Eur J Wildl Res
DOI 10.1007/s10344-011-0524-z

ORIGINAL PAPER

Towards a standardized index of European rabbit abundance in Iberian Mediterranean habitats

Javier Fernandez-de-Simon · Francisco Díaz-Ruiz · Francesca Cirilli · Francisco Sánchez Tortosa · Rafael Villafuerte · Miguel Delibes-Mateos · Pablo Ferreras

Received: 15 December 2010 / Revised: 15 February 2011 / Accepted: 16 February 2011

Abstract European rabbits *Oryctolagus cuniculus* are a keystone species in Iberian Mediterranean ecosystems. However, the reliability of methods for estimating rabbit abundance, particularly when at low numbers, is not well understood. Further, better standardization of these methodologies would allow abundance estimates to be more reliably compared between areas and periods. Consequently, we compared several frequently used methods of estimating rabbit abundance and assessed their advantages and disadvantages. During the summers of 2008 and 2009, in 11 localities of central-southern Spain we undertook (a) driving transect counts of rabbits, either at dusk or at night, (b) linear transects on foot recording rabbit signs, (c) cleared-plot pellet counts at permanent plots, and (d) standing crop counts, both with and without habitat stratification. Density estimated at night from driving transects using the Distance Sampling method (the reference method against which all other indices were compared) varied from 0 to 2.69 rabbits ha^{-1}. Most pellet-count indices were significantly related to the reference method. In particular, cleared-plot pellet counts in permanent plots corrected for pellet persistence showed the best correlation with the reference method. In contrast, latrine counts were not related to the reference method index, and we recommend against their use. A standard methodology based on cleared-plot pellets counts could be used to monitor rabbit abundance on a large scale.

Keywords Abundance indices · Density estimates · Lagomorphs · *Oryctolagus cuniculus* · Reference method · Standardization

Communicated by C. Gortázar

J. Fernandez-de-Simon (✉) · F. Díaz-Ruiz · R. Villafuerte · M. Delibes-Mateos · P. Ferreras
Instituto de Investigación en Recursos Cinegéticos, IREC (CSIC–UCLM–JCCM),
Ronda de Toledo s/n,
13071 Ciudad Real, Spain
e-mail: javier.fernandez@uclm.es

F. Cirilli · F. S. Tortosa
Department of Zoology, University of Córdoba,
Campus de Rabanales Ed. Darwin,
14071 Córdoba, Spain

Introduction

European rabbits *Oryctolagus cuniculus* are a key species in Iberian Mediterranean ecosystems (Valverde 1967; Delibes-Mateos et al. 2007), being consumed by more than 40 predator species (Delibes and Hiraldo 1981; Delibes-Mateos et al. 2008a). Rabbits also act in this area as ecosystem engineers because of their effect in vegetation (creation of open areas, preservation of plant species diversity, increased plant growth by inducing soil fertility) and also because they provide feeding resources (latrines) for many invertebrate species, and nest sites and shelter (burrows) for vertebrates and invertebrates (Delibes-Mateos et al. 2008a; Gálvez et al. 2009). Rabbits are also a popular small game species (Angulo and Villafuerte 2003). Although considered harmful pest species in other parts of the world (Williams et al. 1995; Lees and Bell 2008), European rabbits are only regarded as an agricultural pest in a few localized areas in Spain (Barrio et al. 2010b; Ríos 2010). Rabbit numbers on the Iberian peninsula have declined over recent decades, due mainly to habitat loss (Delibes-Mateos et al. 2010) and the occurrence of two viral diseases, myxomatosis during the 1950s (Muñoz 1960) and rabbit hemorrhagic disease (RHD) at the end of the 1980s (Villafuerte et al. 1995). Since the RHD outbreak in Doñana National Park (southwestern Spain), rabbit numbers have declined by an estimated 90% (Moreno et al. 2007). Most populations in Spain have continued to decline (Delibes-Mateos et al. 2008b), resulting in significant economic and ecological consequences for Iberian Mediterranean ecosystems (Delibes-Mateos et al. 2008a). Monitoring rabbit populations is currently a major challenge for conservation, making the development of widely applicable and reliable monitoring methods particularly important (Delibes-Mateos et al. 2009). For instance, the Iberian lynx (*Lynx pardinus*) is a critically endangered predator specialist on rabbits (Ferreras et al. 2011), and it has been calculated that the autumn mean rabbit density required for Iberian lynx residence is 1 rabbit ha^{-1}, while the spring mean rabbit density required for reproduction is 4.6 rabbits ha^{-1} (Palomares et al. 2001). Hence, large and cost-effective spatial monitoring projects to select areas suitable for lynx re-establishment are dependent on reliable and easy-to-perform indices of rabbit abundance (Ferreira and Delibes-Mateos 2011).

Direct and indirect methods are currently used to estimate rabbit abundances and population trends. Direct methods are based on surveys or counts of the animals, while indirect methods are based on the monitoring of animal signs (Acevedo et al. 2008). One of the most commonly used direct methods is counting individuals along linear transects, which can provide absolute estimates of density (Palomares 2001; Palomares et al. 2001; Martins et al. 2003; Barrio et al. 2010a) by using the Distance Sampling method (Buckland et al. 1993). The accuracy of this method is dependent on several assumptions (e.g., objects on the line or point are detected with certainty, objects do not move, measurements are exact; Thomas et al. 2010) and these may not always be met. A considerable sampling effort is also required to obtain sufficient number of sightings to reliably estimate density in low density populations (Newey et al. 2003). One alternative to the Distance Sampling method is the kilometric abundance index (sightings per kilometer; Beltrán 1991; Moreno et al. 2007; Williams et al. 2007), which is correlated with population density (Palomares et al. 2001; Barrio et al. 2010a) and provide data for a range of abundances. Rabbit counts also have several drawbacks, such as environment variables affecting the rabbit activity patterns (see e.g., Villafuerte et al. 1993; Twigg et al. 1998; Martins et al. 2003). Other direct methods used to estimate densities are based on live trapping, mostly by means of capture–recapture (e.g., Ballinger and Morgan 2002; Marchandeau et al. 2006) and the minimum number of individuals known to be alive (MNA, King and Wheeler 1985; Wood 1988).

Indices from rabbit signs are alternatives to direct methods. Counts of warrens are widely used (Myers et al. 1975) and can be corrected for distances traversed during these counts (Palomares 2001). Pellet counts per unit area can also be used to estimate abundance (Moreno and Villafuerte 1995; Cabezas and Moreno 2007; Delibes-Mateos et al. 2008b), as can latrines per unit of distance (Calvete et al. 2006). The standing crop count method involves counting pellets during only one visit, enabling a large area to be sampled. Nevertheless, to estimate absolute densities it is necessary to correct for defecation rates and pellet persistence (Putman 1984). The cleared-plot count method involves counting pellets that accumulate over a period of time in plots from which pellets had been previously removed (e.g., Palomares 2001). It is important to identify the period during which pellet accumulation and persistence is highest in order to reduce the zero counts in low density populations and the effect of pellet decay in pellet counts. This method is time consuming and labor intensive and it is necessary to optimize the time of visits (Massei et al. 1998). This method can provide reliable estimates of abundance when animal densities are low (Murray et al. 2002), a situation limiting the accuracy of other methods. Other signs, such as scrapes and tracks, are rarely used to obtain indices of rabbit abundance (but see Twigg et al. 2001).

Studies monitoring the abundance and trends of rabbit populations in the Iberian Peninsula have used most of the methods described above (Calvete et al. 2006; Moreno et al. 2007; Williams et al. 2007; Delibes-Mateos et al. 2008b). Unfortunately, due to the variety of methods used, it is not always possible to compare results (i.e., rabbit abundances) among studies (Delibes-Mateos et al. 2009). Several studies have attempted to standardize methodologies by comparing indices with a reference method, which provides less biased estimates of population size and absolute densities, often using live trapping indices (Wood 1988; Ballinger and Morgan 2002; Marchandeau et al. 2006). However, the high costs in human effort, time, and logistical resources prevent the use of live trapping in wide-scale studies, in many localities and in medium/long-term monitoring programs. As a result, most studies have been based on rabbit counts (Myers et al. 1975; Palomares 2001; Palomares et al. 2001; Barrio et al. 2010a), though as the same methods are not simultaneously performed in the same study areas, the optimal method for specific conditions and objectives is unclear. Moreover, fewer studies have used a wider approach than using a locality scale. We therefore sought to compare several commonly used methods, and assessed the advantages and disadvantages of each. We propose common and comparable methodologies for assessing rabbit abundances according to the objectives and scale of each study.

Material and methods

Areas and periods of study

Field work was performed in 11 localities of central-southern Spain (Fig. 1), with different rabbit abundances but similar habitat structures and climate. All localities had a Mediterranean climate, characterized by wet, mild winters and warm, dry summers with marked drought periods. Habitats were composed mainly of Mediterranean scrubland, pastures, croplands, "dehesas" (savanna-like formations that combine pastures with intermittent cereal cultivation in park-like oak woodlands; Blondel and Aronson 1999), and tree plantations. We used the same methods (Table 1, further details below) to estimate rabbit abundance in each locality during the summers (June–August) of 2008 (localities 1–8) and 2009 (localities 9–11). For some indices (density estimates at night, DEN-N; kilometric abundance index at night, KAI-N; cleared-plot pellet counts corrected and uncorrected by persistence, COR and UNC respectively, see below and Table 1) and localities (1–8), data were also collected in the winter–spring (December–May) seasons of 2007–2009 and in summer 2007.

Driving transect counts

Rabbits are active during twilight and at night, with two activity peaks that coincide with sunrise and sunset (Díez et al. 2005). However, rabbit counts at night provide more precise estimates than at dusk (Barrio et al. 2010a). We counted rabbits at dusk (starting 1 h before sunset) and at night (starting 2 h after sunset) in good weather conditions (no strong winds and no rainfall). Surveys were undertaken at each locality and at each of these periods of the day (dusk and night) for three consecutive days, unless climatic or logistic circumstances prevented so. Transects were travelled in an all-terrain pickup at a speed of 15 km/h along dirt tracks varying in minimum length (mean± SE=14.04±1.61 km) and traversing different habitats with good visibility. Most of the sightings were observed within 100 m, though some observations were occasionally obtained up to 200 m approximately. One observer stood at the trunk of an all-terrain pickup observing the 180° area ahead. At night, a 1 million-candle-power halogen spotlight was used. The distance (m) of each rabbit from the observer was measured using a telemeter, and the angle between the transect line and the line from the observer to the animal was measured using a compass. We calculated an average kilometric abundance index (rabbits seen per kilometer, KAI-N and KAI-D) at each locality by pooling the data obtained from the three replicates, and we estimated the rabbit density (rabbits per hectare, DEN-N and DEN-D) with the Distance Sampling method (Buckland et al. 1993), using the Fourier series estimator as detection function in TRANSECT software (Burnham et al. 1980).

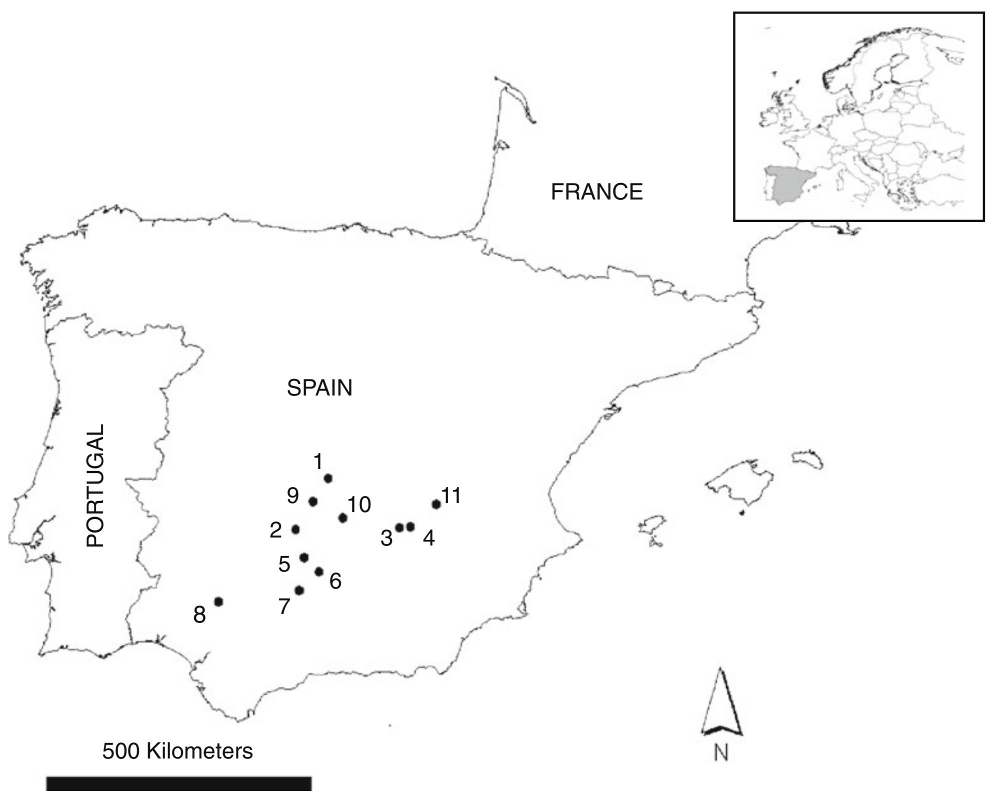

Fig. 1 Location of study areas in the Iberian Peninsula

Table 1 Variables obtained with the different methods

Driving transect counts	
DEN-N	*Density estimates at night (rabbits ha^{-1})*
KAI-N	Kilometric abundance index at night (rabbits km^{-1})
DEN-D	Density estimates at dusk (rabbits ha^{-1})
KAI-D	Kilometric abundance index at dusk (rabbits km^{-1})
Linear transects on foot	
LATR	Latrine index (latrines km^{-1})
WARR	Warren index (warren entrances km^{-1})
RAB	Rabbits seen index (rabbits km^{-1})
SCR	Scrapes index (scrapes km^{-1})
PEL	Standing crop pellet index (pellets m^{-2}) collected along the transect
RDI	Relative density index (PCA from LATR, WARR, RAB and SCR)
Local pellet counts in permanent plots	
COR	Cleared-plot pellet counts corrected by persistence (pellets m^{-2} day^{-1})
UNC	Uncorrected cleared-plot pellet counts (pellets m^{-2} day^{-1})
STA	Standing crop counts (pellets m^{-2}) in the high-density area
Stratified pellet counts	
STR	Standing crop counts (pellets m^{-2}) at stratified transects

The reference method (DEN-N) is highlighted in italics

Linear transects on foot

Four-kilometer transects were walked by two observers traversing areas favorable to rabbits, mainly those ecotones between Mediterranean scrubland and pastureland or cropland (Delibes-Mateos et al. 2008b). The variables assessed along the transect were rabbits (rabbits seen per kilometer, RAB), latrines (latrines per kilometer, LATR), scrapes (scrapes per kilometer, SCR), and warrens (warren entrances per kilometer, WARR) indices. For the latter index, all entrances (either active, inactive, or unknown) were considered, since >90% of the entrances observed were active, with little difference in indices using only active entrances or total entrances. A standing crop pellet index (pellets per square meter, PEL) was obtained from 40 circular plots of 0.5 m^2 each, regularly distributed along the transect (one plot per 100 m). A relative density index (RDI) was obtained as the first axis score from a Principal Component Analysis (PCA, Zar 1984) of four raw variables that were highly correlated (RAB, LATR, SCR, and WARR) following Delibes-Mateos et al. (2008b) and Villafuerte et al. (1998).

Local pellet counts in permanent plots

A 30×90-m grid was set in the area with the highest rabbit abundance at each locality, identified from the 4-km transect on foot (see above). Every month, we performed cleared-plot pellet counts on the sampling areas, which consisted of 40 plots sited as four parallel lines of 10 plots each (Fernandez-de-Simon et al. accepted). The distance between each plot and line was 10 m, resulting in a regular sampling grid. After counting all pellets within the 0.5-m^2 circular plot centered at a wooden stake, we cleared the plot and, approximately 1 month later, we again counted the pellets in each plot. For each month, we obtained an uncorrected daily pellet accumulation rate (UNC) by calculating the average number of pellets per square meter per day (e.g., Catalán et al. 2008). We also estimated the persistence of rabbit pellets for each month and locality by placing 10 pellets marked with nail polish in each of five plots (n=50 marked pellets per locality per month, Fernandez-de-Simon et al. accepted). Remaining marked pellets were counted 1 month later, and daily persistence was calculated by assuming a constant daily decay between counts. The daily pellet accumulation rate corrected for persistence (COR) was calculated according to Palomares (2001), using the formula:

$$\mathrm{COR} = \mathrm{UNC}(\mathrm{DPR} - 1)/\left(\mathrm{DPR}^{\mathrm{nd}} - \mathrm{DPR}\right)$$

where DPR is the daily persistence rate of pellets, and nd is the number of days elapsed since the last count.

We used the data from the month closest to the night counts dates, as the density estimates at night (DEN-N) were used as the reference method (see below). We also measured the standing crop counts (pellets per square meter, STA) in this high density area by counting dung pellets within 40 0.5-m^2 circular plots interspersed halfway among the cleared plots within the same line.

Stratified pellet counts

We walked seven 400-m transects in each locality that contained 40 standing crop counts in regularly distributed 0.5-m^2 circular plots (one plot every 10 m). Transects were stratified among the habitats according to the proportion of the area occupied by each habitat type in each locality. The rabbit abundance index was determined by calculating the average number of pellets per square meter (STR, Table 1) for each locality.

Statistical analyses

Density estimates at night (rabbits per hectare, DEN-N) was considered the reference method for estimating rabbit density when comparing the different techniques (Barrio et al. 2010a). The Distance Sampling method has been used previously to estimate rabbit density (Palomares 2001; Palomares et al. 2001; Martins et al. 2003; Barrio et al. 2010a). Linear regression was used to test the ability of the indices of rabbit abundance (independent variables) to predict DEN-N (dependent variable). First, we calculated linear regression using the data from the summers of 2008 and 2009, when all methods were assessed at each locality. All regressions were forced through zero intercept, as the indices should be zero when the rabbit density is zero. The degree of fitting was assessed using the coefficient of determination (r^2). Relationships were considered significant when P <0.05. Afterwards, we tested whether these relationships were maintained for variables KAI-N, UNC, and COR during other seasons, years, and localities. Thus, a general linear model (GLM) was used separately for each of these three variables to test the effect of locality (1–8, random effect), season (winter–spring and summer, fixed effect), and year (2007 and 2008, random effect) on the slope of the linear regression between the reference method and each index of abundance (dependent variable). Significant effects by season or year would preclude pooling the data in the linear regressions. All analyses were performed using STATISTICA 6.0 (StatSoft Inc. 2001).

Results

Density estimates

In summer and in four localities (6, 2, 9, and 8), the low number of sightings precluded reliable estimation of DEN-N. In locality 6, no rabbits were observed during the three nights. Consequently, in these localities, rabbit densities were indirectly estimated from the linear regression between density estimates (dependent variable) and kilometric abundance index (independent variable, Palomares et al. 2001). The mean±SE rabbit density considering all localities was then 0.85±0.29 rabbits ha^{-1} (n=11, range 0–2.69 rabbits ha^{-1}, Fig. 2). To calculate the density estimates at dusk (DEN-D) in localities with insufficient sightings (2, 6, and 9), we used the linear regression between density estimates at dusk (dependent variable) and kilometric abundance index at dusk (independent variable).

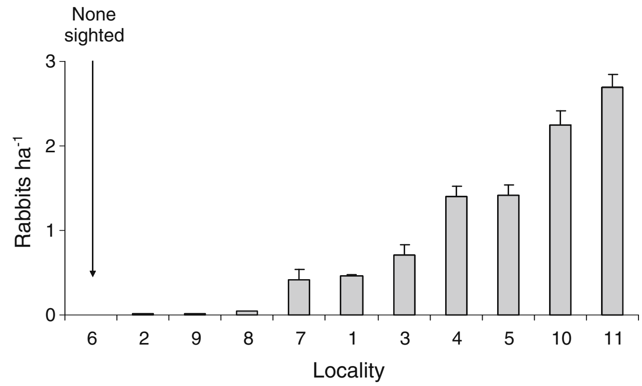

Fig. 2 Rabbit density at each locality estimated from driving transects using the Distance Sampling method at night (DEN-N). *Error bars* represent the standard errors. *Numbers* correspond to the localities codes shown in Fig. 1

Comparisons between indices

Of the variables tested, the pellet-count indices were the most significantly related to the reference method (Table 2, Fig. 3). Among these, COR showed the best fit to the reference method (r^2=0.79, P≤0.001). Nevertheless, we observed a low difference in r^2 among all indices based on pellet counts (see Table 2 and Fig. 3). RAB, based on rabbit counts during linear transects on foot, was also significantly related to the reference method (r^2=0.68, P≤0.001). KAI-N was also significantly associated with the reference method (r^2=0.73, P<0.05), although this was expected because both KAI-N and DEN-N were derived from the same datasets. KAI-D was also significantly related to the reference method, although with lower r^2 than the same index at night (KAI-N). The variables RDI (r^2=0.5, P<0.05) and WARR (r^2=0.45, P<0.05), both based on linear transects on foot, were also significantly related to the reference method. In contrast, SCR, LATR, and DEN-D were not significantly associated with the reference method (P>0.05).

With respect to the model during other seasons, all independent factors except year (P=0.46) significantly affected the slope between KAI-N and the reference method (locality, $F_{7,17}$=3.30, P=0.02; season, $F_{1,17}$=5.78, P=0.03). Pooling data from different years (2007–2009), we found that KAI-N was significantly related to the reference method both in winter–spring (r^2=0.63, P≤0.001, Table 3, Fig. 4) and in summer (r^2=0.78, P≤0.001, Table 3, Fig. 4). DEN-N estimates were obtained from these relationships and for each season separately in cases where there were insufficient observations (see above for details). Because no factor was significant for the UNC and COR models (P>0.05), we pooled the data among seasons,

Table 2 Significant linear regressions ($P<0.05$) between the reference method of rabbit density estimated at night from driving transect spotlight counts (DEN-N, dependent variable), and several rabbit abundance indices in decreasing order of coefficient of determination (r^2)

Independent variable	r^2	Degrees of freedom	F	P value	Equation
COR	0.79	1,10	38.27	≤0.001	$y=0.708x$
PEL	0.77	1,9	30.87	≤0.001	$y=0.018x$
UNC	0.73	1,10	27.5	≤0.001	$y=0.759x$
KAI-N	0.73	1,7	18.64	0.003	$y=0.154x$
STR	0.71	1,9	22.28	≤0.001	$y=0.026x$
STA	0.7	1,10	23.29	≤0.001	$y=0.004x$
RAB	0.68	1,10	21.55	≤0.001	$y=0.153x$
KAI-D	0.5	1,9	9.04	0.02	$y=0.115x$
RDI	0.5	1,10	9.82	0.01	$y=0.798x$
WARR	0.45	1,10	8.11	0.02	$y=0.025x$

Data were collected during summer surveys of 11 localities in central-southern Spain, shown in Fig. 3. Codes of variables are shown in Table 1

years, and localities for each variable. Both UNC ($r^2=0.45$, $P\leq0.001$) and COR ($r^2=0.4$, $P\leq0.001$) were significantly related to DEN-N (Table 3, Fig. 4).

Discussion

Using DEN-N as a reference standard, we evaluated the applicability of most rabbit abundance indices used previously, as described in the literature. We found that most pellet-count indices may reliably estimate abundances, whereas other indices such as scrape and latrine indices and density estimates at dusk were not related to abundance on a regional scale. Among the pellet-count indices, cleared-plot pellet counts corrected and uncorrected by persistence were similarly related to the reference method, as previously demonstrated (Wood 1988; Palomares 2001). Estimating these indices in the zone of highest relative abundance within a locality may minimize the number of zero counts in areas of low rabbit density. In yearly monitoring of Mediterranean habitats, it is advisable to obtain pellet-count indices during early summer, at the start

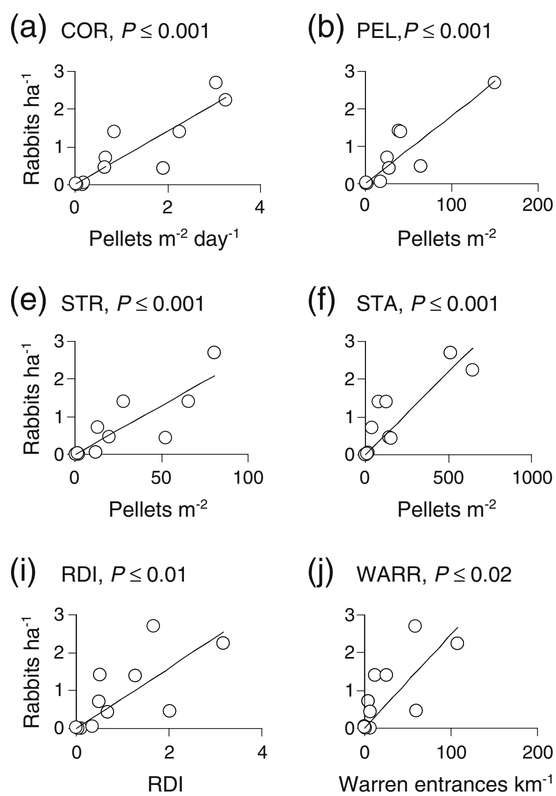

Fig. 3 Relationships (linear regressions) and P values between rabbit density estimates at night (DEN-N, dependent variable and reference method) and the independent variables (**a**–**j**) in summer. The RDI axis lacks units because it was obtained from a principal component analysis of the latrine (LATR), warren (WARR), rabbits (RAB), and scrapes (SCR) indices. Codes of variables are shown in Table 1

Table 3 Significant linear regressions ($P<0.05$) between the reference method of rabbit density estimated at night from driving transect spotlight counts (DEN-N, dependent variable), and several rabbit abundance indices in decreasing order of coefficient of determination (r^2)

Season	Independent variable	r^2	Degrees of freedom	F	P value	Equation
Winter–spring	KAI-N	0.63	1,17	28.5	≤0.001	$y=0.103x$
Summer	KAI-N	0.78	1,14	49.11	≤0.001	$y=0.161x$
Pooled	UNC	0.45	1,41	33.85	≤0.001	$y=0.625x$
	COR	0.4	1,40	26.67	≤0.001	$y=0.515x$

Data were collected during surveys in different seasons (winter–spring and summer) at 11 localities in central-southern Spain, and are shown graphically in Fig. 4. Significant regressions pooling the data from different seasons (pooled) are also exhibited. See Table 1 for codes of variables

of the dry season, thereby reducing the biases associated with differences in rabbit pellet persistence due to the effects of rainfall (Fernandez-de-Simon et al. accepted; Iborra and Lumaret 1997). Standing crop counts in summer were also related to density, resulting in only a slightly lower fit than COR. Therefore, if two visits are not possible owing to logistical constraints, standing crop counts may be a valid option for estimating rabbit abundance. Long-term monitoring of rabbit populations may be accomplished by repeated cleared-plot pellet counts over time because they were found to be related to density during both summer and winter–spring periods. Pellet counts, however, are affected by other factors, such as the non-random distribution of pellets over the area due to heterogeneity in the environment (that may be corrected through stratified transects) or the variation in defecation rates between populations or even between individuals (Putman 1984).

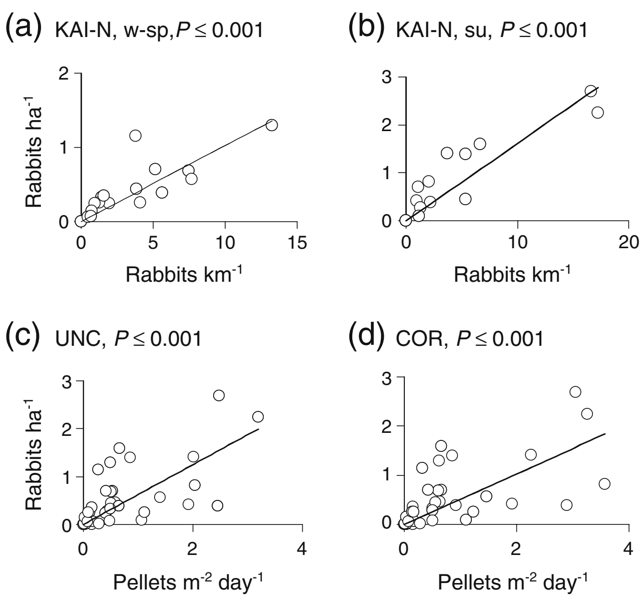

Fig. 4 Relationships (linear regressions) and P values between rabbit density estimates at night DEN-N (dependent variable and reference method) and **a** kilometric abundance index at night (KAI-N) in winter–spring, **b** KAI-N in summer, **c** cleared-plot pellet counts uncorrected by persistence (UNC) with pooled dataset from both seasons, and **d** cleared-plot pellet counts corrected by persistence (COR) with pooled dataset from both seasons

Of the driving transect indices, KAIs were related to the reference method for the seasons tested, although the relationships with DEN-N differed between seasons. Caution must be taken with KAIs because they do not control for habitat variation in rabbit detectability (Marchandeau et al. 2006). Furthermore, sight counts may underestimate rabbit abundance (Twigg et al. 1998; Poole et al. 2003) since the proportion of rabbits seen in a population are inversely related to rabbit density due to social interactions (Twigg et al. 1998). However, at rabbit densities found in the Iberian Peninsula, much lower than those found in Australia (Barrio 2010), the proportion of active rabbits is relatively high during the night and therefore spotlight counts are less affected by inactive rabbits (Twigg et al. 1998). KAI-D was related to the reference method, albeit with a lower fit than KAI-N, as found by Barrio et al. (2010a). DEN-D was not related to the reference method, despite the fact that transects were identical and performed on the same days. This may be related to a rabbit avoidance of dusk-time hours to prevent higher predation risk (hunting and diurnal raptors, Fernández de Simón et al. 2009) or to an avoidance of dirt tracks during the day to prevent human disturbance (similarly as the roe deer *Capreolus capreolus*; Ward et al. 2004). Driving transect counts may be affected by environmental factors such as visibility, wind speed, and rainfall, which may affect rabbit activity (Villafuerte et al. 1993; Twigg et al. 1998; Martins et al. 2003). Similarly, numbers of predators and hunting pressure should be considered when conducting these surveys as rabbits may shift their activity to reduce the predation risk (Fernández de Simón et al. 2009). All these factors make advisable to conduct replicates and to complement these estimates with alternative indices to increase the results reliability.

The lack of significant relationships of LATR and SCR with the reference method may be due to the strong behavioral component of latrine (Monclús and de Miguel 2003) and scrape (Burggraaf-van Nierop and van der Meijden 1984) abundance, which may not be linked to rabbit abundance. Biases concerning latrine counts may also derive from subjective criteria for latrines (Delibes-Mateos et al. 2008b). Although these indices may locally result in reproducible variations in densities, we do not recommend their use in regional monitoring programs if they are not complemented by additional rabbit data (e.g., RDI). Despite LATR being recently adopted as a standardized methodology for monitoring rabbits in Portugal (Ferreira and Delibes-Mateos 2011), our findings indicate this method is not reliable for monitoring abundance on large spatial and temporal scales, being preferable other pellet-count indices.

Recommendations

Overall, DEN-N may be used to provide reliable estimate of rabbit abundance when sufficient resources are available and when terrain, habitat, and weather conditions permit. In other circumstances (wide-scale monitoring programs, many localities surveyed, long-term monitoring), pellet-count indices (except LATR) may be the most reliable for obtaining estimates of abundance when rabbit numbers are low to moderate. In summer and depending on the scale, the best indices for estimating rabbit abundance are the cleared-plot pellet counts corrected by persistence (small scale), the standing crop pellet index collected along a transect (intermediate scale), and the standing crop index at stratified transects (large scale). However, other indices obtained from linear transects on foot (rabbit, relative density, and warren indices) may also be appropriate. Using an abundance index without knowing its relationship to estimated densities throughout the range of abundances likely to be encountered may compromise the reliability of the results. It would also make it difficult to compare these results with those of other studies. Therefore, standardization of abundance indices allows the most appropriate method to be selected, depending on the characteristics and objectives of the study. This can be performed using the methods described in this study, though the standardization with techniques such as density estimates from night counts may not always be possible. In this situation, equations provided here may be useful for rabbit monitoring in other Iberian Mediterranean areas. Further research is also encouraged to standardize the abundance and density indices in other areas or with other reference methods (e.g., live trapping). Here, we concluded that COR may be optimal for monitoring rabbits on a large scale and may be implemented as standardized methods for rabbit conservation programs, as well as for management issues (e.g., hunting, agriculture, etc.). Administrators, researchers, and other personnel monitoring rabbits should promote the use of these methods for uniform monitoring and should abandon less reliable methods such as LATR. Effective monitoring to manage and conserve rabbit populations and their endangered predators is only possible when standardized and comparable monitoring methods are used.

Acknowledgments This work is a partial result from the projects CGL2005-02340, CGL2009-10741, and CGL2009-11665 funded by the Spanish Ministry of Science and Innovation and EU-FEDER funds, and projects POII09-0099-2557 and 07/NAT/E/000742 funded by the Junta de Comunidades de Castilla-La Mancha with EU-FEDER and LIFE funds, respectively. Javier Fernandez-de-Simon benefits from a FPI scholarship and Miguel Delibes-Mateos is currently holding a Juan de la Cierva research contract, both funded by the Spanish Ministry of Science and Innovation and the European Social Fund. We are indebted to the landowners and personnel of the study areas where we undertook this study. We thank F. Aparicio, I.C. Barrio, J.A. Blanco-Aguiar, F. Buenestado, A. Camarero, J. Cañadilla, F. Castro, B. Cooke, F. Fernández de Simón, C. Ferreira, J.E.C. Flux, J.L. García-Moreno, C. Iriarte, L. E. Mínguez, R.P. Pech, P. Peiro, S.O. Petrovan, N. Reid, P. Rodríguez, and C. Rouco for their kind collaboration and comments. G. Nugent and an anonymous reviewer provided interesting suggestions that improved the manuscript.

References

Acevedo P, Ruiz-Fons F, Vicente J, Reyes-García AR, Alzaga V, Gortázar C (2008) Estimating red deer abundance in a wide range of management situations in Mediterranean habitats. J Zool 27:37–47

Angulo E, Villafuerte R (2003) Modelling hunting strategies for the conservation of wild rabbit populations. Biol Conserv 115:291–301

Ballinger A, Morgan DG (2002) Validating two methods for monitoring population size of the European rabbit (*Oryctolagus cuniculus*). Wildl Res 29:431–437

Barrio IC (2010) The rabbit as an agricultural pest in its native distribution range. Dissertation, Universidad de Castilla-La Mancha

Barrio IC, Acevedo P, Tortosa FS (2010a) Assessment of methods for estimating wild rabbit abundance in agricultural landscapes. Eur J Wildl Res 56:335–340

Barrio IC, Bueno GC, Tortosa FS (2010b) Alternative food and rabbit damage in vineyards of southern Spain. Agr Ecosyst Environ 138:51–54

Beltrán JF (1991) Temporal abundance pattern of the wild rabbit in Doñana, SW Spain. Mammalia 55:591–599

Blondel J, Aronson J (1999) Biology and wildlife of the Mediterranean region. Oxford University Press, Oxford

Buckland ST, Anderson DR, Burnham KP, Laake JL (1993) Distance sampling: estimating abundance of biological populations. Chapman and Hall, London

Burggraaf-van Nierop YD, van der Meijden E (1984) The influence of rabbit scrapes on dune vegetation. Biol Conserv 30:133–146

Burnham KP, Anderson DR, Laake JL (1980) Estimation of density from line transect sampling of biological populations. Wildl Monogr 72:1–202

Cabezas S, Moreno S (2007) An experimental study of translocation success and habitat improvement in wild rabbits. Anim Conserv 10:340–348

Calvete C, Pelayo E, Sampietro J (2006) Habitat factors related to wild rabbit population trends after the initial impact of rabbit haemorrhagic disease. Wildl Res 33:467–474

Catalán I, Rodríguez-Hidalgo P, Tortosa FS (2008) Is habitat management an effective tool for wild rabbit (*Oryctolagus cuniculus*) population reinforcement? Eur J Wildl Res 54:449–453

Delibes M, Hiraldo F (1981) The rabbit as prey in the Iberian Mediterranean ecosystem. In: Myers K, MacInnes CD (eds) Proceedings of the World Lagomorph Conference. University of Guelph, Ontario, pp 614–622

Delibes-Mateos M, Redpath SM, Angulo E, Ferreras P, Villafuerte R (2007) Rabbits as keystone species in southern Europe. Biol Conserv 137:149–156

Delibes-Mateos M, Delibes M, Ferreras P, Villafuerte R (2008a) Key role of European rabbits in the conservation of the western Mediterranean basin hotspot. Conserv Biol 22:1106–1117

Delibes-Mateos M, Ferreras P, Villafuerte R (2008b) Rabbit populations and game management: the situation after 15 years of rabbit haemorrhagic disease in central-southern Spain. Biodivers Conserv 17:559–574

Delibes-Mateos M, Ferreras P, Villafuerte R (2009) European rabbit population trends and associated factors: a review of the situation in the Iberian Peninsula. Mamm Rev 39:124–140

Delibes-Mateos M, Farfán MA, Olivero J, Vargas JM (2010) Land use changes as a critical factor for long-term wild rabbit conservation in the Iberian Peninsula. Environ Conserv 37:169–176

Díez C, Pérez JA, Prieto R, Alonso ME, Olmedo JA (2005) Activity patterns of wild rabbit (*Oryctolagus cuniculus*, L. 1758), under semi-freedom conditions, during autumn and winter. Wildl Biol Pract 1:41–46

Fernandez-de-Simon J, Díaz-Ruiz F, Villafuerte R, Delibes-Mateos M, Ferreras P (2011) Assessing predictors of pellet persistence in rabbits: towards reliable population estimates from pellet counts. Wildl Biol

Fernández de Simón J, Díaz-Ruiz F, Cirilli F, Sánchez F, Villafuerte R, Ferreras P (2009) Relación entre la presión de depredación y los ritmos de actividad del conejo de monte: aplicación a las estimas de densidad. In VIII Jornadas de la Sociedad Española para la Conservación y Estudio de los Mamíferos. Sociedad Española para la Conservación y Estudio de los Mamíferos, Málaga, pp 72

Ferreira C, Delibes-Mateos M (2011) Wild rabbit management in the Iberian Peninsula: state of the art and future perspectives for Iberian lynx conservation. Wildl Biol Pract. Available at http://socpvs.org/journals/index.php/wbp/article/view/146/212. Accessed 15 February 2011

Ferreras P, Travaini A, Zapata SC, Delibes M (2011) Short term responses of mammalian carnivores to a sudden collapse of rabbits in Mediterranean Spain. Basic Appl Ecol. doi:10.1016/j.baae.2011.01.005

Gálvez L, Belliure J, Rebollo S (2009) European rabbits as ecosystem engineers: warrens increase lizard density and diversity. Biodivers Conserv 18:869–885

Iborra OL, Lumaret JP (1997) Validity limits of the pellet group counts in wild rabbit (*Oryctolagus cuniculus*). Mammalia 61:205–218

King DR, Wheeler SH (1985) The European rabbit in south-western Australia I. Study sites and population dynamics. Aust Wildl Res 12:183–196

Lees AC, Bell DJ (2008) A conservation paradox for the 21st century: the European wild rabbit *Oryctolagus cuniculus*, an invasive alien and an endangered native species. Mamm Rev 38:304–320

Marchandeau S, Aubineau J, Berger F, Gaudin JC, Roobrouck A, Corda E, Reitz F (2006) Abundance indices: reliability testing is crucial—a field case of wild rabbit *Oryctolagus cuniculus*. Wildl Biol 12:19–26

Martins H, Barbosa H, Hodgson M, Borralho R, Rego F (2003) Effect of vegetation type and environmental factors on European wild rabbit *Oryctolagus cuniculus* counts in a southern Portuguese montado. Acta Theriol 48:385–398

Massei G, Bacon P, Genov PV (1998) Fallow deer and wild boar pellet group disappearance in a Mediterranean area. J Wildl Manage 62:1086–1094

Monclús R, de Miguel FJ (2003) Distribución espacial de las letrinas de conejo (*Oryctolagus cuniculus*) en el monte de Valdelatas (Madrid). Galemys 15:157–165

Moreno S, Villafuerte R (1995) Traditional management of scrubland for the conservation of rabbits *Oryctolagus cuniculus* and their predators in Doñana National Park, Spain. Biol Conserv 73:81–85

Moreno S, Beltrán JF, Cotilla I, Kuffner B, Laffite R, Jordán G, Ayala J, Quintero C, Jiménez A, Castro F, Cabezas S, Villafuerte R (2007) Long-term decline of the European wild rabbit (*Oryctolagus cuniculus*) in south-western Spain. Wildl Res 34:652–658

Muñoz G (1960) Anverso y reverso de la mixomatosis. Dirección General de Montes, Caza y Pesca Fluvial, Madrid

Murray DL, Roth JD, Ellsworth E, Wirsing AJ, Steury TD (2002) Estimating low-density snowshoe hare populations using fecal pellet counts. Can J Zool 80:771–781

Myers K, Parker BS, Dunsmore JD (1975) Changes in numbers of rabbits and their burrows in a subalpine environment in south-eastern New South Wales. Aust Wildl Res 2:121–133

Newey S, Bell M, Enthoven S, Thirgood S (2003) Can distance sampling and dung plots be used to assess the density of mountain hares *Lepus timidus*? Wildl Biol 9:185–192

Palomares F (2001) Comparison of 3 methods to estimate rabbit abundance in a Mediterranean environment. Wildl Soc Bull 29:578–585

Palomares F, Delibes M, Revilla E, Calzada J, Fedriani JM (2001) Spatial ecology of Iberian lynx and abundance of European rabbits in southwestern Spain. Wildl Monogr 148:1–36

Poole DW, Cowan DP, Smith GC (2003) Developing a census method based on sight counts to estimate rabbit (*Oryctolagus cuniculus*) numbers. Wildl Res 30:487–493

Putman RJ (1984) Facts from faeces. Mamm Rev 14:79–97

Ríos CA (2010) Los Planes Técnicos de Caza de Castilla-La Mancha y su aplicación en la gestión y conservación de las especies cinegéticas. Dissertation, Universidad de Castilla-La Mancha

StatSoft, Inc. (2001) STATISTICA (data analysis software system), version 6. Available at http://www.statsoft.com

Thomas L, Buckland ST, Rextad EA, Laake JL, Strindberg SS, Hedley SL, Bishop JRB, Marques TA, Burnham KP (2010) Distance software: design and analysis of distance sampling surveys for estimating population size. J Appl Ecol 47:5–14

Twigg LE, Lowe TJ, Gray GS, Martin GR, Wheeler AG, Barker W (1998) Spotlight counts, site fidelity and migration of European rabbit (*Oryctolagus cuniculus*). Wildl Res 25:113–122

Twigg LE, Gray GS, Massam MC, Lowe TJ, Kirkpatrick W, Bendotti G, Chester DR (2001) Evaluation of bait stations for control of urban rabbits. Wildl Res 28:299–310

Valverde JA (1967) Estructura de una comunidad Mediterránea de vertebrados terrestres. Monografías de la Estación Biológica de Doñana 1. Consejo Superior de Investigaciones Científicas, Madrid

Villafuerte R, Kufner MB, Delibes M, Moreno S (1993) Environmental factors influencing the seasonal daily activity of the European rabbit (*Oryctolagus cuniculus*) in a Mediterranean area. Mammalia 57:341–347

Villafuerte R, Calvete C, Blanco JC, Lucientes J (1995) Incidence of viral hemorrhagic disease in wild rabbit populations in Spain. Mammalia 59:651–659

Villafuerte R, Viñuela J, Blanco JC (1998) Extensive predator persecution caused by population crash in a game species: the case of red kites and rabbits in Spain. Biol Conserv 84:181–188

Ward AL, White PCL, Critchley CH (2004) Roe deer *Capreolus capreolus* behaviour affects density estimates from distance sampling surveys. Mamm Rev 34:315–319

Williams CJ, Parer I, Coman BJ, Burley J, Braysher ML (1995) Managing vertebrate pests: rabbits. Bureau of Rural Resources/ CSIRO Wildlife & Ecology, Australian Government Printing Service, Canberra

Williams D, Acevedo P, Gortázar C, Escudero MA, Labarta JL, Marco J, Villafuerte R (2007) Hunting for answers: rabbit (*Oryctolagus cuniculus*) population trends in northeastern Spain. Eur J Wildl Res 53:19–28

Wood DH (1988) Estimating rabbit density by counting dung pellets. Aust Wildl Res 15:665–671

Zar JH (1984) Biostatistical analysis. Prentice-Hall, New Jersey