New contributions to the knowledge of the marsupials (Didelphimorphia: Didelphidae) of southern Santa Fe province, Argentina

Nuevos aportes al conocimiento de los marsupiales (Didelphimorphia: Didelphidae) del sur de la provincia de Santa Fe, Argentina

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ABSTRACT
In Argentina, the increasing demand of land for agricultural production, together with the effects of population growth, have been important factors in the alteration of the environments of the Pampas region. Marsupials represent a group of mammals with a long evolutionary history in this territory. Few species have been historically recorded in the province of Santa Fe, and most of these records come from the north and center of the province. In the present contribution we analyze the distribution patterns and habitat preference of didelphids present in the south of Santa Fe province. The study area corresponds to the Carcarañá river basin and the methodology used was: standardized line transects in search of signs of activity, camera traps and analysis of *Tyto furcata* pellets. In order to establish associations between the records of presence and the environmental variables studied (distance to roads and wire fences, ground cover, etc.), a geographic information system was generated. As a result, 3 species were recorded: *Didelphis albiventris*, *Lutreolina crassicaudata* and *Monodelphis dimidiata*. The principal component analysis showed that distance to urban centers, type of cover and vegetation heterogeneity are the variables that best explain the presence records of the species under study. Although the number of specimens is low, the results are relevant because these species have been little studied in Argentina and there are few or no records in the area. The transformation and degradation of the natural habitat due to the advance of the agricultural frontier is the main threat to these species.

Keywords: Argentina, ecology, mammals, Santa Fe.

RESUMEN
En Argentina, la creciente demanda de tierras para la producción agrícola, junto con los efectos del crecimiento demográfico, han sido factores importantes en la alteración de los ambientes de la región pampeana. Los marsupiales representan un grupo de mamíferos con una larga historia evolutiva en este territorio. Pocas especies han sido registradas históricamente en la provincia de Santa Fe, y la mayoría de estos registros provienen del

norte y centro de la provincia. En la presente contribución se analizan los patrones de distribución y preferencia de hábitat de los didélfidos presentes en el sur de la provincia de Santa Fe. El área de estudio corresponde a la cuenca del río Carcarañá y la metodología utilizada fue: transectas lineales estandarizadas en busca de signos de actividad, cámaras trampa y análisis de egagrópilas de Tyto furcata. A fin de establecer asociaciones entre los registros de presencia y las variables ambientales estudiadas (distancia a caminos y alambrados, cobertura del suelo, etc.), se generó un sistema de información geográfica. Como resultado, se registraron 3 especies: Didelphis albiventris, Lutreolina crassicaudata y Monodelphis dimidiata. El análisis de componentes principales mostró que la distancia a núcleos urbanos, el tipo de cobertura y la heterogeneidad de la vegetación son las variables que mejor explican los registros de presencia de las especies objeto de estudio. Si bien el número de ejemplares es bajo, los resultados son relevantes ya que estas especies han sido poco estudiadas en Argentina y existen escasos o nulos registros en la zona. La transformación y degradación del hábitat natural debido al avance de la frontera agrícola es la principal amenaza para estas especies.

Palabras clave: Argentina, ecología, mamíferos, Santa Fe.

1 INTRODUCTION

It is now considered that there is no place on the terrestrial biosphere without the human footprint, which has generated a point of no return in the loss of biodiversity in terrestrial and marine ecosystems (Hautier et al., 2015; Venter et al., 2016).

Wildlife is increasingly threatened by this development, which is pushing habitat fragmentation and loss to the limit (IUCN et al., 2008; Wallace et al., 2010; Ojeda et al., 2012). Mammals present different levels of sensitivity to this alteration, depending on their space requirements, their feeding needs and their behavior in the face of landscape changes resulting from anthropization (Rimoldi, 2015).

In response to these processes, man has attempted to measure, evaluate and lessen the impact of the causes of this crisis through theoretical and practical approaches (Primack et al., 2001).

Their study has become more relevant in recent years due to the accelerated process of modification that biological communities are undergoing as a result of human activities (Maclaurin & Sterelny, 2008 from Moreno et al., 2011).

In our country, the growing demand for land for agricultural production, together with the effects of population growth, have been important factors in the alteration of the environments of the Pampas region (Bilenca et al., 2012).

According to its conservation status, this region has been categorized by the World Wildlife Foundation (WWF) as an "endangered" area, and is assigned the highest

conservation priority due to its high biological diversity, its high degree of alteration and the scarce presence of natural protected areas (Bó et al., 2002).

In the south of the province of Santa Fe, the region presents a degree of fragmentation and land use that transformed its original aspect, becoming a plain where grain and oilseed crops predominate (Biasatti et al., 2019). In general, this region can be considered an area of high agricultural production, with a marked growth to the detriment of livestock and in an overexploitation of the soil with intensive practices of double annual crops, such as wheat and soybeans (Venencio, 2007).

Mammals present different levels of sensitivity to this alteration, depending on their space requirements, their feeding needs and their behavior in the face of landscape changes resulting from anthropization (Rimoldi, 2015).

Marsupials represent a group of mammals with a long evolutionary history in South America. In Argentina, the diversity amounts to 27 species and is mostly located in the northern ecosystems: the Yungas, the Paranaense Forest and the Chaco. Three species have been historically recorded for the province of Santa Fe, with most of these records coming from the north and center of the province. For the southern part of the province, marsupial records are restricted almost exclusively to the weasel (Didelphis albiventris Lund, 1840), which historically is one of the most characteristic mammals of this region. The objective of this contribution is to present new records of presence, distribution patterns and habitat preference for three species of marsupials.

2 MATERIALS AND METHOD

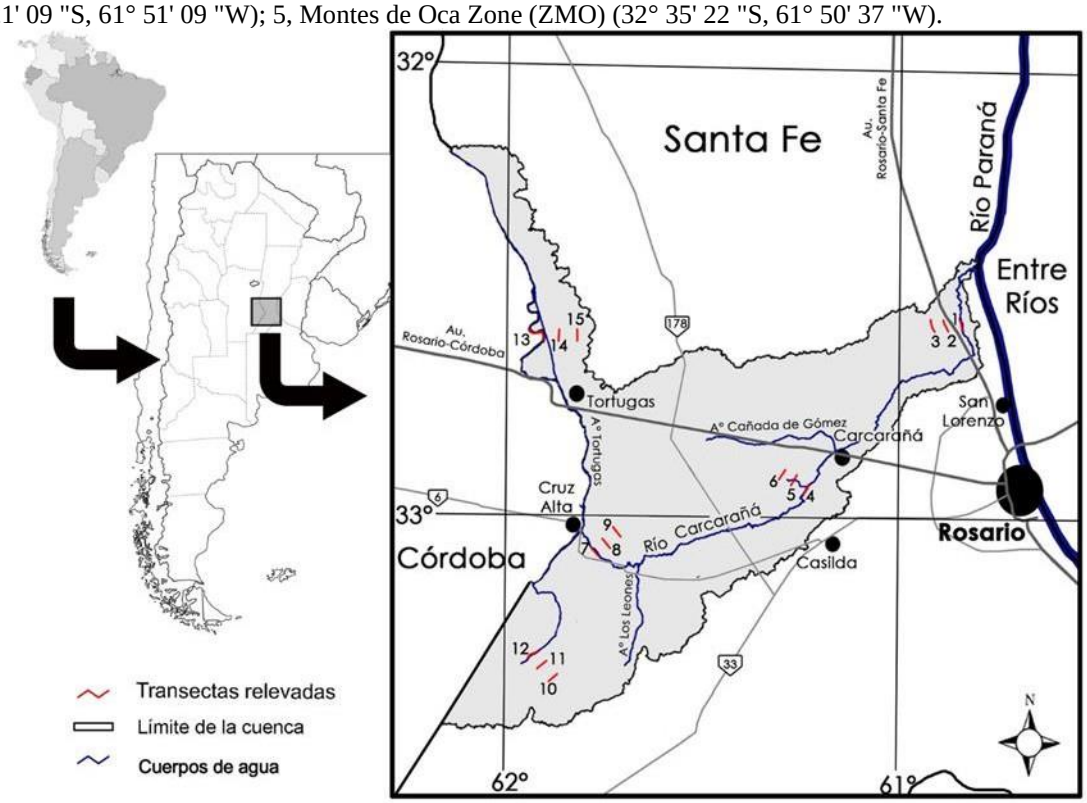
Study area. The area corresponds to the Carcarañá river basin in the province of Santa Fe, which is bordered to the west by the province of Córdoba and to the east by the Paraná river. It covers an area of 4,575 km^2 and is located between 32º26' and 33º20' S and 62º04' and 60º36' W. This area is part of the Belgrano and Iriondo departments, to the north of the Carcarañá River; and Caseros, San Lorenzo and General López to the south of it. Average annual temperatures range between 14 ºC and 20 ºC, with most of the precipitation concentrated in spring and summer. Annual precipitation in the region averages 970 mm and is distributed by isohyets oriented from north to south, with the maximum in the eastern region and the minimum to the west (Coronel and Sacchi, 2006).

Selection of sampling sites. Five sampling zones were established within the basin (distant from each other by 50 km), both on the banks of the Carcarañá river and its tributaries. Each zone for this work took the name of the nearest locality as a

reference. Thus, the names were established for each sampling zone as follows: 1-Oliveros Zone (ZO) (32°34'30 "S, 60°54'11 "W), 2-Berreta Zone (ZB) (32°53'48 "S, 61°16'24 "W), 3-Villa Eloísa

Zone (ZVE) (33°01'54 "S, 61°42'45 "W), 4-Zona Berabevú (ZBe) (33°21'09 "S, 61°51'09 "W) and 5-Zona Montes de Oca (ZMO) (32°35'22 "S, 61°50'37 "W) (Fig.1). These sectors were defined with the objective of obtaining a good representation of the total extension of the basin, taking into account that all the existing environmental units in the study area are represented. Based on this, three 3 km long transects were established in each study zone, taking into account that the first one is located on the river/tributary margin, while the remaining ones are parallel to it with a distance of 5 and 10 km respectively (Fig. 1). Thus, the methodology used was based on standardized line transects in search of signs of activity (Aranda, 2012).

Figure 1. Detail of the Carcarañá river basin in the province of Santa Fe (Argentina). Some reference localities and transect sites are indicated: 1, Oliveros Zone (32° 34' 30 "S, 60° 54' 11 "W); 2, Berreta Zone (32° 53' 48 "S, 61° 16' 24 "W); 3, Villa Eloísa Zone (33° 01' 54 "S, 61° 42' 45 "W); 4, Berabevú Zone (33° 21' 09 "S, 61° 51' 09 "W); 5, Montes de Oca Zone (ZMO) (32° 35' 22 "S, 61° 50' 37 "W).



Data collection. Field work was carried out during two consecutive years (2020-2021), seasonally (autumn, winter, spring and summer), with a periodicity of two days per month. The methodology used was the survey of indirect evidence (tracks and feces)

and the collection of information from direct evidence (sightings). Transect walks were conducted on foot, during daylight hours and at an average speed of one km/h, and were carried out during one day for each zone. A total of 80 days of field work and a total sampling effort of 360km were completed in the four environmental units proposed. In 2021, photo- trapping was incorporated as a support method for species confirmation.

Analysis of pellets. The analysis of the content of pellets, non-digestible material regurgitated by raptors in the form of pellets, is considered a useful tool to describe diet in terms of the structure of small mammal communities, the availability of prey according to the season of the year, as well as in the estimation of the relative abundances of prey populations in a given area and time. In this work we considered samples that could be attributed without doubt to *Tyto furcata*, possibly produced by a single specimen, a pair, or a pair with young in each environment under study. Between January and December 2020, pellets were collected monthly at previously established points in the sampling sites. One collection point was established for each sampling site. In all cases, the total number of pellets was collected leaving the perch clean, which ensured that, for each sampling, the material collected corresponded to the period between the previous and the current sampling. The jaws and skulls of the captured prey were compared with samples identified in osteological collections and specialized literature. Pairs of mandibles of the same species and/or skull were considered as one individual.

Spatial analysis. To establish associations between the records of didelphid presence with the environmental variables studied (distance to roads and fence edges, land cover, etc.), a geographic information system was generated to integrate information from various sources. Landsat TM 5 satellite images of March 28, 2020 were used to generate the maps and the approach scale was 1:100,000. The satellite images were obtained from the Data Distribution Center of the Instituto Nacional de Pesquisas Espaciais. The Gauss- Krüger coordinate reference system (Belt 5) defined by POSGAR WGS84 was used. The image was geometrically corrected with a first degree polynomial and 40 control points. The RMS (root mean square error) of the geometric correction process was 0.7 pixels. The programs used for spatial analysis were ArcGis 10.0 (ESRI, Redlands, CA, USA), IDRISI Selva GIS (Clark University, Worcester, MA, USA), Quantum GIS 1.7.4 and gvSIG 1.11.0.

Generation of land cover map. For the identification of vegetation units, an unsupervised classification with 15 classes was performed. Subsequently, each of the classes generated was assigned to the different vegetation and environment units based

on the spectral characteristics of the image. For the correct assignment, we used our own field information and high-resolution images available from Google Earth. These classes were then regrouped based on the data collected in the field. In this process, the different spectral classes could be reassigned to some of the different environmental units according to the existing bibliography for the south of the province of Santa Fe (croplands, xerophytic forests, halophytic communities and urban environments). For validation, a random draw of 100 points was made and these were corroborated by field information and sources of higher spatial resolution, obtaining an accuracy in the cover map of 90%.

3 RESULTS

Didelphis albiventris (Lund, 1840) (Fig. 2B). In total, 34 records of presence of *D. albiventris* were obtained, during the years 2020 - 2021, finding traces in all seasons, five (14.70%) in winter, seven (20.58%) in autumn, ten (29.41%) in spring and 12 (35.29%) in summer. Significant differences were found in the presence of traces of the species between seasons (Chi-square = 3.41, gl = 1, P-Value = 0.065) since the goodness-of-fit test to a uniform distribution yields P-Value less than 0.10, thus, the hypothesis of fit to a discrete uniform distribution can be rejected with a 90% confidence level. *D. albiventris* was recorded in all environmental units sampled, 15 (42.12%) evidences were found in xerophytic forest (6 in summer, 6 in spring, 2 in autumn and one in winter), eleven (32.35%) in croplands (two in autumn, three in winter, three in spring and three in summer), six (17.65%) in urban environments (two in summer, two in spring, one in autumn and one in winter) and two (5.88%) in halophilic communities of the sainfoin type (one in summer and one in autumn, with no records of presence in the sparse or impoverished salt meadows type.

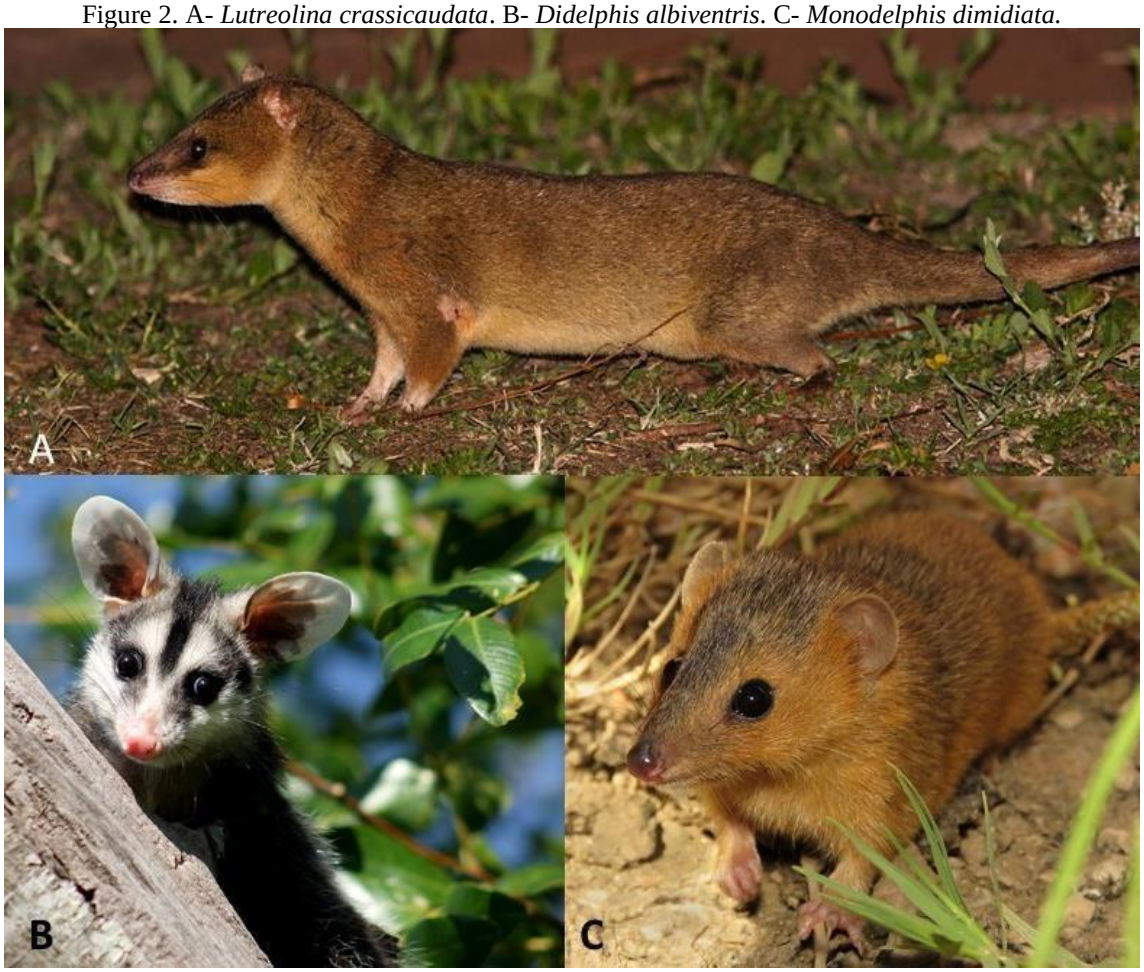
The total sampling effort for cropland was 240 km travelled, 48 km for xerophilous forest, 24 km for halophilous communities of the Espartillar type and 24 km for urban environments, giving a relative abundance of *D. albiventris* of 0.31 traces/Km for xerophilous forest, 0.25 traces/Km for urban environments, 0.08 traces/Km for halophilous communities of the Espartillar type and 0.04 traces/Km for cropland. With respect to the seasons of the year, no significant differences were obtained in the comparison between these and the environmental units (Chi-Square = 5.74, gl = 9, p = 0,7652). In relation to this, the analysis by environmental unit allowed us to establish that xerophytic forest was the only unit that showed significant differences between seasons for

this species (chi-square, $p<0.05$), with a greater presence in spring-summer. The records of *D. albiventris*, from the point of view of spatial analysis, were very heterogeneous. In this sense, the distance to water bodies varied from 0 to 9364m; however, 50% were recordedat less than 100m (96.51m) (RQ=4859.27). Similarly, the distance to roads varied from 0 to 8082.87m, with 50% of the data recorded below 1030m (RQ=1054.85). With respect to the distance to localities, the data ranged from 0 to 11511m. Fifty percent of the data were below 3182.42m (RQ=2398.24). All the records were at an average altitude of 65.35m (SD=30.01). Fifty percent of the records had an average slope grade of 1.04% (RQ=0.907). Principal component analysis was applied to the 5 environmental variables. The variables that had higher positive or negative values with higher loadings are indicated below, i.e., those variables that have a greater association with the principal components and, therefore, are of greater importance in explaining spatial variation. The first two components explain 82.2% of the variance of the 5 variables considered. Within component 1, which is responsible for 60.3% of the variance, is the effect of distance to localities and altitude. In component two, which is responsible for 21.9% of the variation, distance to water was associated with a high and negative value.

Lutreolina crassicaudata (Desmarest, 1804) (Fig. 2A): Five records were obtained for this species. Three from the analysis of *Tyto furcata* pellets. They occurred in summer, in the environmental unit of halophilic communities of the esparto grassland type. In this unit, 24 km of transects were covered, giving a relative abundance for the species of 0.04 tracks/km. From the point of view of spatial analysis, we can only mention that traces were found 8 km from the nearest road and at a distance of 11 km from Montes de Oca (nearest locality). They were 3m from the Tortugas stream with a height of 30 meters above sea level and a terrain slope of 0.741 meters.

Monodelphis dimidiata (Wagne, 1847) (Fig. 2C): Only one record in xerophytic forest was obtained from the analysis of *Tyto furcata* pellets.

Figure 2. A- *Lutreolina crassicaudata*. B- *Didelphis albiventris*. C- *Monodelphis dimidiata.*



4 DISCUSSION AND CONCLUSIONS

Didelphis albiventris: As observed by other authors (Parera, 2002; Pautasso, 2008; Teta and De Tomasso, 2009) this medium-sized mammal occupied all environments present in the basin, including those heavily modified by man (Teta and De Tomasso, 2009). Pautasso (2008), in reference to this, locates it for the Pampean region of Santa Fe in anthropized areas: landscaped areas with exotic and native woody species, with permanent and weekend human dwellings; farmhouses and periurban sites; bordering agricultural pastures. In this work, the highest relative abundance occurred in xerophytic forests. This can be explained by the preference of this species for areas close to water and with the presence of trees that it can climb in search of safety (Parera, 2002; Flores et al., 2006), conditions that are present in this environmental unit associated with the Carcarañá River.

The environmental unit called urban and peri-urban environments was presented as the second environmental unit in terms of relative abundance for this species, well above the results obtained for the other two environmental units (halophyte communities

and farmland). In this regard, Teta and De Tomasso (2009) note that this species, following the advance of urbanization and aided by its ecological versatility, has colonized areas where it was previously scarce, expanding its domain thanks to man, who indirectly offers it shelter and food in areas that were originally less favorable (Parera, 2002). In addition, it was possible to establish a differential use of the environments, particularly in the xerophytic forests where the highest records of presence occurred in the spring and summer seasons. This coincides with the reproductive season of the species (Parera, 2002), which could be interpreted as the use of these forested areas as a refuge, a fact already observed for the species in the province of Entre Ríos (Udrizar Sauthier et al., 2008) and Buenos Aires (Lucero et al., 2011). In addition to what has been mentioned so far, it is interesting to mention that *D. albiventris* is the species with the greatest amplitude in terms of the environmental variables studied. Its records were found on roads, in urban centers and on the banks of bodies of water, but also at distances exceeding 10,000 meters from any of the aforementioned variables. From the lowest zones (24 meters above sea level) to the highest zones of the basin (125 meters above sea level), there was evidence of their activity. All of the above reinforces the idea already proposed by other authors (Parera, 2002; Teta and de Tomasso, 2009; Wallace et al., 2010) that *D. albiventris* is one of the few medium-sized mammals characteristic of this ecosystem that has adapted, largely to the changes made by man.

Lutreolina crassicaudata: With a more restricted distribution than the previous species at the national level, its status in the province of Santa Fe is controversial. Some authors in general mappings (e.g. Parera, 2002; Canevari and Vaccaro, 2007) cite it for the entire provincial territory. Pautasso (2008) in the distribution maps of this marsupial indicates five records for southern Santa Fe, in which he does not include the Carcarañá river basin in his geonemics.

In the present investigation, five records were obtained in the environmental unit halophilic communities of the esparto grassland type. This site agrees with the distribution of the species proposed by Massoia et al. (2000) for southern Santa Fe. The conditions of this environmental unit, with *Spartina spartinae* associated with the Tortugas creek, are similar to those of the mentioned by Parera (2002) and Wallace (2010) about the preference of this species for open areas with grasslands associated with bodies of water. This has already been observed for the species in the Submeridional Lowlands (Pautasso, 2011), where all the records obtained by the author were associated with large areas of open grassland associated with water bodies.

S. *spartinae* sainfoin.

Monodelphis dimidiata: Although the number is low, this result is relevant because this species has been little studied in Argentina and there are few or no records in the area. The transformation and degradation of the natural habitat due to the advance of the agricultural frontier is the main threat to this species. In this work, the records were presented in a relict of xerophytic forest associated with the Carcarañá river, which agrees with what has been mentioned by several authors that in the Pampean region some populations have been fragmented, occupying isolated patches of native vegetation or with low intensity of use.

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