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Short notes

Mastomys natalensis, *Cricetomys gambianus* and *Taterillus* sp. were found PCR positive for *Leishmania major* in Burkina Faso, West Africa

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ABSTRACT. Ouagadougou, the capital city of Burkina Faso, was recognized as a focus of zoonotic cutaneous leishmaniosis in April 2000. *Leishmania major* was the only strain isolated in this focus. We conducted a prospective study to detect *L. major* in rodents, animals which are described as reservoir of the parasite. Rodents were caught in five city areas from November 2005 to October 2006. Giemsa stained smears were realized from the cutaneous lesions when present after macroscopic examination of external lesions. The spleen of each rodent was sterilely removed and split into 3 parts for microscopic examination of smears, culture on NNN media and PCR, respectively. A total of 101 rodents belonging to 9 genera were trapped. All the direct examinations and cultures were negative. By using PCR of lesions and spleen samples, three animals were found infected by *L. major*: one out of 24 (4.2%) *Mastomys natalensis*; one out of 8 (12.5%) *Taterillus* sp. and one out of three *Cricetomys gambianus*. This is the first detection of *L. major* in rodent species in Burkina Faso. Further studies are needed to confirm their role as reservoirs of *L. major*.

Keywords: *Leishmania major*, *Mastomys natalensis*, *Taterillus* sp., *Cricetomys gambianus*, Burkina Faso

Introduction

Since 1996 many cases of zoonotic cutaneous leishmaniosis (ZCL) have been reported in Ouagadougou, capital of Burkina Faso, with a particular increase from 2003 to 2004 (2095 human cases) and during the period from 2005–2006 (1783 human cases) [1–3]. This increase was not rooted in any co-infection with the Human Immunodeficiency Virus [4]. Therefore, eco-epidemiological investigation has been carried out from 2006 in order to understand the increase of leishmaniosis prevalence. The entomological survey revealed that *Phlebotomus duboscqi* is the only species of genus *Phlebotomus* present (11.2% from 4676 trapped specimens), a well-known vector of ZCL in West Africa [5]. Concurrently, we conducted a parasitological investigation to isolate *L. major* from rodent species, potential reservoir of the parasite. This paper reports the results of this research.

Materials and Methods

Study sites. Ouagadougou, capital city of Burkina Faso, is situated between latitude 12°22'N

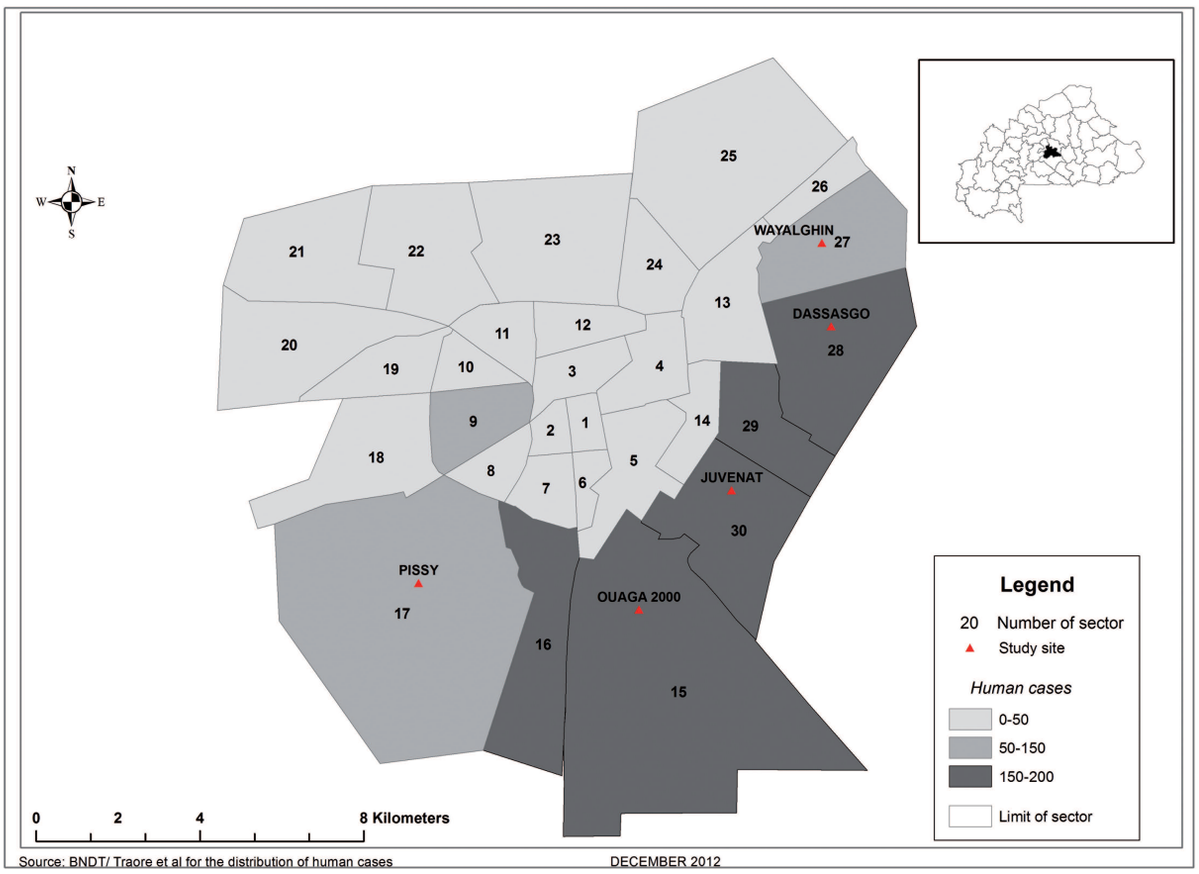

Figure 1. Map of Ouagadougou showing the location of trapping sites in different areas

and longitude 1°31'W, 300 meters above sea level. It had a population of around 1,086,000 inhabitants in 2006. The annual rainfall is 750 to 900 mm. The rainy season is between June and October, the cold and dry season is between November and January, and the hot and dry season is between February and May. Ouagadougou has a hydrographic network less developed. Artificial lakes located in peripheral areas provide a green belt with unusual vegetation in Sahel. There are also in peripheral areas spontaneous houses surrounded by vegetable crops. The trapping sites were selected in peripheral areas where the numbers of ZCL among inhabitants were the highest [1]. These areas also showed numerous rodent burrows and seemed to be good biotopes for the sandfly (high humidity of air, temperature around 26–30°C) (collecting sites in Figure 1). The site of Pissy, in the West, is in contact with the dam "*Boulmiougou*" and is bordered by rice fields. The sites of Dassasgho and Wayalgin in the East are wetland area with many mud houses. The site of Juvenat, always in the East of the city, is an area of vegetable crop. The site of "Ouaga2000" in the South is an area of food crop, which is currently undergoing urban reconstruction.

Rodents trapping. Rodents were trapped from November 2005 to October 2006 using Montpellier, Schermann and Chauvency live traps baited with bread and peanut butter. Areas were identified using a geographic information system [6]. Five trapping sorties were done each month for a sortie per site. In the selected transects, traps were placed around rodent burrows, from the sunset. Traps were gathered up early the following morning and brought back to the laboratory.

Organs removal and *Leishmania* diagnosis. Each rodent was killed according to the Guiding Principles for Biomedical Research involving Animals [7]. Cutaneous lesions were searched from head, ears, snout, paws and tail. When present, lesions were carefully disinfected with ethanol 70% and sterilely collected in a Petri dish. The animal was then sterilely dissected and the spleen removed and placed in a Petri dish. Each spleen sample was divided into three fragments. The first one served to detect amastigotes under a microscope through standard Giemsa staining method. The second fragment was cultured in supplemented NNN medium as described by Deniau et al. [8]. Culture

Table 1. Distribution of rodent lesions according to their topography

Topography of lesions	Number	%
Tail	12	40
Legs	6	20
Ears	5	16.7
Nose	4	13.3
Head	3	10
Total	30	100

was kept at 25°C, checked for promastigotes after 7 days. It was subcultured weekly for 5 weeks before being considered having no growth. Biopsies of lesions and the 3rd fragment of spleen were stored in 70% alcohol and sent to the Department of Parasitology at the University Paris XII for *Leishmania* DNA detection and species identification using PCR [9]. DNA was extracted using the Qiamp DNA Mini kit (Qiagen) and the diagnosis procedure included a real-time PCR assay targeted at the 18S rRNA gene, followed by a species identification step based on sequencing of the cytochrome b (cyt b) gene directly from the DNA extracted from the specimen.

Rodent's identification. Genera of rodents were determined morphologically at the Museum National d'Histoire Naturelle de Paris (France). It took into account the length of the body/tail, the space between the ears and the ear length. Then a panel of each genus was sent to Montpellier (France) and a PCR determination confirmed the species.

Results and Discussion

A total of 101 rodents were trapped from November 2005 to October 2006. The species were: *Gerbillicus* (*Tatera*) *guinea* (26), *Mastomys natalensis* (24), *Rattus rattus* (10), *Praomys daltoni* (9), *Taterillus* sp. (8), *Nannomys (Mus) musculus* (7), *Cricetomys gambianus* (3) and *Arvicanthis ansorgei* (2). Twelve rodents could not be identified. Their labels were detached and mixed, thus their origins could not be associated with any site.

The largest population of rodents came from sites located in South part (61) and West part (30) of the city. Observation made in an area of 10 m¸ have high density of 11 burrows in Ouaga2000 (1100 burrows per hectares) and 9 in Pissy (900 orifices per hectare). This abundance of burrows and rodent inhabitants in the south site provide favourable feeding opportunities for *P. duboscqi*, the probable main vector, which is present in Ouagadougou all the yearlong [5]. Thus, close dwelling association between rodents and sandflies maintains the continuous life cycle of *L. major*.

Lesions were observed in 30 rodents and mostly located on the tail (12) (Table 1). They could be caused by *Leishmania* infection. However it is important to notice that lesions could also result from fighting between animals. Smear examination and culture in NNN medium were negative for all rodents. However, one out of 24 *Mastomys natalensis*, one out of eight *Taterillus* sp. and one out of three *Cricetomys gambianus* were found PCR positive for *Leishmania major*. This underlines the interest of PCR as a screening and diagnostic tool. The three positive animals belong to genera already known to be a putative reservoir for *L. major*, i.e. *Mastomys*, *Taterillus* and *Cricetomys* genera. Our results are similar to those of Dedet et al. [10] in Senegal who found an infection rate with *L. major* of 4/5 (80%) for *Mastomys natalensis*, and to those of Githure et al. [11] in Kenya who found a 2/7 (28.6%) for *Taterillus emini*. *Cricetomys gambianus* has also been described as a reservoir for *L. aethiopica* in Ethiopia and Kenya [12]. The *Mastomys natalensis* and *Taterillus* sp. infected come from the same site in the South of the town (Ouaga 2000). They were captured in August 2006, the time we encountered many cases of LC from men in that site.

Mastomys natalensis (Muridae, Murinae) and *Taterillus* sp. (Muridae, Gerbilidae) were more captured than *Cricetomys gambianus*. However the size of sample is not sufficient for a dynamic analysis of the reservoir status. Studies with larger sample sizes would provide additional information for reservoir hosts. But our initial results support more direct control strategies against the endemic cutaneous leishmaniosis by targeting rodent populations in Ouagadougou.

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References

[1] Traoré K.S., Sawadogo N.O., Traoré A., Ouedraogo J.B., Traoré K.L., Guiguemdé T.R. 2001. Étude préliminaire de la leishmaniose cutanée dans la ville de Ouagadougou de 1996 – 1998 [Preliminary study of cutaneous leishmaniasis in the town of Ouagadougou from 1996 to 1998]. *Bulletin de la Société de Pathologie Exotique* 94: 52-55 (in French). http://www.pathexo.fr/documents/articles-bull/T94-1-2221.pdf

[2] Gouba A. 2007. Étude rétrospective de la leishmaniose cutanée dans la ville de Ouagadougou de 1999 – 2005. *Thèse de Pharmacie, Université de Ouagadougou* 74: 117 (in French).

[3] Sawadogo W. 2007. Etude rétrospective de la leishmaniose cutanée dans la ville de Ouagadougou de 2006 ŕ 2012: aspect épidémiologique. *Thèse de Pharmacie, Université de Ouagadougou* 196: 141 (in French).

[4] Guiguemdé R.T., Sawadogo O.S., Bories C., Traore K.L., Nezien D., Nikiema L., Pratlong F., Marty P., Houin R., Deniau M. 2003. *Leishmania major* and HIV co-infection in Burkina Faso. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 97: 168-169. doi:10.1016/S0035-9203(03)90109-X

[5] Sangaré I., Gantier J.C., Koalaga G., Deniau M., Ouari A., Guiguemdé R.T. 2009. Sandflies of the south part of Ouagadougou city, Burkina Faso. *Parasite* 16: 231-233. doi:10.1051/parasite/2009163231

[6] Openshaw S. 1996. Geographical information systems and tropical diseases. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 90: 337-339. doi:10.1016/S0035-9203(96)90500-3

[7] Council for International Organization of Medical Sciences (CIOMS). 1985. The international guiding principles for biomedical research involving animals. CIOMS, Geneva.

[8] Deniau M., Cańavate C., Faraut-Gambarelli F., Marty P. 2003. The biological diagnosis of leishmaniasis in HIV infected patients. *Annals of Tropical Medicine and Parasitology* 97: 115-133. doi:10.1179/000349803225002598

[9] Foulet F., Botterel F., Buffet P., Morizot G., Rivollet D., Deniau M., Pratlong F., Costa J.-M., Bretagne S. 2007. Detection and identification of *Leishmania* species from clinical specimens by using a real-time PCR assay and sequencing of the cytochrome *b* gene. *Journal of Clinical Microbiology* 45: 2110-2115. doi:10.1128/JCM.02555-06

[10] Dedet J.P., Hubert B., Desjeux P., Derouin F. 1982. Ecologie d'un foyer de leishmaniose cutanée dans la région de Thiès (Sénégal, Afrique de l'Ouest). 5. Infestation spontanée et rôle de réservoir de diverses espèces de rongeurs sauvages [Ecology of a focus of cutaneous leishmaniasis in the Thies region (Senegal, West Africa). 5. Spontaneous infestation and the role of the reservoir of various species of wild rodents]. *Bulletin de la Société de Pathologie Exotique* 75: 599-605 (in French). https://gallica.bnf.fr/ark:/12148/bpt6k9774954b/f645.image

[11] Githure J.I., Beach R.F., Lightner L.K. 1984. The isolation of *Leishmania major* from rodents in Baringo District, Kenya. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 78: 283. doi:10.1016/0035-9203(84)90305-5

[12] Le Blancq S.M., Schnur L.F., Peters W. 1986. *Leishmania* in the Old World. 1. The geographical and hostal distribution of *L. major* zymodemes. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 80: 99-112. doi:10.1016/0035-9203(86)90206-3

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