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Journal of Infection and Public Health xxx (2020) xxx–xxx

Contents lists available at ScienceDirect

Journal of Infection and Public Health

journal homepage: http://www.elsevier.com/locate/jiph

Short Report

First report of human *Leptospira santarosai* infection in French Guiana

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ARTICLE INFO

Article history:
Received 27 January 2020
Received in revised form 24 March 2020
Accepted 28 March 2020

Keywords:
Leptospira santarosai
Leptospirosis
Human
French Guiana

ABSTRACT

Leptospirosis is a zoonotic disease with worldwide distribution. In French Guiana, a French overseas department of South America, this bacterial infection is endemic with the increase of human cases since the last 5 years. Nevertheless, the epidemiological data on the circulating infecting strain remains scarce due to the lack of specific symptoms and the used diagnostic approaches. We report a severe case of leptospirosis in a 52-years-old male, working as a street cleaner, hospitalized at the Intensive Care Unit of city capital hospital of French Guiana because of hemodynamic, neurological, renal, and respiratory failure. At ICU admission, the patient was comatose, his temperature was 37.3 °C, heart rate 104 beats per minute, blood pressure 84/45 mm Hg, and oxygen saturation 95% while under mechanical ventilation. Retrospective exploration using molecular and serological approaches from the samples allowed reporting an infection by *Leptospira santarosai* serogroup Sejroe serovar Hardjo. To our knowledge, this is the first human case infected with this species in French Guiana. Through these analyses, this report provides new epidemiological information about the *Leptospira* strains circulating in French Guiana. In particular, this emphasizes the importance of accurate diagnosis to characterize with more precision the circulating *Leptospira* strains in this department.

Introduction

Leptospirosis is among the most common bacterial zoonotic diseases worldwide. The genus Leptospira includes at least 64 species grouped into four subclades: pathogens (P1 and P2) that are the causative agents of leptospirosis and saprophytes (S1 and S2) [1]. Currently, there are more than 250 pathogenic serovars. Human infection occurs through direct contact with animal reservoirs (mostly rodents) or, more frequently, water or soil contaminated with their urine [2,3]. After an incubation period of 2 to 30 days, the disease generally begins by fever, which is difficult to dissociate from other multiple causes of fever, particularly in tropical areas, and evolve toward chills, headache, myalgia, and more severe clinical signs [3]. Globally, the clinical course of leptospirosis is variable, from mostly mild and self-limited cases to some severe and potentially fatal cases, approximately 5 to 14 days after infection [3].

In French Guiana, a French overseas department located in the North-East of South America, leptospirosis is endemic although epidemiological data on this disease are seldom reported [4]. This can be explained by the absence of specific clinical signs to discriminate it from other tropical infectious diseases circulating in this region and a late diagnosis restricted to antibody detection using commercial assays. Nevertheless, the French National Reference Center (NRC), which monitors human leptospirosis in France and French overseas territories, reported a significant increase in the number of cases in French Guiana since 2014, from 6 to 18 cases per year between 1996 and 2012 to 92 cases in 2014 and 67 in 2018 [5]. Although no strain has been isolated in the last decades, the NRC reported the serogroup Icterohaemorrhagiae as the most prevalent among the few samples tested by microscopic agglutination test (MAT).

Case report

On March 2014, a 52-year-old man living in Cayenne (Capital city) working as a street cleaner presented at the Emergency Department of the Andrée Rosemon General Hospital Cayenne,

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https://doi.org/10.1016/j.jiph.2020.03.020

Table 1
Laboratory results at ICU admission and diagnostic investigations.

Biological parameter at ICU admission	Normal range	Value	Days after onset of symptoms	Diagnostic results		
				Real-time PCR (*Lfb1* gene)	ELISA IgM	MAT titer
Hematocrit, %	39–53	30	Day 7	Pathogenic *Leptospira* (*L. santarosai by sequence analysis of rrs and secY products)*	Equivocal 18 U/mL (*) (Panbio test)	Negative (***)
Leucocytes, x 10^9 cells/L	4–11	10.1				
Neutrophils, x 10^9 cells/L	1.4–7.7	8.5				
Lymphocytes, x 10^9 cells/L	1–4.8	0.6				
Plateled count, x 10^9 cells/L	150–400	136				
Serum protein, g/L	66–87	52				
Sodium, mmol/L	136–145	135				
Potassium, mmol/L	3.5–5.1	3				
Chlorides, mmol/L	98–107	96				
Bicarbonates, mmol/L	22–29	19	Day 55	Not done	Positive 98.4 U/mL (**) (Serion test)	Serogroup Sejroe Serovar Hardjo (***) (1/800)
Calcium, mmol/L	2.15–2.55	1.99				
Glycemia, mmol/L	3.9–6.4	8.9				
Urea nitrogen, mmol/L	2.8–8.1	20.1				
Creatinine, µmol/L	59–104	528				
Lactate, mmol/L	<2	4.9				
Total bilirubin, µmol/L	<21	87.2				
Direct bilirubin, µmol/L	<5	74.9				
Indirect bilirubin, µmol/L	<21	12.3				
Alanine aminotransferase, UI/L	<41	17	Day 78	Not done	Positive 68.1 U/mL(**) (Serion test)	Serogroup Sejroe Serovar Hardjo (***) (1/800)
Aspartate aminotransferase, UI/L	<40	36				
Gamma-glutamyltransferase, UI/L	<60	85				
Alkaline phosphatase, UI/L	40–130	103				

(Positivity thresholds: 19 U/mL* for the Panbio test, 15 U/mL** for the Serion test, and ≥1/100*** for the MAT titer).
UI/L: International Unit/liter.

French Guiana. He reported initial symptoms of dry cough, fever, and myalgia, diffuse headache, and hiccough over six days and a recent trip to Oyapock, an Amazonian river marking the border between French Guiana and the North Brazilian state of Amapa. When admitted to hospital, the patient presented confusion, stiff neck, a Glasgow Coma Score Scale of 11/15, and a temperature of 39.4 °C. Biological tests showed renal failure, leukocytosis, hyperlactacidemia, and slightly elevated bilirubin levels. The patient was rapidly transferred to the intensive care unit (ICU) because of hemodynamic, neurological, renal, and respiratory failure. At admission, the patient was comatose, his temperature was 37.3 °C, heart rate 104 beats per minute, blood pressure 84/45 mm Hg, and oxygen saturation 95% while under mechanical ventilation (FiO_2: 100%). The remainder of the clinical examination was normal. Results of laboratory showed renal impairment and an increase in inflammatory parameters evocative of infectious disease (Table 1). Cerebrospinal fluid (CSF) analysis showed a protein level of 0.29 G/L, glucose ratio of 0.66, lactate concentration of 2.4 mmol/L. CSF pleocytosis showed six white blood cells (WBC)/mm^3, Gram staining was negative, and microbiological culture was sterile. The patient's management included mechanical ventilation, norepinephrine fluid infusion, sedation, and broad spectrum anti-infectious drugs (ceftriaxone and acyclovir). The patient gradually recovered and was weaned from norepinephrine at day 8, from sedation at day 20, and from mechanical ventilation at day 30 after ICU admission. He fully recovered and left the unit at day 42 and discharged from the hospital on day 74. His clinical symptoms and recent outdoor activities on the Amazonian river led suspicion of a tropical infection.

At the ICU, various samples (serum, CSF, nasopharyngeal swab, bronco-alveolar lavage) were collected for microbiological investigations, including those consistent with the regional epidemiological context (Arboviruses, malaria, Q fever, Hantaviruses, Trypanosoma, etc.), according to the standard patient-care protocol. For specific leptospirosis diagnosis and follow-up, sequential serum samples collected at days 7, 55, and 78 after disease onset were tested for IgM serological analyses. The first sample showed equivocal IgM using an enzyme immuno-assay technique (Panbio, Australia). The two other samples collected respectively at day 55 and 78 after disease onset, showed positive IgM using the same serological approach but a different diagnostic test (Serion, Germany) (Table 1). MAT was performed by the NRC using 24 leptospiral antigens representative of Leptospira serogroups for confirmation [6]. The acute serum sample was negative but a high agglutination titer with serovar Hardjo (serogroup Sejroe) was found in the two convalescent sera (Table 1). Laboratory testing for other infectious agents also showed a concomitant acute infection of the patient by an adenovirus.

For epidemiological purpose, we retrospectively performed molecular investigations of the serum collected at day 7 after onset of disease to obtain potential new data on leptospirosis strains circulating in French Guiana. After serum DNA extraction using a QIAamp DNA mini kit (Qiagen, Germany), we obtained a positive PCR based on the *lfb1* gene [7] (Table 1). Then, we performed molecular typing by sequencing the PCR products of the *rrs* (16S rRNA) and *secY* genes [8,9]. The obtained sequences were aligned and analyzed with BioNumerics (version 7.6; 153 Applied Maths N.V., Sint Martens-Latem, Belgium) and compared with available representative sequences for all Leptospira species. These analyses showed 99% and 100% identity for rrs and secY with the pathogenic species L. *santarosai* sequences available in GenBank, respectively.

Conclusion and discussion

Although the incidence of leptospirosis has been recently increasing in French Guiana, knowledge of the circulating strains is scarce and reported epidemiological data are mostly based on MAT. Here, compilation of retrospective molecular and serological investigations identifies, for the first time, the *L. santarosai* serogroup Sejroe in human case infected in French Guiana. Several studies reported the detection of this strain in human and animal (cattle, dog) samples in Latin America (Colombia and Brazil), and the French West Indies. However, none reported an association in humans with this serogroup in these regions [6,10–14].

The epidemiological data reported here provide new information about the *Leptospira* strain circulating in French Guiana. This also emphasizes the importance of investigating reservoirs, such as rodents and cattle, in parallel with increasing awareness of lep-

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tospirosis differential diagnosis among clinicians to better estimate the true epidemiological impact of this disease in the French Guiana population.

Authors contributions

HK and SM wrote the manuscript.
PB, MP and SM performed the analysis.
HK, PB, CM, SH, DH, MP, VC and SM analyzed the data.
PB, MP and VC participated in the critical reading of the manuscript.
All authors read and approved the final version of the manuscript.

Funding

This study was supported in part by public funds allocated for the National Reference Center for Leptospirosis by the French Ministry of Health and also by the EMERGUY research program supported by European funds (FEDER) and assistance from the Region Guyane and Direction Régionale pour la Recherche et la Technologie.

Competing interests

None declared.

Ethical approval

Ethical approval was given by the Comité de Protection des Personnes Sud-Ouest et Outre-Mer III corresponding to the "Emergences virales et syndromes sévères en Guyane-EMERGUY" and was registered on 26 September 2012 as RCB: 2012/86. Informed consent was obtained from the patient.

Acknowledgments

We are thankful to the Genotyping of Pathogens Pole of the Environment and Infectious Risks Research Unit, Institut Pasteur for their support and encouragement.

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