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Reliability of methods used to estimate rodent pest densities in agricultural systems: the case of common vole (*Microtus arvalis*) in NW Spain

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Resumen

Uno de los aspectos más importantes a la hora de mejorar la gestión de plagas y reducir de forma eficaz los daños que causan en cultivos, minimizando a su vez los daños colaterales causados al medio ambiente, es disponer de herramientas de monitorización fiables. El topillo campesino es una de las plagas agrarias mas importantes en Castilla y León (NO de España). En el presente estudio se compararon las estimaciones de densidad del topillo campesino obtenidas mediante dos índices indirectos: el índice PSpS (Presence Signs per Square) previamente evaluado con la metodología de trampeo considerada mas fiable para estimar densidades de roedores, y el índice MI propuesto por el Ministerio de Agricultura, Alimentación y Medio Ambiente, recomendado como principal herramienta de monitorización de dicha especie, pero para el que no existe aparentemente ninguna evaluación de precisión o eficacia. En condiciones de baja abundancia de topillo campesino se observó que el índice MI estimaba densidades de topillos muy superiores a las proporcionadas por el índice PSpS (14 veces mayores en promedio), aunque ambos índices estaban altamente correlacionados entre sí. Estos resultados indican que el índice MI tiene una alta probabilidad de sobreestimar la abundancia de topillos en condiciones de densidad baja. En conclusión, las estimaciones de abundancia obtenidas con el índice MI deben ser consideradas como sobreestimaciones potenciales de la densidad real del topillo campesino, debiendo ser corregidas si se quiere mantener dicho índice como la principal herramienta para monitorizar las poblaciones de topillo campesino. Los resultados obtenidos refuerzan la importancia de evaluar la eficacia y precisión de este tipo de herramientas, para reducir el riesgo de realizar campañas de control innecesarias, minimizando así los costes económicos y ambientales asociados a las mismas.

Palabras clave: control de plagas, herramientas de monitorización, índices indirectos, protección de cultivos.

Abstract

For pest management strategies to effectively prevent crop damage while at the same time causing the least environmental collateral damage, reliable abundance monitoring tools are required. The common vole is a major agricultural pest in Castilla y León (NW Spain). In this study we compared common vole density estimates provided by two indirect indices, the Presence Signs per Square (PSpS) index, previously evaluated by a comparison with the trapping methodology most accepted to estimate rodent densities, and another one (MI), proposed by the Spanish Ministry of Agriculture Food and Environment, which is currently recommended as main monitoring tool for this pest, but that apparently has not been evaluated in a similar way. We found that at low vole abundance, MI generated much larger vole density estimates (on average 14 times) as compared to PSpS, although both indices were highly correlated. These results strongly support that abundance estimates provided by MI must be considered potential overestimates of real vole density and must be corrected accordingly in the future, if the method is to be kept as a main monitoring tool of vole density. Our results reinforce the importance of evaluating this kind of monitoring tools in different density scenarios to prevent the start or continuation of control campaigns when they are unnecessary, thus reducing their associated economic and environmental costs.

Keywords: crop protection, indirect indices, monitoring tools, pest management.

Introduction

Natural systems are inherently complex and obtaining accurate data that can be used to take appropriate management decisions is often a challenging task. This is particularly true for agricultural pest species, whose management (e.g. pest control) is often complex. For instance, accurately determining threshold densities for implementing specific management actions remains a key issue, and uncertainties may lead to mistakes during the decision making process (Ludwig *et al.* 1993). To address the risks and uncertainties of decision making under imperfect knowledge, different strategies have been developed like adaptive management or management strategies evaluation (Bunnefeld *et al.* 2011, Parkes *et al.* 2006).

In both types of management strategies, one of the most common uncertainties is the lack of adequate knowledge about the population dynamics of the species under management. This is of crucial importance in order to decide the number of individuals in a given population that could be extracted at a given time without compromising sustainable future exploitation (Holland 2010, Milner-Gulland 2011) or which control measures are effective and when they should be applied in a pest management scenario (Parkes *et al.* 2006). Thus, an inaccurate knowledge of population abundance and trends would increase the chances of making erroneous management decisions in pest control programs, inducing undesired actions either by the application of unnecessary control treatments, or by not applying a treatment when required (Gent *et al.* 2011).

An adequate knowledge of population dynamics requires a reliable method to measure population size or abundance (Krebs 1999, Witmer 2005). In the case of pest management, the method should also be simple enough to be applied at a large scale and should allow a fast monitoring (Delattre *et al.* 1990, Jareño *et al.* 2014). As the knowledge of the studied species increases it becomes important to review the reliability of the methods used to measure abundance (Lisicka *et al.* 2007, Embleton & Petrovskaya 2013), in order to improve decision-making.

The common vole (*Microtus arvalis*) colonized the agricultural plains of Castilla y León (NW Spain) since the end of 1970s, and regular population outbreaks have since caused important agricultural damages (Luque-Larena *et al.* 2013). During a common vole outbreak in 2007-08, an activity index was used by the Regional Government to estimate vole density and guiding implementation of pest control actions, and this method has been subsequently advocated as the main large-scale monitoring tool for common vole populations in Spain (Dirección General de Recursos Agrícolas y Ganaderos, 2009). However, to our knowledge, the efficacy and accuracy of this method had not been reliably tested in the study area, and could have potentially lead to erroneous management decisions, such as applying large-scale chemical treatments when unnecessary, with important environmental side-effects caused by massive use of rodenticides that affected to non-target species too (Olea *et al.* 2009, Jubete 2011, Sánchez-Barbudo *et al.* 2012). Our aim here is to assess whether the monitoring tool proposed by the Ministry of Agriculture, apparently following the experience by Castilla y León regional government mentioned above, is accurate enough for estimating common vole density under conditions of low to mid vole abundance and thus, if it can be considered an adequate management tool allowing correct decisions about the density thresholds from which control programs should be started. For this purpose, we compared common vole abundance estimates derived from the index currently recommended by the Spanish Ministry of Agriculture with those obtained using another abundance estimation method recently tested in the same agricultural area, under similar conditions of vole abundance and on the same habitats (Jareño *et al.* 2014).

Material and methods

The study took place during August 2012. We compared two indirect methods of estimating common vole abundance in 60 plots, characterized by three different types of agricultural habitats (non irrigated alfalfa, fallows and cereal stubble; with a total of 20 plots per habitat type). The study plots (fields) were located in three villages of Castilla y León: Boada de Campos, San Martín de Valderaduey and Villalar de los Comuneros. The habitats used in this study were selected for their relevance for the common vole: alfalfa is the most favorable habitat for this species in the agricultural areas of Castilla y León, fallows represent the biggest areas of natural vegetation in agricultural areas (and thus a potential stable refuge free of agricultural perturbances) and cereals occupy most of the agricultural surface in the study area and are the crop where more damage claims are registered in outbreak years (Jareño *et al.* 2014, AGROSEGURO, pers. com.).

Presence Signs per Square (PSpS) method

This method estimates the number of vole Presence Signs per Square (PSpS from now on), a method recently developed and tested in this region and study area (Jareño *et al.* 2014). This index is based on the presence/absence of vole activity signs (fresh dropping and/or clippings) within 15 squares (each 30x30 cm), at locations randomly selected every 5 m along a transect line located on the edge of each sampled agricultural parcel (5 sampled squares in the field margin, a habitat highly favorable for this species see Jareño *et al.* 2014) and along another perpendicular transect line, from the edge towards the centre of the field (10 sampled squares inside the field; the two lines forming a "T" design, see details in Jareño et al., 2014). This index showed a good correlation with vole densities estimated by trapping (capture-marking-recapture, CMR density estimates, a method considered as reliable to estimate rodent densities, Krebs 1999), so it can be considered a reliable sampling method to estimate vole densities, at least for densities up to 100 voles/ha (see Jareño et al. 2014). The regression function between the index and density estimates obtained from trapping was used to estimate vole density (number of voles/ha) at the plot level from PSpS index, using the formula: voles/ha= $\exp[(1.79 \pm 0.39 + (3.34 \pm 0.86) \times \text{PSpS})] - 1$, obtaining the variable PSpS_d (Jareño *et al.* 2014). Thus, this index provided density estimates calibrated with the best know method to estimate small mammal abundances (CMR trapping) in the same habitats, study areas and under similar conditions of vole abundance than those found in this study (<200 voles/ha).

Ministry of Agriculture (MI) method

This method, proposed by the Spanish Ministry of Agriculture Food and Environment (hereafter referred to as MI method) is based on the sampling of 33 rectangles, each 3 meters long by 1.5 meters wide along a 100-m transect line going from the edge to the center of the sampled plot. It consists of looking for active vole burrows (a burrow being active when recent activity signs in the form of clipping or droppings are found near burrow external holes within each rectangle). The vole abundance estimates (voles/ha) are obtained by multiplying the number of rectangles with active burrows by 40 (Dirección General de Recursos Agrícolas y Ganaderos 2009). This method seems to be based on an indirect index developed elsewhere (Delattre *et al.* 1990), with the recommendations to be used only in areas where vegetation height does not exceed 15 cm. We calculated abundance estimates using the MI method and squares sampled inside the field only, as field edges in our study area often have dense vegetation taller than 15 cm, and because in the reference work for this method sampling in filed edges is not considered.
For each of the 60 study plots, we obtained estimates for both indices consecutively, over the same central axis.

Statistical analyses

We used Spearman's rank correlations to test for associations between abundances estimates derived from the PSpS method (standardized before analysis by dividing them by the number of quadrats used, 15) and the MI method (standardized before analysis by dividing it by the total number of rectangles, 33). Correlations were tested for all habitats pooled, as well as for each habitat separately. Paired Wilcoxon signed-rank tests were used to determine whether the density estimates provided by each index (being PSpS_d the density estimate for the PSpS method and MI_d the density estimate for the MI method) were significantly different. We also performed a linear regression between PSpS_d and MI_d to obtain a new density estimate for the index MI; we used linear models since the relationship between both densities was apparently linear (Fig. 1). All statistical analyses were done using R 2.14.1 (R Development Core Team, 2008).

Results

We found that abundance estimates derived from the presence of activity signs with both methods (MI and PSpS) were highly and significantly correlated (Table 1).

When we analyzed the relationship between PSpS and MI in each habitat separately, we found that in alfalfas and especially in fallows PSpS and MI had a good correlation between them, but they were not significantly correlated in stubble (Table 1). Then we analyzed mean densities estimated

Table 1. Spearman correlations between the ministry index (MI), and presence signs index (PSpS), in all habitats (All, n=60), alfalfa (n=20), fallow (n=20) and stubble (n=20). Significant correlation coefficients are highlighted in bold (p<0.1; * p <0.05; ** p<0.01; *** p<0.001).

	Habitat	Index PSpS
Index MI	All	0.76 ***
	Alfalfa	0.70 ***
	Fallow	0.87 ***
	Stubble	0.29

by both methods, and found marked differences between the estimates derived from the MI and PSpS methods. Average vole densities estimated by the MI index were 14 times higher than those obtained using the PSpS index (PSpS_d vs. MI_d, V= 1554 p= 2.5E-06, Fig. 2). When we analyzed these differences in each habitat separately we also found higher mean densities estimates for MI_d than for PSpS_d in all habitats (Fig. 1), although the differences were significant in fallow (V= 182, p= 4.2E-03) and alfalfa (V= 204, p= 2.3E-04), but not in stubble (V= 119, p= 0.61).

Since the densities estimates provided by both indices were highly correlated, and apparently had a linear relationship between them (Fig. 2), we used a linear model to test if PSpS_d could be obtained from MI_d, the results were significant (R^2= 62.95%, $F_{1,58}$= 98.53, p= 4.1E-14):

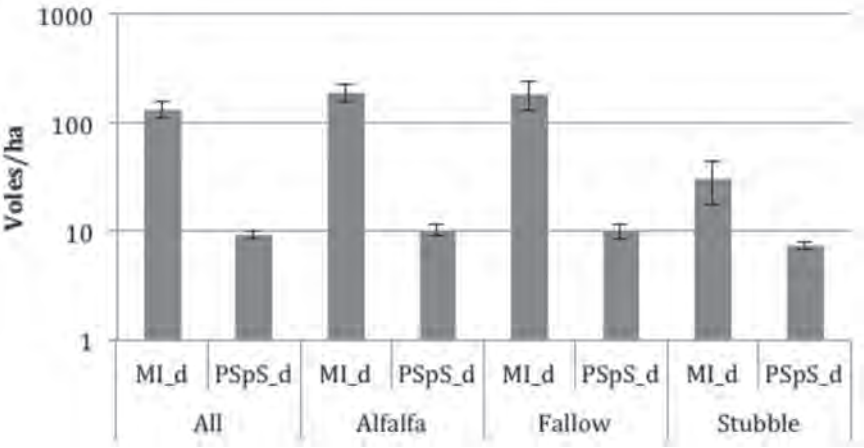

Figure 1. Common vole (*Microtus arvalis*) mean abundance estimates produced by each indirect activity index; PSpS_d (density estimate for the PSpS index), and MI_d (density estimate for the MI index), for all habitats (All, n=60) and for each individual habitat (Alfalfa, Fallow and Stubble, each one with n=20). We include standard error on the top of bars.

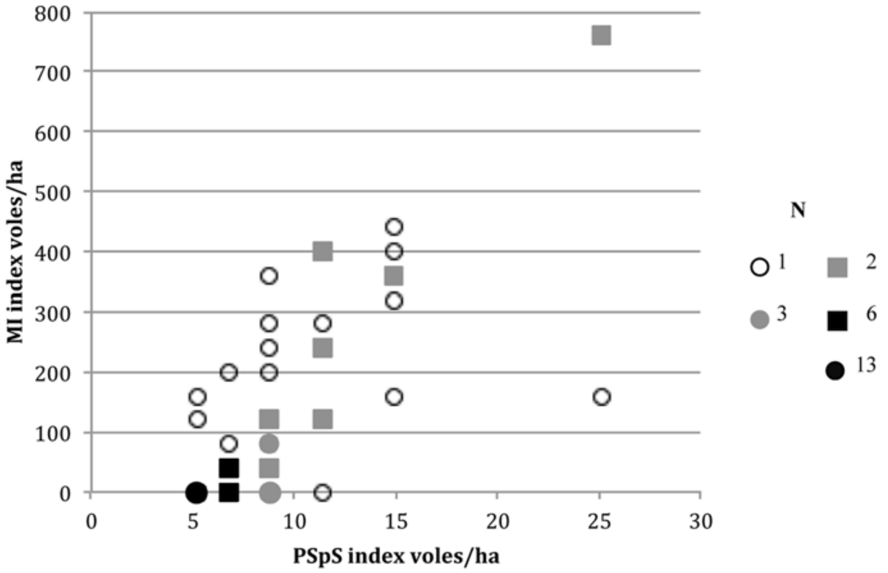

Figure 2. Dispersion graphic for the densities estimates obtained from MIand PSpS (MI_d and PSpS_d respectively). The different colors and shapes indicate the number of data points.

$$PSpS\_d = (6.4 \pm 0.47) + (0.021 \pm 0.002)*MI\_d.$$

Thus a direct transformation from MI to MI_d would be:

$$MI\_d_{PSpS\_d} = (6.4 \pm 0.47) + (0.84 \pm 0.08)*MI$$

Discussion

The common vole density estimates obtained with the two activity signs methods (PSpS and MI) were highly correlated, what supports the idea that the relative abundances of common vole in agricultural fields estimated by both methods are similar. This could be expected "a priori", as both methods are based on recording similar activity signs to estimate vole abundance. However, the density estimates derived from each method were markedly different, as the MI method provided values much higher than those obtained using the PSpS method (as much as18 times higher; Fig. 1). The density estimates obtained by PSpS have been found to be reliable at medium and low vole abundance (maximal densities of 126 voles/ha, a relatively high value, but still far form maximum recorded density in optimal agrarian habitat, >1000 voles/ha) by comparing them with CMR trapping results, but the reliability of this index at higher densities is doubtful (Jareño *et al.* 2014). However, during our study, vole density was extremely low, with estimated mean densities under 10 voles/ha following PSpS results. Given that vole densities estimated from PSpS were found to be reliable when vole densities were lower than 100 voles/ha (Jareño *et al.* 2014), and were obtained in the same study areas and habitats, the estimates derived by PSpS method can be considered reliable for the densities found in this study. Furthermore, trapping data obtained in the same areas one month before also indicated a very low vole density (3.18 voles per 100 traps; data from ongoing long-term monitoring data, J.J. Luque, unpub. data).

It could be argued that density estimates derived from PSpS index could be underestimating real population density, because the method was calibrated with a CMR methodology that may tend to underestimate real population sizes in small mammals, although available information about reliability of these estimates is scarce (Hilborn *et al.* 1976, Krebs *et al.* 2011). In this case, MI would not overestimate so much vole density, because CMR method, and consequently PSpS index, would produce density underestimates. However, recent modeling exercises indicate, on the contrary, that when all assumptions of CMR models are met in virtual populations (something that rarely happens completely in real wild populations) CMR estimates may indeed largely overestimate real population densities (Rees *et al.* 2011). In any case, field studies evaluating reliability of vole population densities produced by CMR methods have reported density underestimations usually between 10% and 30% of real densities, rarely higher (Hilborn *et al.* 1976, Peterjohn *et al.* 1981, Manning *et al.* 1995, Parmenter *et al.* 2003, Krebs *et al.* 2011). In contrast, common vole densities estimated by MI index were more than 1000% higher than density estimates produced by PSsP index. Thus, even assuming that PSsP index would underestimate real vole densities by 100% (an amount higher than the largest underestimate we have found in literature for voles, the 75% reported by Parmenter *et al.* 2003), MI index would still produce vole density estimates 8-10 times higher than "real" densities.

Thus, the MI method appears to largely overestimate vole populations, at least under conditions of low vole abundance, most probably due to the equation used to estimate density (finding just one active vole burrow in a plot would produce an estimate of 40 voles/ha, a density considerably high, and the MI index due to its use of rectangles of 4.5 m^2, as well as the higher number of rectangles sampled is probably more sensitive than PSpS to detect variations of vole activity at low densities). No information about how this formula has been produced has been provided by the Ministry, but it is different to that provided by the original work on which this monitoring method has presumably been based (Delattre *et al.* 1990, work on french pastures comparing line trapping data and faeces index). Perhaps the formula was generated from data obtained during the 2007 vole outbreak, under conditions of very high vole abundance (and this would suppose that the formula used to estimate densities from this index could vary depending on vole abundance as detected for the case of PSpS (Jareño *et al.* 2014). In any case, it seems clear that this formula overestimates vole densities under conditions of low vole abundance, so, based on the results presented above, we can provide a correction for the density estimate of MI (6.4 + 0.84×MI instead of 40 × MI) for its possible future consideration in monitoring programs using this sampling method. Nonetheless this correction should be taken with caution as it has been developed during a period of extremely low vole abundance, thus its usefulness could be limited to low vole abundance (not higher than 15 voles/ha, see Fig. 2). Additional work under conditions of higher vole densities is necessary to definitely correct vole density estimates provided by MI index (as for PSpS, see Jareño *et al.* 2014). Otherwise, the MI seems to perform at least as well as PSpS in terms of detecting abundance variation across different agricultural fields or crops, so if the problem of overestimation apparently induced by that equation is corrected, probably it can be considered an index at least as useful as PSpS may be.

Given that a monitoring program is usually aimed to detect the threshold densities when decision making is necessary, this potential overestimation of vole density under conditions of low vole abundance induced by the official monitoring method could lead to the application of unnecessary control measures before vole density has reached a problematic level. In our case, MI would have estimated an average density of 200 voles/ha, which would be perceived as a serious threat that could cause damage to crops (Dirección General de Recursos Agrícolas y Ganaderos 2009), while PSpS would provide a density estimate closer to 10-20 voles/ha (20-40 considering potential underestimation of real density that his index could have, as described above), which can be considered non-problematic. Alternatively, the acting threshold should be different depending of which method is used, or density calculations completely discarded, just using relative vole abundance known to be a potential problem. Since, to our knowledge, for the case of vole populations in Spain the threshold to act has not been accurately identified, the discrepancy in abundance estimates between methods is potentially even more risky, as different estimates and thresholds may lead to take incorrect measures. For example, some Spanish technical works have indicated that control campaigns should be undertaken when vole populations exceed 50 voles/ha in winter (Arenaz 2006), a treshold that would be estimated by the MI index with just 2 rectangles with activity, an activity easlily found in favourable habitats like alfalfas, even under conditions of general low vole abundance, and which could prompt unecessary control measures if alternative information about acting tressholds is lacking. Under these circumstances it would be easy to start unnecessary control campaigns, or to continue campaigns that have already been successful, thus extending their usually high economical cost (Jacob & Tkadlec 2010), as well as the environmental damage associated with large-scale use of rodenticides. In fact, the use of this monitoring system probably providing overestimated vole densities could explain why during the 2007-2008 vole outbreak, control campaigns based on rodenticide use were deployed when vole density had already declined to non-risky levels (Olea *et al.* 2009, Jubete 2011).

When we considered each habitat separately, we found that MI and PSpS were significantly correlated in fallows and alfalfas, but not in stubble. This was probably due to stubbles being a suboptimal habitat for voles in our study area (Jareño *et al.* 2014), thus in a year with low vole abundance there is practically no vole presence inside the field, being restricted mostly to edges.

Finally, both indices are not well suited to estimate vole abundances in field edges or road ditches (Delattre *et al.* 1990, Jareño *et al.* 2014), that are micro-habitats extremely important to maintain vole populations in the agricultural ecosystem of the study area (Jareño *et al.* 2014). Thus, additional research is required to improve current methodology to estimate vole abundance in field edges, something of critical importance in a monitoring program of this pest species.

This study emphasizes the importance for reviewing and improving pest monitoring tools, as proposed by adaptive management or management strategies evaluation, in order to correct imprecise abundance estimation methods and reduce uncertainty that may lead to inaccurate management decisions with their associated environmental and economic costs. Specific work at different levels of abundance to validate this kind of monitoring tools is strongly recommended.

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