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Agricultural land-use intensity and its effects on small mammals in the central region of Argentina

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Abstract Agriculture intensification is one of the main threats to biodiversity in agricultural systems. The effectiveness of conservation strategies in these systems depends mainly on the compromise between biodiversity conservation and agricultural land use. The aim of this study was to assess the effect of land-use intensity, characteristics of linear habitats (field borders) and their surrounding fields (landscape) on small mammal richness and abundance in agricultural systems of Argentina. In autumn 2009, we performed a removal sampling in 60 traplines located in the field borders of high or low land-use intensity regions. In traplines under high land-use intensity we found seven species while under low land-use intensity we found ten. Characteristic species of grasslands and woodlands such as *Monodelphis dimidiata*, *Thylamys pallidior*, *Necomys lasiurus* and *Graomys griseoflavus* were only captured in traplines under low land-use intensity. Higher numbers of *Calomys musculus* species (habitat generalist) were observed under high land-use intensity while *Akodon azarae* and *Oxymycterus rufus* species, known as habitat specialists, were more frequently found in the low land-use intensity region. Border width and height, as well as land use of both sides of the border, were major variables for explaining small mammal abundances. Our results suggest that conservation of wide field borders with characteristics similar to

those of natural habitats would be crucial for sustainable management of Pampean agricultural systems which hold high richness and abundance of small mammal species.

Keywords Field borders · High land-use intensity · Low land-use intensity · Population abundance · Species richness

Introduction

Land-use activities have caused declines in biodiversity through habitat fragmentation, degradation of soil and water and overexploitation of native species (Pimm and Raven 2000). The conversion of natural landscapes in croplands and pastures constitutes one of the most worldwide land-use activities (Foley et al. 2005). This conversion introduces alterations of habitat quality and suitability, producing agricultural landscapes widely variable in their degree of spatial heterogeneity (Fahrig et al. 2011). More heterogeneous landscapes are characteristic of traditional farming systems where many different production cover types are interspersed with more natural ones. In contrast, intensive agricultural systems are characterized by only few crop types in large uniform fields (Sirami et al. 2007; Fahrig et al. 2011). The effects of agricultural intensification on biodiversity are observed at local scale due to an increase in the use of pesticides and fertilizers. At landscape scale, loss, degradation and/or subdivision of natural or semi-natural habitats could be found (Hole et al. 2005; Lindenmayer and Fischer 2006; Concepción et al. 2008; Firbank et al. 2008; Geiger et al. 2010). The responses of species to agricultural intensification depend on its biology, behaviour and habitat requirements. Thus, a given species can be more prone or more resilient to extinction in modified landscapes (Ims 1995; Lindenmayer and Fischer 2006; Didham 2010). The effects of agriculture intensification could

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vary with the degree of habitat specialization of species. Specialist species are more dependent on habitat quality, and they suffer more from habitat disturbance than generalists, which are able to use other habitats and resources (Filippi-Codaccioni et al. 2010; Fischer and Schröder 2014).

The effect of agricultural intensification on biodiversity has been investigated at local and landscape scales in different animal taxa (Donald et al. 2001; Bilenca et al. 2007; Medan et al. 2011; Fuentes-Montemayor et al. 2012). Among species groups that coexist in agricultural landscapes, small mammals are crucial due to their contribution to well-structured food webs (Salamolard et al. 2000; Michel et al. 2006; Baraibar et al. 2009), the consumption and dispersal of plant products (Carey et al. 1999) and mycorrhizal fungi (Maser et al. 1978) and the consumption and control of invertebrates (Elkinton et al. 1996). In conventionally managed European fields, Fischer et al. (2011) found that complex landscapes increased richness and abundances of small mammal species, indicating the importance of landscape complexity for small mammals under agricultural intensification.

In Argentina, agricultural systems comprise large arable fields and sparse linear habitat networks (Baldi et al., 2006; Poggio et al. 2010). In intensively managed agricultural landscapes, the maintenance of undisturbed linear habitat networks can attenuate the effects of agricultural intensification by providing suitable habitats for biodiversity conservation (Simone et al. 2010; Gomez et al. 2011; Coda et al. 2014; Coda et al. 2015). An increase in agriculture intensification affected small mammal diversity and abundance in the Pampean region (Medan et al., 2011), with habitat generalist species such as the Cricetidae rodent *Calomys laucha* (Fischer, 1814) and *Calomys musculinus* (Thomas, 1913) being favoured and habitat specialist species such as *Akodon azarae* (Fischer, 1829) being negatively influenced (Bilenca and Kravetz 1995; Cavia et al. 2005; Fraschina et al. 2012).

Córdoba province is located in the centre of Argentina and occupies about the 6 % of the country total territorial extension. The centre-south of this province corresponds to Espinal and Inland Pampa ecoregions and has undergone a marked transformation due to intensive agriculture and increasing livestock practices. Considering the impact of environmental changes produced by agricultural intensification on linear habitats as well as the need to identify the environmental variables associated with richness and abundance of small mammal, the development of biodiversity conservation strategies in fragmented landscapes of the central region of Argentina seems to be a fundamental issue. To cope with this aim, the objective of this study was to assess the effect of land-use intensity, characteristics of linear habitats (field borders) and their surrounding fields (landscape) on small mammal richness and abundance. We predicted that the effects of land-use

intensity on small mammal populations would vary with the degree of specialization of the species. We expected higher species richness and abundance of specialist species in linear habitats of low land-use intensity regions than of high land-use intensity regions.

Material and methods

Study area

The study was carried out in agricultural systems of the centre-south of Córdoba Province, Argentina. This region corresponds to a low elevation plain (600–900 m) with vegetation dominated by algarrobo (*Prosopis alba*, *Phyllostachys nigra*), accompanied by quebracho blanco (*Aspidosperma quebracho blanco*), mistol (*Ziziphus mistol*) and itín (*Portulaca kuntzie*). The vegetation, however, has undergone marked alterations as a result of agriculture and cattle farming. At present, the landscape consisted of a matrix of crop/pasture fields surrounded by field borders, railways and other types of linear habitats. Despite the influence of crop fields, borders have a homogeneous plant cover of about 85 % throughout the year and a high availability of seed in the soil (Priotto et al. 2002). Study area (85000 ha approximately) was located among La Invernada (32° 48' S, 64° 36' W), Alcira (32° 41' S, 64° 43' W), General Cabrera (32° 48' S, 63° 52' W), Washington (33° 52' S, 64° 41' W), General Paunero (33° 52' S, 65° 01' W) and Chaján (33° 33' S, 65° 00' W) localities. This area has different states in relation to land-use intensity. The centre of the province is included in a high land-use intensity region where cultivated hectares (mainly with soybean and corn) double pasture hectares for livestock and many of the linear habitats were removed to enlarge agricultural fields. The south-west of the province is considered a low land-use intensity region since the number of hectares for pastures and for crops is similar and due to the presence of dunes with native grasslands and forest patches of *Prosopis* spp. (Cisneros et al. 2008; Martínez et al. 2014).

In this region, the small mammal assemblage is mainly represented by the Cricetidae rodents *C. musculinus*, *Calomys venustus* (Thomas, 1894), *C. laucha*, *A. azarae*, *Akodon dolores* (Thomas, 1916), *Oxymycterus rufus* (Fischer, 1814) and *Oligoryzomys flavescens* (Waterhouse, 1837) (Simone et al. 2010). Rodent species were ranked from generalists to specialists considering species-specific habitat specialization, ranging from habitat generalist (species occur in almost all habitats within the agriculture landscape) to habitat specialist (species occur mainly in habitats with high vegetation cover): *C. musculinus*, *C. laucha*, *A. azarae*, *Oligoryzomys flavescens*, *C. venustus*, *A. dolores* and *Oxymycterus rufus* (Martínez et al. 2014).

Field work

We performed a removal sampling through the use of 60 traplines during three consecutive weeks in April 2009 (austral autumn) in the two regions: high land-use intensity and low land-use intensity. Autumn was chosen as the sampling season due to the fact that most species present their annual abundance peak in it (Simone et al. 2010, Gomez et al. 2011). Every sampling week, we set up 20 lines of 20 traps (similar to Sherman live traps) each in the field borders located between fields and secondary roads. In the region with high land-use intensity, a total of 40 traplines were randomly set up, and the remaining 20 traplines were randomly located in the low land-use intensity region (Fig. 1). Traps were baited with a mixture of peanut butter and cow fat. Contiguous traplines were separated by a distance of at least 1 km, traps within lines were separated by a distance of 10 m, and each line is

200 m long. Traps were active during four consecutive nights each week. Traplines were considered as replicates for statistical analyses. This trap design was set in order to obtain reliable data for a study of the genetic structure of rodent species.

All specimens collected were carried out to the laboratory for their identification and to perform the measurement of external features (weight and length of body and tail). Captured animals were also used for other studies that included DNA and skull morphometric analyses (Martínez et al. 2014), and they were treated in humane manner according to current Argentinean Laws (National Law 14346). Considering that the studied species have small body sizes, cervical dislocation was used as euthanasia method (Sikes and Gannon 2011). The abundance values of each species were obtained as the number of individuals in each trapline, and richness was calculated as the number of different captured species within each trapline.

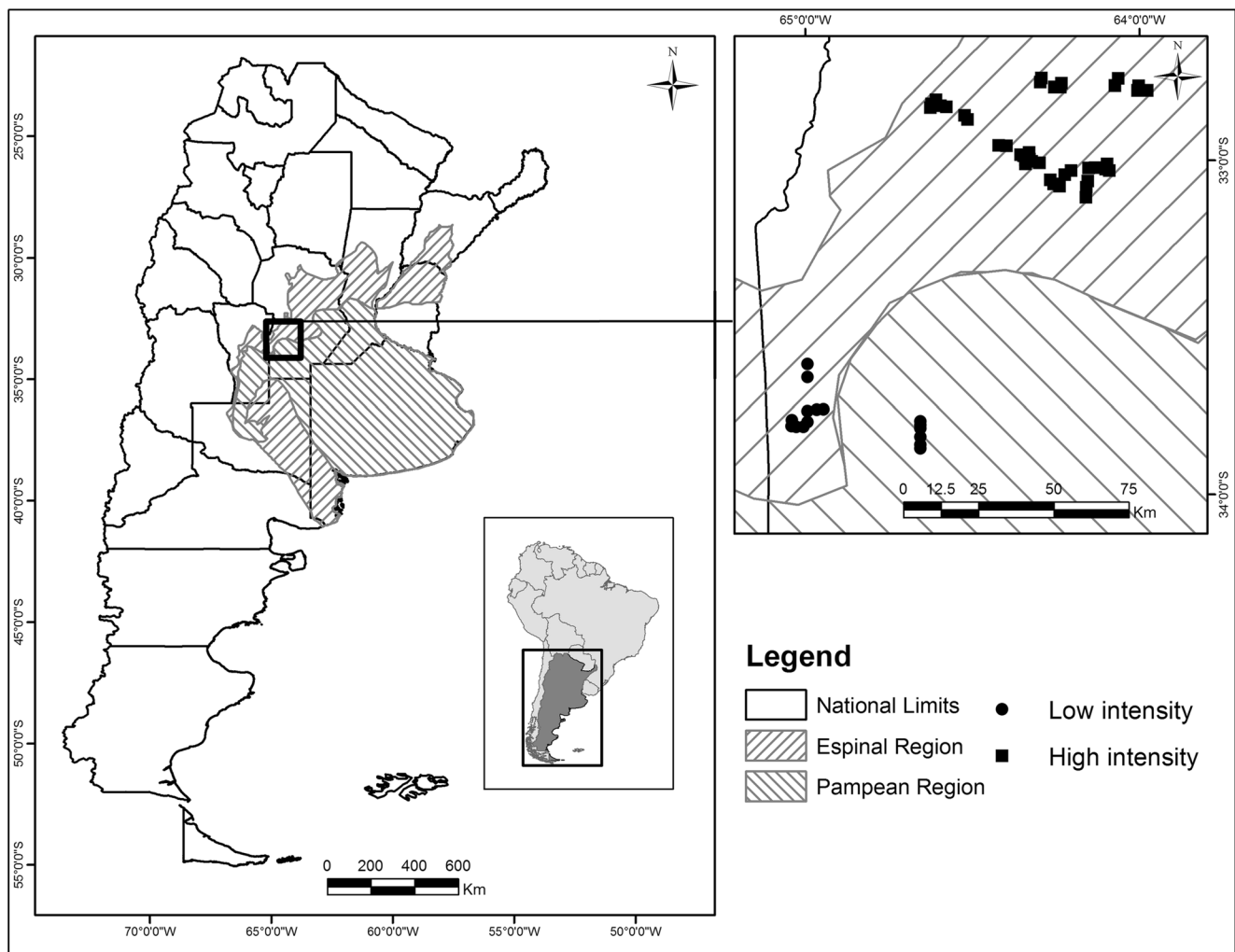


Fig. 1 Area of study, comprised within the agricultural systems of the centre-south of Córdoba province. *Squares*: traplines located in the high land-use intensity region; *circles*: traplines located in the low land-use intensity region

Habitat characteristics

Each line was classified according to the region where it belongs (high or low land-use intensity), and for each line, three local heterogeneity variables related to habitat fragmentation were obtained: field border width, border height and land use on both sides of the border. Border widths (m) were measured from field wire fences until secondary road, border heights (m) were measured from the road line up to the base of the wire fences, and land use was classified as follows: crop, stubble or pasture. In addition, a heterogeneity index was defined to assess landscape heterogeneity, and it was selected from a set of heterogeneity indexes previously defined for this study area (Martinez et al. 2014). Heterogeneity index was based on the quantification of the number of different classes included in an observational kernel (Baldi et al. 2006; Mora et al. 2010). To calculate this index, one size kernel of 6×6 pixels (180×180 m or 3.24 ha) was delimited around each trapline. Images were filtered from the different Landsat bands through a Sobel high-pass filter (Simone et al. 2012). Subsequently, the number of different classes (“border types”) surrounding each line within this kernel was registered. The higher the number of different borders, the greater the spatial heterogeneity obtained. Finally, two environmental variables were estimated for each line: Normalized by Difference Vegetation Index (NDVI) and land surface temperature (LST). These variables were obtained from a Landsat 5 TM satellite image (path/row: 229/83), acquired at the beginning of the fieldwork date. The image was processed using ENVI 3.5 (System Research), and a “stack image” was generated from the seven original Landsat bands and the NDVI. Each georeferenced trapline was uploaded in the stack image, and all variables values were exported. LST is defined as the thermal emission of the surface, including the top cover layer for areas with vegetation and other surfaces, such as bare soils, and it was estimated from Landsat TM band 6 (Schott and Volchok 1985; Markham and Barker 1986; Wukelic et al. 1989). NDVI constitutes a spectral vegetation index derived from the red/near-infrared reflectance ratio (Tucker et al. 1986; Chuvieco 1996).

Statistical analyses

The contribution of land-use intensity, local and landscape heterogeneity variables and environmental variables on small mammal richness and abundance of each species was evaluated using GLMs with Poisson distribution and the log-link function. The set of models contained each explanatory variable and the additive effects of two, three, four and five predictor variables. The null model with intercept only was also considered. Analyses were performed using the “MuMIn” package, implemented in R 2.15.2 software (R Development Core Team 2013). Models were compared using Akaike

information criterion corrected for small sample size (AICc), Δ AICc values and Akaike weights (Burnham and Anderson 2002). Weighted parameter estimates were produced for the variables using model averaging. Finally, hierarchical partitioning was conducted to determine the relative influence of the predictor variables (Mac Nally 2000) by using the package “hier.part” implemented in R 2.15.2 software (R Development Core Team 2013). Since model residuals may be spatially non-independent, we used Moran’s *I* tests to evaluate the spatial autocorrelation of the model residuals.

Results

A total of 634 individuals belonging to seven small mammal species were trapped in traplines located in the region with high land-use intensity, whereas in those located in the low land-use region, a total of 233 individuals belonging to ten small mammal species were captured (Table 1). *Necomys lasiurus* (Lund, 1840), *Graomys griseoflavus* (Waterhouse, 1837), *Monodelphis dimidiata* (Wagner, 1847) and *Thylamys pallidior* (Thomas, 1902) were only found in traplines placed in the region with low land-use intensity while *C. laucha* was only captured in traplines within the high land-use intensity region. The most frequently captured rodents belonged to *C. musculinus* in traplines under high land-use intensity and *A. azarae* in traplines of the remaining region (Table 1). Since for richness analyses, four models had similar statistical support (Δ AICc ≤ 2 ; Table 2), we used weighted parameter estimates for variables using model averaging. Finally, to explore the relative influence of the predictor variables, we used hierarchical partitioning. This analysis showed that LST (coefficient=0.037, SE=0.049, % I_y =33.421), land use on both sides of the border (pasture: coefficient=0.040, SE=0.208; stubble: coefficient=-0.082, SE=0.168; % I_y =25.758) and border height (coefficient=-0.072, SE=0.108; % I_y =21.349) were the main factors explaining richness. Richness was higher in the borders associated with pasture fields and was positively influenced by LST and negatively affected by border height (BH).

The analyses of abundances were performed by using the most abundant species: *C. musculinus*, *C. venustus*, *A. azarae* and *Oxymycterus rufus*, and the resulting best models are shown in Table 2. Two models, with similar statistical support, explained abundance of *C. musculinus* and *A. azarae*; thus, we applied model averaging and hierarchical partitioning. This analysis showed that land use on both sides of the border (pasture: coefficient=-1.094, SE=0.376; stubble: coefficient=0.368, SE=0.139; % I_y =36.498) and land-use intensity (low-intensity coefficient=-0.568, SE=0.212, % I_y =21.087) were the main factors explaining *C. musculinus* abundance. For explaining *A. azarae* abundance, the main factors were BH (coefficient=-0.416, SE=0.100, % I_y =32.619) and BW

Table 1 Species richness and abundances of each species by high ($n=40$) and low ($n=20$) land-use intensity regions

	High land-use intensity		Low land-use intensity	
	Mean (\pm SE)	Total	Mean (\pm SE)	Total
Richness	3.550 (\pm 0.932)	7	3.26 (\pm 1.73)	10
Species abundance	Mean (\pm SE)	Total (capture%)	Mean (\pm SE)	Total (capture%)
<i>Akodon azarae</i>	5.150 (\pm 3.906)	206 (32.492)	5.05 (\pm 4.54)	96 (41.2)
<i>A. dolores</i>	0.100 (\pm 0.496)	4 (0.631)	0.47 (\pm 1.72)	9 (3.9)
<i>Calomys musculinus</i>	7.575 (\pm 7.012)	303 (47.792)	2.37 (\pm 3.96)	45 (19.3)
<i>C. laucha</i>	0.275 (\pm 0.751)	11 (1.735)	–	–
<i>C. venustus</i>	1.375 (\pm 1.807)	55 (8.675)	0.11 (\pm 0.32)	2 (0.9)
<i>Oxymycterus rufus</i>	0.850 (\pm 1.099)	34 (5.363)	2.21 (\pm 2.35)	42 (18)
<i>Oligoryzomys flavescens</i>	0.525 (\pm 0.905)	21 (3.312)	0.32 (\pm 0.58)	6 (2.6)
<i>Necomys lasiurus</i>	–	–	1.16 (\pm 2.67)	22 (9.4)
<i>Graomys griseoflavus</i>	–	–	0.26 (\pm 0.93)	5 (2.1)
<i>Monodelphis dimidiata</i>	–	–	0.26 \pm 0.56	5 (2.1)
<i>Thylamys pallidior</i>	–	–	0.05 \pm 0.23	1 (0.4)
Total		634		233

Mean values with standard error and total values are given

(coefficient=0.021, SE=0.005, % $I_y=30.136$). This species was captured in large numbers in traplines of both regions, but capture percentages were higher in low land-use intensity region (41 % of total captures versus 32.5 % in high land-use intensity region). There were five models with similar statistical support for explaining *C. venustus* abundance; in this case, model averaging and hierarchical partitioning showed

that land-use intensity (low-intensity coefficient=-2.495, SE=0.735, % $I_y=70.904$) was the only factor with a significant effect. The abundance of *Oxymycterus rufus* was explained by four models in which land use on both sides of the border (pasture: coefficient=0.834, SE=0.426; stubble: coefficient=0.709, SE=0.350; % $I_y=23.602$) and land-use intensity (low-intensity coefficient=1.239, SE=0.371, % $I_y=$

Table 2 Generalized linear models (GLM) describing richness and species abundances as a function of land-use intensity, local and landscape heterogeneity variables and environmental variables

Models	AICc	Δ AICc	w_i
Richness (null model)	197.572	0	0.099
Richness (LST)	199.192	1.621	0.044
Richness (BH)	199.238	1.667	0.043
Richness (BW)	199.456	1.885	0.039
<i>Calomys musculinus</i> (BW + U + HI + NDVI + LUI)	424.433	0	0.224
<i>C. musculinus</i> (BW + U+ NDVI + LUI)	425.668	1.235	0.121
<i>Calomys venustus</i> (LUI)	164.832	0	0.144
<i>C. venustus</i> (LST + LUI)	165.347	0.515	0.111
<i>C. venustus</i> (NDVI + LUI)	166.605	1.773	0.059
<i>C. venustus</i> (HI + LUI)	166.792	1.960	0.054
<i>C. venustus</i> (BW + LUI)	166.829	1.997	0.053
<i>Akodon azarae</i> (BH + BW + HI + LST)	331.463	0	0.237
<i>A. azarae</i> (BH + BW + HI + LST + NDVI)	333.192	1.730	0.099
<i>Oxymycterus rufus</i> (BH + BW + HI + LUI)	164.460	0	0.183
<i>Oxymycterus rufus</i> (BW + U + HI + LUI)	165.785	1.325	0.094
<i>Oxymycterus rufus</i> (BH + BW + U + HI + LUI)	165.954	1.494	0.087
<i>Oxymycterus rufus</i> (BH + BW + HI + LST + LUI)	166.373	1.913	0.070

Land use on both sides of the border (U), border width (BW), border height (BH), land-use intensity (LUI), heterogeneity index (HI), Normalized by Difference Vegetation Index (NDVI) and land surface temperature (LST). Models are ordered by Δ AICc, only models with Δ AICc ≤ 2 . Weight (w_i)

28.177) were the most relevant variables. Thus, *C. musculus* and *C. venustus* species were more abundant in the region with high land-use intensity, whereas *Oxymycterus rufus* prevailed under low land-use intensity. *C. musculus* and *Oxymycterus rufus* abundances varied in relation to the field type associated to the borders. The highest abundance of *C. musculus* was registered in stubbles, and the lowest was detected in pasture fields. In contrast, *Oxymycterus rufus* was more abundant in pasture fields (Fig. 2a). *A. azarae* abundance showed a negative association with BH (Fig. 2b), and it was positively related to BW (Fig. 2c).

Discussion

In this study, we expected that linear habitats belonging to low land-use intensity region would have higher richness and abundance of specialist species than those of high land-use intensity one. The results showed that a higher number of species constituted the small mammal assemblage in a region with low land-use intensity when compared to a high land-use intensity one. However, average richness values were similar in both regions and land-use intensity did not explain richness, as the number of species per border was low with small variation between traplines located in different regions. The composition of small mammal assemblage reflected a clear trend of increasing relative abundance of the specialist species (*A. azarae* and *Oxymycterus rufus*) associated with low land-use intensity region and, on the other hand, an opposite downward trend of relative abundance of the more generalist species (*C. musculus*).

In the low land-use intensity region, there was a greater proportion of grasslands and xerophytic woodlands, characteristics of the Espinal ecoregion described by Brown et al. (2006). This fact possibly explains the presence of

M. dimidiata, *T. pallidior*, *N. lasiurus* and *G. griseoflavus* in traplines of this region, considered as distinctive species of the mentioned ecoregion. In concordance with results presented by Millán de la Peña et al. (2003) and Michel et al. (2006, 2007), we found that variations in species richness were higher among the field borders than between sampling regions. This fact may explain the lack of a clear response to land-use intensity.

Habitat specialization has been outlined as a key concept to predict the adaptive response of populations in heterogeneous and/or fluctuating environments (Levins 1968). In our study, the effects of landscape intensification on population abundances were species-specific, while specialist species may mostly be affected by environmental stressors such as habitat fragmentation or chemicals utilized in agroecosystems; generalist species may benefit from competition relaxation in these systems. Thus, specialist species as *Oxymycterus rufus* and *A. azarae* may suffer more from environmental stress and decline their population numbers with high levels of land-use intensity. *Oxymycterus rufus* is considered the most specialized within the four mentioned species due to its semi-fossorial habits, and the fact that it is exclusively captured in grasslands often associated to watercourses (Pardiñas et al. 2001). The higher representativeness of *A. azarae* in less-disturbed habitats has been observed by other researchers for the Pampean region corresponding to Buenos Aires province (González Fischer et al. 2012). On the other hand, generalist species as *C. musculus* may be benefited and increase its abundance under high land-use intensity. Different studies reported that agricultural intensification and expansion over Espinal and Pampean regions have favoured *C. musculus* species since it is a good settler of disturbed habitats as agricultural systems due to its habitat and trophic niche width (Bilenca and Kravetz 1995; Busch et al. 2000; Sommaro et al. 2010). Thus, the association between *Oxymycterus rufus*

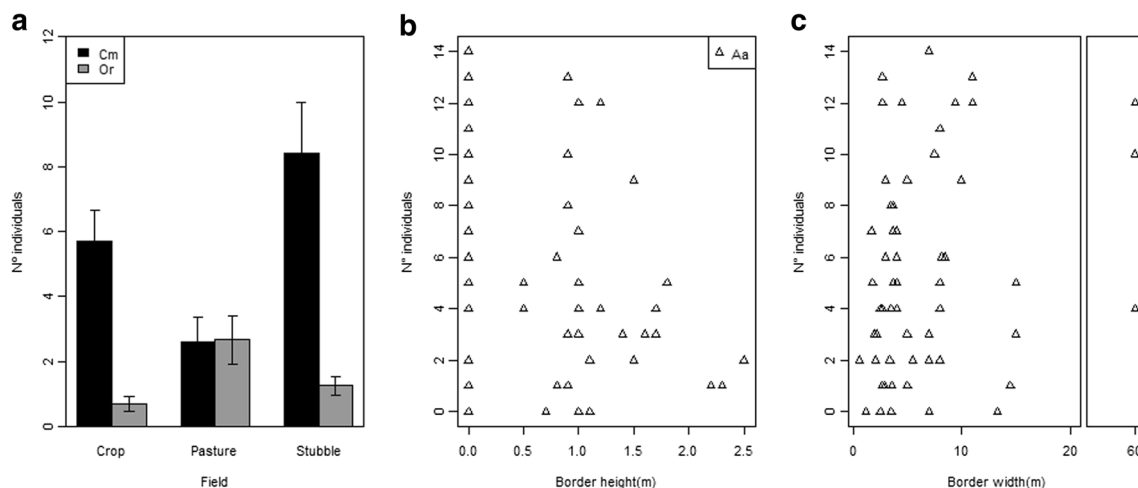


Fig. 2 a Number of individuals of *Calomys musculus* (Cm) and *Oxymycterus rufus* (Or) in relation to the land use on both sides of the border (mean ± SD); relationship between *Akodon azarae* (Aa) abundance and b border height and c border width

abundance and low land-use intensity as well as between *C. musculus* abundance and high land-use intensity may be explained by their habitat specialization grades. Agricultural expansion and intensification may have favoured dispersal and abundance of opportunistic species such as *C. musculus* in detriment of other small mammals during the last century (Pardiñas et al. 2000, 2004). The low capturability observed for the habitat specialist *C. venustus* within the low land-use intensity region could be related to its area of distribution, since the analyzed area was located in the boundaries of the southern geographic range of distribution described for this species (Polop 1996).

Additionally, local heterogeneity characteristics (border width and height and land use on both sides of the border) were related to abundances of habitat generalist and habitat specialist species. The relative importance of these variables varied among species. Borders width and height were important factors in determining the abundances of the habitat specialist *A. azarae*. It was also established that the widening of field borders increased its suitability as habitats for some small mammal species (Yletyinen and Norrdahl 2008). In this sense, the higher abundance values registered in wider borders would reflect the higher suitability of these linear habitats, as previously revealed in a study of movement distances of *A. azarae* (Gomez et al. 2011). The higher abundance registered in low borders may be related to the fact that they would facilitate movements of these small mammals, increasing colonization rates of these kinds of borders. Land use of both sides of the border was an important variable for explaining the abundance of species in the extremes of the specialization grade. Higher abundance of the most generalist species *C. musculus* was registered in the borders adjacent to unstable habitats as crops and stubbles, whereas the most specialist species *Oxymycterus rufus* was more abundant in more stable borders (with high vegetation cover throughout the year) associated to pastures. Abundance peaks of specialist species are registered in the field borders in autumn; thus, during this season, *C. musculus* would be displaced to more unstable borders (without vegetation cover) due to the dominance exerted by specialist species as *A. azarae* (Simone et al. 2010). Environmental variables such as NDVI and LST showed low incidence on species abundances, possibly due to the short variation range that they possessed during the sampling season. Other studies have revealed a relationship between NDVI and abundances of *A. azarae*; nonetheless, this relationship was observed in spring and summer (Simone et al. 2010) or during inter-annual dynamics (Andreo et al. 2009) when reductions in the plant cover may negatively affect the population size of this species.

In conclusion, the variable land-use intensity was a good predictor of species abundances and allowed a clear separation of species with different specialization grade. High land-use intensity regions seem to favour more opportunistic and

generalist species, while specialist species may be more frequently associated to less-disturbed and unmodified habitats. The characteristics related to linear habitats, such as border width and height and land use of both sides of the border, were relevant variables that allowed explanation of species abundances.

Due to the constant increase in world food and biofuel demand, the reversion of the current trend of agricultural intensification and expansion would be a utopia. Nevertheless, based on our data, we can support the idea that the implementation of some minor labours such as the preservation of wide field borders with characteristics similar to those of natural habitats would be crucial to ensure a sustainable management of agricultural systems. These linear habitats supporting high abundance of small mammal populations contribute to the maintenance of agroecosystem biodiversity.

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Compliance with ethical standards

Ethical approval All procedures performed in studies involving animals were in accordance with the current Argentinean Laws (National Law 14346, www.sarem.org).

Informed consent Informed consent was obtained from all individual participants included in the study.

Conflict of interest The authors declare they have no conflict of interests.

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