

HUMAN LEPTOSPIROSIS CAUSED BY SEROTYPE *ALEXI* IN BRAZIL

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SUMMARY

The results of a serologic study of a patient serum and the isolation of leptospira from hemo culture are reported. Cross-agglutination and agglutinin absorption tests revealed the isolated strain to be identical to serotype *alexi*.

This is the first known isolation of this serotype in Latin America. This case was a very mild atypical case of leptospirosis.

INTRODUCTION

The first isolations of the members of the *pyrogenes* group in the Western Hemisphere were made by YAGER & GOCHENOUR ¹⁵ in 1952.

Until a few years ago only serotype *icterohaemorrhagiae* had been incriminated in human leptospirosis in Brazil. The importance of leptospirosis has been more widely recognized in recent years in this country since other serotypes such as *wolffi* ⁵ and *andamana* ⁶ have been isolated from clinical cases.

Canicola ³ and *grippityphosa* ⁴ have been incriminated recently as the cause of human leptospirosis in the Brazilian area. Also serologic studies on humans indicated the presence of a multiplicity of leptospiral serotypes, mainly in the State of São Paulo ^{2, 7, 8, 9, 11, 12, 13, 14}.

The results of a 1968 serologic study of a patient serum and the isolation and identification of leptospira from his blood are reported in this paper.

MATERIALS AND METHODS

In 1968, an adult male arrived from the hinterland of Brazil looking for a doctor in a private clinic in Recife, State of Pernambuco, Brazil.

His complaints were severe headache (there was no evidence of meningeal irritation), elevated temperature, abdominal pains, and a rash on the abdomen. Typhus fever was suspected at first.

Stool and hemo cultures for Salmonellosis were carried out and blood was inoculated into tubes of Fletcher's semi-solid medium as a check for leptospire. The patient's serum was also tested for typhus fever and leptospirosis. The microscopic agglutination test employing live antigens was the basic serological technique used with a selected battery of 22 different serotypes (Table I). The serum dilution titer was defined to be the highest dilution in which 50 percent or more of the cells were agglutinated.

The method of preparing antisera in rabbits and the conduct of agglutinin absorption tests

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T A B L E I
Serotypes used as antigens in microagglutination test

Serotype	Strain
1 — icterohaemorrhagiae	RG A
2 — canicola	Hond Utrecht IV
3 — pomona	Pomona
4 — grippotyphosa	Moskwa V
5 — tarassovi	Perepelicin
6 — sejroe	M 84
7 — australis	Ballico
8 — bataviae	Van Tienen
9 — mini	Sari
10 — ballum	Mus 127
11 — wolffi	3705
12 — panama	CZ 214 K
13 — pyrogenes	Salinem
14 — javanica	Veldrat Bat. 46
15 — autumnalis	Akyiami A
16 — butembo	Butembo
17 — celledoni	Celledoni
18 — poi	Poi
19 — andamana	CH 11
20 — saxkoebing	Mus 24
21 — brasiliensis	LT 966
22 — patoc	Patoc I

are described in detail elsewhere¹⁰. Antigen and antisera from recognized serotypes were used in the cross-agglutination and agglutinins absorption tests.

The isolated strain was designated as LT 22-69, it was tested preliminarily by the microscopic agglutination test and then by agglutinin-absorption tests.

The illness lasted only one week. There was no jaundice and the patient recovered with no specific therapy.

R E S U L T S

Stool and hemo cultures were negative for salmonellae, as were the serological results from the Widal test. Leptospire were seen in Fletcher's semi-solid medium five days after inoculation.

Serologic studies for leptospirosis revealed agglutinating antibodies against serotypes *tarassovi* 1:3200, *pyrogenes* 1:1600, *poi* 1:400 and *saxkoebing* 1:200.

Results of preliminary tests (Table II) indicate that the isolated serotype was serologically identical with serotype *alexii*. This was later confirmed by the agglutinin-absorption test performed at the Center for Disease Control in Atlanta, Georgia, U.S.A. (Table III).

T A B L E I I

Cross-agglutination reactions of leptospiral strain LT 22-69 with antisera against 30 leptospiral serotypes and *pyrogenes* serogroup

Antiserum	Homologous	Titer against antigen 22-69	LT 22-69 antisera against antigens in column 1
ballum	6,400	—	—
canicola	12,800	—	—
copenhageni	25,600	—	—
bataviae	25,600	—	—
grippotyphosa	25,600	—	—
autumnalis	51,200	—	—
pomona	25,600	—	—
wolffi	12,800	—	—
australis	6,400	—	—
tarassovi	6,400	—	—
georgia	12,800	—	—
javanica	6,400	—	—
celledoni	12,800	—	—
fort-bragg	12,800	—	—
sentot	25,600	—	—
djasiman	12,800	—	—
borincana	12,800	—	—
cynopteri	12,800	—	—
<i>Pyrogenes serogroup</i>			
pyrogenes	25,600	25,600	800
zanoni	25,600	400	400
abramis	12,800	1,600	3,200
biggis	12,800	25,600	400
hamptoni	12,800	1,600	800
alexii	12,800	25,600	6,400
robinsoni	6,400	1,600	400
manilae	6,400	—	200
myocastoris	6,400	100	800
varela	12,800	—	400
cam-lo	25,600	6,400	6,400
guaratuba	25,600	800	400

(-) Negative at 1:100 final dilution

T A B L E I I I

Results of cross agglutinin-absorption tests on leptospiral strain LT 22-69 with members of the *Pyrogenes* serogroup

Antiserum		Reciprocal of titer against antigen			
Serotype	Absorbed with	Homologous		Absorbing strain	
		Before	After	Before	After
pyrogenes	22-69	25,600	12,800	25,600	100
abramis	22-69	12,800	51,200	800	—
hamptoni	22-69	25,600	12,800	800	—
robinsoni	22-69	3,200	3,200	1,600	—
biggis	22-69	12,800	25,600	6,400	—
alexi	22-69	25,600	800	25,600	100
cam-lo	22-69	25,600	25,600	12,800	—
guaratuba	22-69	3,200	3,200	800	—
LT 22-69	pyrogenes	3,200	1,600	800	—
LT 22-69	abramis	3,200	1,600	1,600	200
LT 22-69	hamptoni	3,200	1,600	6,400	200
LT 22-69	alexi	12,800	—	12,800	—
LT 22-69	myocastoris	3,200	1,600	800	—
LT 22-69	cam-lo	3,200	1,600	6,400	200
LT 22-69	guaratuba	3,200	3,200	400	200
LT 22-69	zanoni	3,200	3,200	400	—
LT 22-69	LT 22-69	3,200	0	100	9

(-) Negative at 1:100 dilution (final dilution)
(0) Was not done (not needed)

DISCUSSION

This case was a very mild atypical case of leptospirosis caused by serotype *alexi*; this is the first known isolation of this serotype in Latin America.

The clinical features of human leptospirosis vary considerably; the illness can be severe as fulminating hepatitis and as mild as influenza. The classic manifestations that suggest Weil's disease are fever, severe prostration, icterus, renal decompensation, hemorrhagic phenomena, and vascular collapse. All these are found in Brazilian leptospirosis. Milder forms of leptospirosis are more difficult to recognize, but a leptospiral etiology should be considered in any febrile patient when severe headaches, vomiting, myalgia, conjunctival injection, proteinuria and neutrophilia are noted. Leptospirosis is to be suspected even more whenever there is a history of exposure to those environmental conditions that favor infection.

Leptospirosis can easily be mistaken for typhoid fever, acute sinusitis, and influenza among other infections. However, there are some clinical manifestations in the early phase that should suggest leptospirosis, namely, an explosive onset with chills, a rapidly rising fever, severe conjunctivitis, and marked myalgia accompanied by nausea and vomiting. The illnesses attributed to infection by serotype *alexi* can range from mild to severe¹.

This case also exemplifies the value of blood culture in the laboratory diagnosis of leptospirosis. When the patient was first seen, typhoid fever was considered a diagnostic possibility. Stool specimens were cultured for enteric pathogens and blood specimens were cultured in Fletcher's medium to check for leptospires. Five days after the blood culture was made leptospires were found.

The immunologic response of the patient is very interesting in that high serum titers were obtained against serotype *tarassovi* 1:3200 and

pyrogenes 1:1600 with cross-agglutination with *poi* and *saxkoebing*. The serological results also corroborated the cultural result.

R E S U M O

Leptospirose humana causada pelo sorotipo alexi, em Recife

Os Autores relatam um caso de leptospirose humana, benigna e atípica. Através de hemocultura, em meio de Fletcher, foi isolada uma amostra de leptospira. Pela tipagem sorológica completa (testes de aglutinação cruzada e absorção de aglutininas) a amostra foi classificada como sorotipo *alexi*. Com o soro do paciente foi feito o teste de aglutinação microscópica obtendo-se o seguinte resultado: *tarassovi* 1:3200, *pyrogenes* 1:1600, *poi* 1:400 e *saxkoebing* 1:200. Este é o primeiro isolamento do sorotipo *alexi* na América Latina.

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