P-52

Unraveling *Leishmania* Reservoirs: An Exploration of Wild Micromammal Diversity in Southern and Central Areas of Tunisia

Sarah Chavez-Fisa[1*], Hichem Kacem[2,3], Xavier Roca-Geronès[1], Cristina Riera[1], Jordi Miquel[1,4], M. Magdalena Alcover[1], Roser Fisa[1]

1. Laboratori de Parasitologia, Departament de Biologia, Sanitat i Medi Ambient. Facultat de Farmàcia i Ciències de l'Alimentació. Universitat de Barcelona. 2. Laboratoire Écologie et Environement, Faculté des Sciences de Gabès, Université de Gabès Zrig, Tunisia. 3. Département des Sciences de la Vie, Faculté des Sciences de Sfax, Université de Sfax, Tunisia. 4. Institut de Recerca de la Biodiversitat (IRBio), Facultat de Biologia, Universitat de Barcelona.
*schavezf23@gmail.com

Background

Leishmaniasis is an endemic public health issue in Tunisia, attributed to the species *Leishmania major*, *L. infantum*, and *L. tropica* [1]. Micromammals play a pivotal role in the transmission of these diseases, underscoring the critical importance of identifying novel species as reservoir hosts for *Leishmania* parasites. The aim of this study is to enhance our understanding of the diverse array of wild animals serving as reservoirs for *Leishmania* spp. in the southern and central areas of Tunisia.

Materials and Methods

A total of 162 biological samples (ear=54, spleen=54, blood=54), were analyzed from 54 asymptomatic wild micromammals representing 10 different species (Table 1) from different localities of Southern and Central Tunisia. Enzyme-linked immunosorbent assay (ELISA) and Western blot (WB) techniques were conducted on blood samples for the detection of *Leishmania* antibodies [2]**.** WB was considered positive when immunoreactivity against the 14 and/or 16 kDa *L. infantum* fraction was observed and when ELISA is ≥20 U. For ear and spleen samples, after DNA extraction with the High Pure PCR Template Preparation Kit (Roche Applied Science), a Real-Time PCR targeting kinetoplast DNA was performed [3].

Results

All samples studied by Real-Time PCR were negative. Serology was positive in six out of the 54 micromammals, corresponding to a rate of 11.11% (CI 95%: 4.83% - 22.55%). Among the positive cases, five were positive in both ELISA and WB techniques, while one sample demonstrated positivity exclusively in the WB analysis. Four out of the six positive cases identified through serological analysis were attributed to *Ctenodactylus gundi,* one to *Atelerix algirus* and one to *Mus spretus* (Table 1). These positive results are in agreement with findings from other studies in southern and central areas of Tunisia supporting their identification as potential reservoirs for *Leishmania tropica*, *L. major*, and *L. infantum* in northern Africa and southern Europe [4,5,6].

Conclusion

Although a positive result through immunological techniques does not necessarily indicate an active infection, seroprevalence analyses could help advance our understanding of the leishmaniosis transmission dynamics in these regions. Furthermore, they may serve as indicators of prior contact with *Leishmania*, expanding our knowledge in this field.

Table 1. Micromammals analysed and serological results.

Specimens studied	Number specimens	WB		ELISA	
		Positive/total	Bands kDa	Positive/total	U
Atelerix algirus	2	1/2	14/16/18/24/36	1/2	72
Ctenodactylus gundi	4	4/4	14/24 14/24 14/24 14/24/36	4/4	69 50 63 182
Gerbillus campestris	13	0/13	Neg	0/13	<20
Gerbillus gerbillus	12	0/12	Neg	0/12	<20
Meriones shawi	1	0/1	Neg	0/1	<20
Mus spretus	12	1/12	16	0/12	<20
Petrosaltator rozeti	2	0/2	Neg	0/2	<20
Psammomys obesus	3	0/3	Neg	0/3	<20
Rattus norvegicus	1	0/1	Neg	0/1	<20
Rattus rattus	4	0/4	Neg	0/4	<20

WB: Western Blot; ELISA: enzyme-linked immunosorbent assay; U: units

Funding: This research received no external funding.

Conflict of interest: The authors declare no conflict of interest.

References:

1. Ben Othman S, Ghawar W, Chaouch M, et al., First detection of *Leishmania* DNA in *Psammomys obesus* and *Psammomys vexillaris*: Their potential involvement in the epidemiology of leishmaniasis in Tunisia. Infect Genet Evol. 2018; 59:7-15.
2. Riera C, Valladares JE, Gállego M, et al., Serological and parasitological follow-up in dogs experimentally infected with *Leishmania infantum* and treated with meglumine antimoniate. Vet Parasitol 1999; 84(1-2):33–47.
3. Martín-Ezquerra G, Fisa R, Riera C, et al., Role of *Leishmania* spp. Infestation in nondiagnostic cutaneous granulomatous lesions: report of a series of patients from a Western Mediterranean area. Br J Dermatol. 2009; 161 (2): 320–325.
4. Ghawar W, Bettaieb J, Salem S, et al., Natural infection of *Ctenodactylus gundi* by *Leishmania major* in Tunisia. Acta Trop. 2018; 177:89-93.
5. Alcover MM, Ribas A, Guillén MC, et al., Wild mammals as potential silent reservoirs of *Leishmania infantum* in a Mediterranean area. Prev Vet Med. 2020; 175:104874.
6. Chemkhi J, Souguir H, Ali IB, et al., Natural infection of Algerian hedgehog, *Atelerix algirus* (Lereboullet 1842) with *Leishmania* parasites in Tunisia. Acta Trop. 2015; 150:42-51.