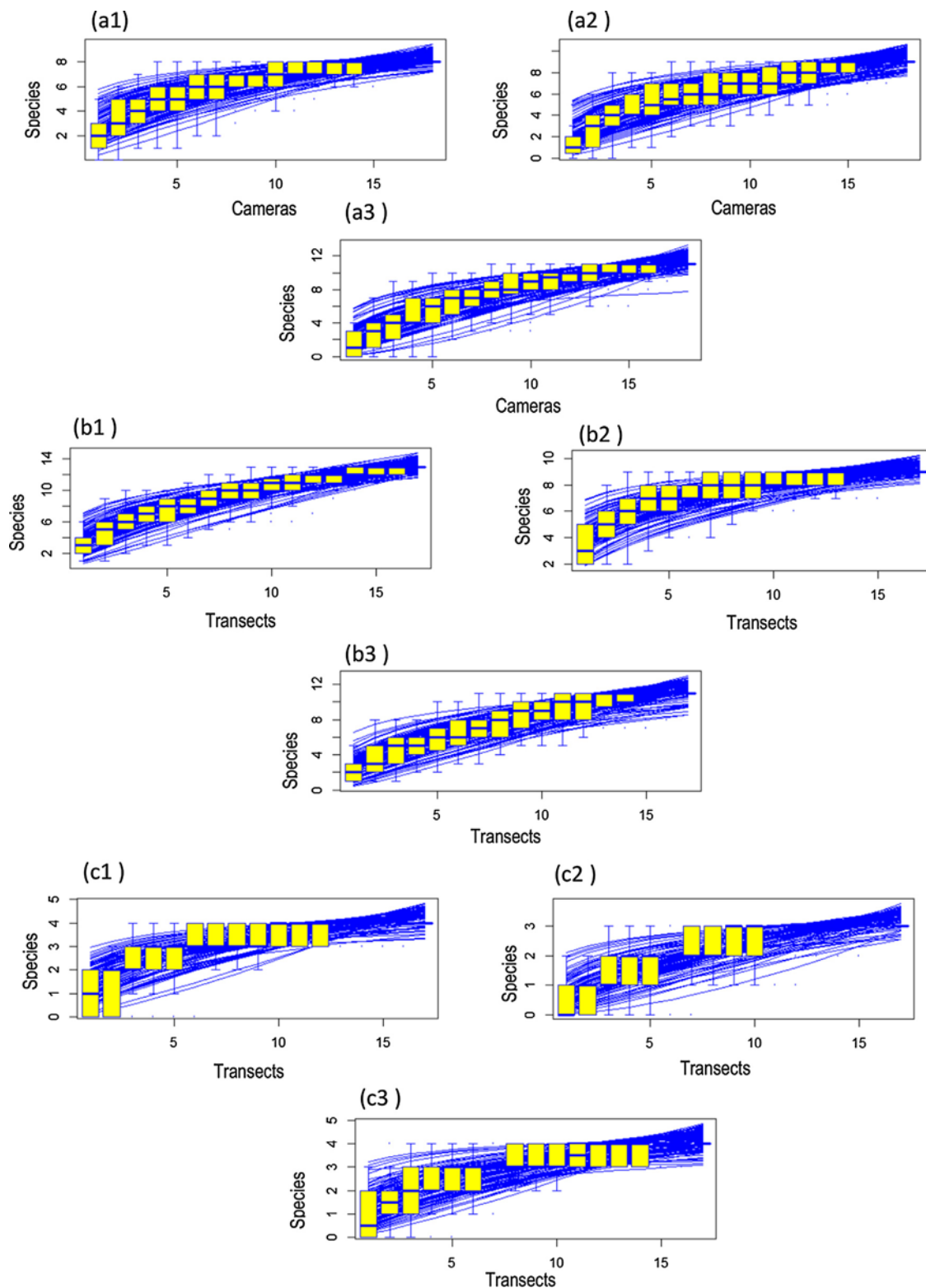


Fig. 2. Species accumulation curves using the “exact” method with the boxplots with confidence intervals, showing the increase in species richness throughout the sampling effort. (a1) Camera trapping in winter; (a2) Camera trapping in spring; (a3) Camera trapping in summer; (b1) Indirect index in winter; (b2) Indirect index in spring; (b3) Indirect index in summer; (c1) Spotlight count in winter; (c2) Spotlight counts in spring; (c3) Spotlight counts in summer.

Variation among Methods

There were no significant differences (p -value > 0.05) when comparing camera trapping with the indirect index for all three seasons analysed (winter, spring and summer with 0.053, 0.122 and 0.978

p -values respectively). However, there were significant differences (p -value < 0.05) among the three seasons when comparing camera trapping and spotlight counts (for winter, spring and summer 2.1E-07, 1.1E-08 and 3.9E-07 p -values respectively) and when comparing indirect index and spotlight counts (for winter, spring and

Table 1
Description of success as regards observation of animals/signs of presence for each method (Cam – Camera trapping; II – Indirect index; SC – Spotlight counts) and for each season sampled (winter, spring and summer). Number of observations (presence/absence) and percentage of success in relation to the number of cameras present in the field or the number of transects performed.

Species	Cam w/ observations (% of success)	Cam w/ observations in Winter (% of success)	Cam w/ observations in Spring (% of success)	Cam w/ observations in Summer (% of success)	II w/ observations (% of success)	II w/ observations in Winter (% of success)	II w/ observations in Spring (% of success)	II w/ observations in Summer (% of success)	SC w/ observations (% of success)	SC w/ observations in Winter (% of success)	SC w/ observations in Spring (% of success)	SC w/ observations in Summer (% of success)
<i>Lepus granatensis</i>	7 (12.9)	3 (16.7)	2 (11.1)	1 (5.6)	8 (15.7)	1 (5.9)	4 (23.5)	3 (17.6)	9 (17.6)	5 (29.4)	2 (11.8)	2 (11.8)
<i>Oryctolagus cuniculus</i>	8 (14.8)	2 (11.1)	2 (11.1)	3 (16.7)	19 (37.3)	5 (29.4)	7 (41.2)	7 (41.2)	15 (29.4)	6 (35.3)	4 (23.5)	5 (29.4)
<i>Vulpes vulpes</i>	26 (48.1)	13 (72.2)	7 (38.9)	5 (27.8)	49 (96.1)	16 (94.1)	17 (100)	16 (94.1)	13 (25.5)	4 (23.5)	2 (11.8)	7 (41.2)
<i>Sus scrofa</i>	7 (12.9)	2 (11.1)	2 (11.1)	2 (11.1)	22 (43.1)	9 (52.9)	10 (58.8)	3 (17.6)	1 (1.9)	1 (5.9)	0 (0)	0 (0)
<i>Capreolus capreolus</i>	2 (3.7)	0 (0)	0 (0)	1 (5.6)	6 (11.8)	2 (11.8)	2 (11.8)	2 (11.8)	1 (1.9)	0 (0)	0 (0)	1 (5.9)
<i>Martes foina</i>	16 (29.6)	6 (33.3)	5 (27.8)	4 (22.2)	10 (19.6)	5 (29.4)	4 (23.5)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Genetta genetta</i>	5 (9.3)	1 (5.6)	1 (5.6)	2 (11.1)	2 (3.9)	1 (5.9)	1 (5.9)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Felis silvestris</i>	0 (0)	0 (0)	0 (0)	0 (0)	3 (5.9)	1 (5.9)	0 (0)	2 (11.8)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Meles meles</i>	15 (27.8)	6 (33.3)	5 (27.8)	3 (16.7)	20 (39.2)	9 (52.9)	8 (47.1)	3 (17.6)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Lutra lutra</i>	2 (3.7)	0 (0)	0 (0)	1 (5.6)	5 (9.8)	2 (11.8)	2 (11.8)	1 (5.9)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Mustela lutreola</i>	0 (0)	0 (0)	0 (0)	0 (0)	2 (3.9)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Mustela putorius</i>	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Mustela nivalis</i>	0 (0)	0 (0)	0 (0)	0 (0)	1 (1.9)	1 (5.9)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Erinaceus europaeus</i>	0 (0)	0 (0)	0 (0)	0 (0)	1 (1.9)	0 (0)	0 (0)	1 (5.9)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Castor fiber</i>	0 (0)	0 (0)	0 (0)	0 (0)	1 (1.9)	0 (0)	0 (0)	1 (5.9)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Felis domesticus</i>	4 (7.4)	0 (0)	1 (5.6)	2 (11.1)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>Canis lupus familiaris</i>	5 (9.3)	2 (11.1)	1 (5.6)	1 (5.6)	1 (1.9)	1 (5.9)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)

summer 8.2E-10, 2.4E-13 and 6.2E-08 *p*-values respectively). In fact, spotlight counts had a poorer richness value when compared with the other two methods (Fig. 3).

Variation around the Year

There were no significant differences among the three seasons when applying the same sampling methodology (*p*-value >0.05 – for camera trapping, spotlight counts and indirect index for winter and spring comparison was of 0.748, 0.065, 0.064 respectively; for winter and summer comparison was of 0.083, 0.530, 0.090 respectively; and for spring and summer comparison was of 0.164, 0.075, 0.779 respectively). The maximum number of species (thirteen species) was seen in the winter sampling when using the indirect index (tracks and faeces sampling), as also occurred with the maximum number of species potentially seen (asymptote with 32 species – although we were surveying only 17 species, this represents method potential and the relation with the survey effort made) (Fig. 3).

Discussion

Taking into account our results, the method with the best cost-performance balance for monitoring mammals in a Mediterranean farmland was the indirect index. The Shannon index revealed to be a well-suited index for this type of work, giving us useful graphical information to assess methods performance that we discuss over this section. When testing the same method (for the three methods tested), there were no significant differences in the number of species detected among seasons, meaning that it is possible that the season chosen to perform the sampling is not relevant when considering the monitoring of the mammal community on Mediterranean farmlands. This is not in accordance with the premise that weather conditions affect indirect methodologies counts (Lyra-Jorge et al. 2008). The other two methods were not carried out when weather conditions were particularly harsh (e.g. rain), although detectability also varies with and within seasons, even with favourable weather conditions, owing to the behavioural and ecological traits of species (Fragoso et al., 2016). The indirect index was significantly different when compared with the spotlight counts, which may be related to the most powerful detection of species registered when using this type of sampling, due to a greater detectability when dealing with objects, rather than animals (with a maximum of 13 species seen, versus 4 species when using spotlight counts).

The two methodologies used to extract the species richness (random and exact method), gave us similar results, which means that at a first sight sampling design was correct and the conclusions can be meaningful. Asymptotes were achieved with a different sampling effort for the three methods. However, it is worth noting that these values refer to the effort required to find the maximum number of species based on those which were registered in each case scenario, which varies widely (from 3 species seen in spotlight counts in spring to 13 species registered using the indirect index method in winter). Although the camera trapping and indirect methods registered close values for species richness in all three seasons, the asymptotes (mainly as regards the winter sampling) seem to be a little far apart, with the indirect index asymptote attaining more than 30 species. Although we were prospecting only 17 species, this indicates that this type of indirect indices has the potential to detect a greater number of species. Taking into account the need to choose a method based in several aspects of the survey, as the ecology of the studied species and logistical and budget issues (Mayle et al. 1999, Valente et al. 2014), we drawn a global picture for the three tested methods. Indeed, we can observe that the method with the best cost-performance balance is the indirect

Table 2
Mean Shannon index values with confidence intervals, standard deviation and asymptote values (from exact method, see text for details) for the three different methods (camera trapping, indirect index and spotlight counts) in the three seasons sampled (winter, spring and summer).

		Mean Shannon Index	Lower 95% confidence interval	Upper 95% confidence interval	Standard deviation	Asymptote
Camera	Winter	0.576	0.316	0.837	0.523	10.578
	Spring	0.344	0.103	0.585	0.485	13.868
	Summer	0.413	0.112	0.714	0.605	16.689
Indirect index	Winter	0.899	0.582	1.216	0.616	32.281
	Spring	1.010	0.830	1.191	0.351	10.860
	Summer	0.583	0.287	0.878	0.575	21.712
Spotlight counts	Winter	0.257	0.091	0.422	0.323	4.474
	Spring	0.024	0.000	0.075	0.099	3.699
	Summer	0.190	0.028	0.352	0.315	5.116

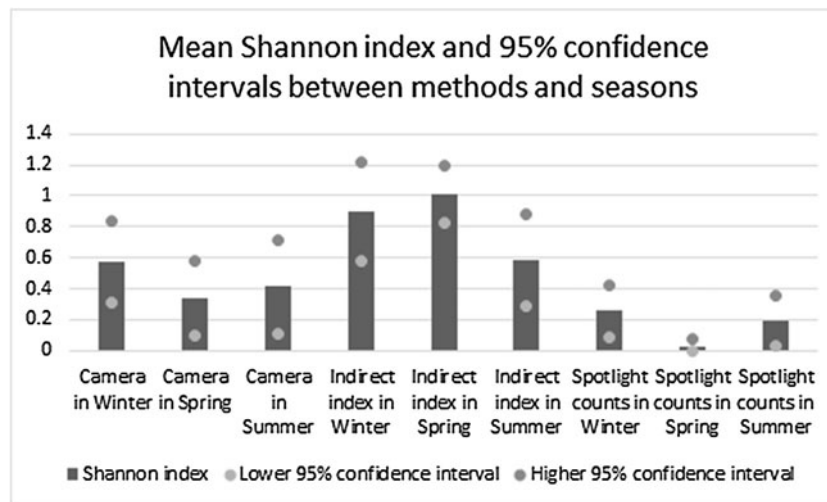


Fig. 3. Mean Shannon index values and 95% confidence intervals values for the three methods tested in the three tested seasons.

index which, with 24 hours of sampling and a low financial cost, can achieve relatively satisfactory results. This indicates that, in addition to providing robust results and being the cheapest of the three methodologies tested, this method could be the most appropriate with which to sample large study areas with potentially good results. The spotlight count method, meanwhile, involved the smallest amount of field work, but also attained the poorest results with a median financial cost. It is, on the other hand, worth noting the great amount of labour hours (and consequently financial costs and logistical constraints, being indeed the most expensive method) required with the camera trapping method (84 hours). However, the survey effort appears to be related to the results achieved, which although limited by the reduced number of cameras in the field, accorded with the species seen and the asymptotes attained. Nevertheless, [Tobler et al. \(2008\)](#) showed that camera trapping requires a substantial effort to register some species. In their study, 3 or less photos were taken of some species in 3840 camera days, while our study consisted of a total of 378 camera days. In contrast, [Rowcliffe et al. \(2008\)](#) consider that in some cases, only 100 camera days can yield robust results, depending on the density of the species studied (however, there were some species for which 1000 camera days still did not show reasonable results). Furthermore, [Lyra-Jorge et al. \(2008\)](#) confirmed that detection by camera traps may be dependent on the animal's body mass, i.e. they attain a major detectability for large species. This is in accordance with our results, since, for example, none of the mustelids present in the study area were found when using this method. Limitations such as this are also applicable to direct observations (such as spotlight counts), since animals with a large body size are the easiest to see and are consequently easily recorded ([Silveira et al. 2003](#)). The indirect index also has its drawbacks, as species identification

in track plots is strongly dependent on the researcher's ability to identify the material recorded ([Lyra-Jorge et al. 2008](#)), which can have direct implications as regards the results achieved, however the field work was performed by trained biologists with posterior identification of species faeces that can originate ambiguous situations, which diminish the bias from this source. In order to reduce other bias sources, we are aware of the power and benefits of modelling such data ([MacKenzie et al. 2002](#), [Rich et al. 2017](#)), however this does not potentially make sense in a smaller area such as this one, in which we do not make inferences for contiguous areas.

Conclusions

The search for the best methodology with which to monitor wild mammal populations is not always straightforward. This work, although limited in space and by the field effort carried out (which was constrained by budget limitations), constitutes a contribution to the search for an optimum methodology with which to assess species distribution (presence/absence) on Mediterranean farmland, which we considered to be the indirect index (i.e. with the best detectability for mammals, no significant seasonal-dependence and with the best cost-performance balance). Monitoring populations is indeed a mandatory phase in order to build management plans that reflect the true population status and can follow population dynamics and ecosystem balance, thus this work constitutes a solid step in this search.

Declarations of interest

None.

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