

1 **Title: Leptospirosis in Spanish travelers returning from Chiang Mai: a case series**

2 **Keywords: Leptospirosis;Thailand;imported diseases;zoonosis;travel**

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27 **Abstract**

28 Background: Leptospirosis is an important zoonosis worldwide, nevertheless is often
29 poor recognized in non tropical settings. In Thailand is becoming an emerging disease
30 and Chiang Mai could become a popular spot to acquire the disease amongst travelers.

31 Methods: We describe three cases of imported leptospirosis undifferentiated fever after
32 travelling to Thailand during the summer of 2015 diagnosed at two Spanish hospitals.

33 Results: Our three patients probably acquired leptospirosis while swimming in
34 freshwater around Chiang Mai, a Thailand's northern region with moderate incidence of
35 leptospirosis. Travelers had normal white blood cell counts and low platelets, suggesting
36 leptospirosis after ruling out other imported diseases such as malaria, dengue or
37 typhoid.

38 Conclusion As recent findings point out, low platelets and normal white blood cell
39 counts are clinical features that could help the clinician to suspect Leptospirosis
40 infection. It should be always considered as a cause of fever, particularly if travelers
41 come from a tropical country and have had contact with water or flooding, especially
42 during rainy season.

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45 Leptospirosis is an important zoonosis worldwide, nevertheless is often poorly recognized
46 in non-tropical settings. In Thailand it is becoming an emerging disease(1) and Chiang
47 Mai could become a popular spot to acquire the disease amongst travelers. It should be
48 always considered as a cause of fever, particularly if travelers come from a tropical
49 country especially in those who performed any whitewater sport or other activities such
50 as trekking, adventure races or spelunking.(2)

51 **Case 1**

52 A healthy 34-year-old Spanish male was treated at the Tropical Medicine outpatient
53 clinic in Hospital Clinic of Barcelona after a trip to Thailand, from July 27 to August 17
54 2015. He visited Bangkok, Chiang Mai, Krabi and Kho Tao. On August 12th he started
55 with high-grade fever and severe myalgia and he sought medical attention in a
56 Bangkok's hospital. There, the patient reported trekking and on August the 8th, he swam
57 in freshwater in Chiang Mai, at Mae Taman River spot area. Physical examination was
58 normal and the main laboratory findings were: 120.000 platelets/mm³, normal white
59 blood cell count (WBC) (7300 WBC/mm³), lymphopenia (513 lymphocytes/mm³),
60 aspartate transaminase (AST) 67 U/L, alanine transaminase (ALT) 71 U/L, creatinine
61 1.14mg/dL, and urea 7.7mg/dL. Chest X-ray and urinalysis did not show abnormalities.
62 Ceftriaxone 2g plus doxycycline 200mg, once daily, were initiated empirically by
63 Thailand doctors, after ruling out malaria by antigen rapid test. Influenza virus serology,
64 polymerase chain reaction (PCR) for dengue in serum and PCR for leptospira in urine
65 were negative, while PCR for leptospira in serum was positive. He was discharged five
66 days later without any medical complication. On August 27, leptospirosis was confirmed
67 by microscopic agglutination test (MAT) 1/320 at Hospital Clinic in Barcelona, the
68 highest titre was for serogroup Canicola.

69 **Case 2**

70 A 24 year-old Spanish healthy female was seen at the outpatient tropical medicine clinic
71 in Barcelona after a trip to Thailand. From July 30 to August 16 she visited Bangkok (3
72 days), Ayutthaya (1 day), Sukhothai (1 day), Chiang Mai (5 days) where she practiced
73 trekking and also swam in freshwater, Krabi (1 day, where she visited Railay) and Ko
74 Lipe (1 day). On August 17th she started with low-grade fever, lumbar pain, myalgia
75 and she visited a regional hospital in Barcelona, there a bloodsmear showed no malaria
76 parasites. On August 20 she presented at the Hospital Clinic in Barcelona with high
77 fever, vomiting and diarrhoea and she was hospitalized.

78 Physical examination revealed skin lesions consistent with insect bites on the legs.

79 Blood test showed WBC 8500/mm³, haemoglobin 11.2g/L, thrombocytopenia

80 (88000/mm³), alkaline phosphatase 342U/L, Gamma-glutamyl transferase 337, ALT

81 88U/L, bilirubin 2.1mg/dL. Antigen rapid test and thick smear for malaria as

82 well as PCR for dengue in serum were negative. No antibiotic was initiated at that time.

83 High fever persisted, and on August 26 a lumbar puncture was made, showing

84 pleocytosis with lymphocytic predominance with negative Gram stain and culture. The

85 results were consistent with aseptic meningitis. Ceftriaxone plus doxycycline were

86 started empirically. Serology confirmed the diagnosis (initial MAT 1/80 and 1/640 9

87 days after), although no specific serogroup could reliably be identified and PCR for

88 leptospira in serum. Two days after being diagnosed of meningitis, she started with

89 bilateral ptosis and ocular discomfort and acute anterior uveitis was confirmed by

90 Ophthalmology, probably associated to leptospirosis, she was treated with topical

91 steroids. She was discharged on August 26. The patient was completely recovered one
92 month after discharge

93

94 **Case 3**

95 A 28 year-old man, previously healthy, was visited at the outpatient clinic in the
96 Hospital Virgen de la Salud in Toledo (Spain), three days after his return from a trip to
97 Thailand-Cambodia, from August 8 to the 23rd 2015. He visited Bangkok for two days,
98 Chiang Mai for three days, including an elephant park, where he performed activities
99 such as trekking and swimming in natural pools and waterfalls and also had contact
100 with elephants. He visited Angkor Valley in Cambodia for three days and he finally
101 spent 4 days in Ko Phi Phi. On August the 23rd, he developed high fever (41°C), along
102 with chills and shivers, migratory arthralgia, intense muscular pain (in the lumbar area
103 and calves) and prostration. Physical examination did not show abnormalities.
104 Laboratory findings showed 120.000 platelets/mm³, normal number of WBC (5500
105 WBC / mm³), AST 56 U/L, ALT 108 U/L, creatinine 0.88 mg/dl, urea 15mg / dL,
106 total bilirubin 3.0 mg/dL; direct bilirubin 2.40 mg/dL. Urinalysis: red blood cells 250/
107 µL; bilirubin 3 mg/dL; white blood cells: 100/µL; proteins 500 mg/dL; urine culture was
108 negative. Malaria smear, Dengue PCR, chest x-ray and an abdominal ultrasound did not
109 show any anomalies. Doxycycline 100mg every 12h was initiated empirically. During
110 the treatment, a slight transaminase increase was detected. The patient was managed at
111 the Travel Clinic, ending with clinical and laboratory recovery. MAT serology was
112 found at high titres in patient from a non-endemic country (MAT>1280) suggesting
113 Leptospirosis as the probable cause of the clinical picture.

114 **Discussion**

115 Leptospirosis is one of the major bacterial zoonosis worldwide, caused mainly by direct
116 or indirect contact (through water, food or soil) with urine from an infected animal,
117 particularly mammals. It is recently estimated that leptospirosis causes 1.03 million
118 cases (95% CI 0.43-1.75) and 58900 deaths (95% CI 23800-95900) per year globally,
119 significantly distributed in tropical countries.(3)

120 Leptospirosis is increasing amongst travelers returning from South-east Asia(4) . It is
121 also an emergent zoonosis in Thailand (1), (5), which is considered a high incidence
122 country where cases are up to 48.9 leptospirosis per million population annually (6).

123 Thailand is a very common destination among Spanish travellers. In our travel clinic
124 during 2015 in Hospital Clinic in Barcelona, 10.1% out of 14977 travellers seeking pre-
125 travel advice visited Thailand, being the most common destination among our travelers
126 that year. Once in Thailand, Chiang Mai is one of the favourite destinations. Chiang
127 Mai is a rural area of northern Thailand where travelers often go to trek, to visit
128 Elephant game parks and to perform water recreational activities; thus increasing the
129 odds of leptospirosis transmission by contact with moist soils, flooding , water or
130 animals(7). Incubation periods of our patients along with epidemiology suggest that
131 leptospirosis was probably acquired in Chiang Mai during rainy season (July-August),
132 when most of autochthonous leptospirosis cases occur in the north in Thai
133 population(8),(9) . Patient number 1 had a 4 days incubation period after swimming in
134 Chiang Mai and he only visited Bangkok before became ill, where he did not have any
135 epidemiological risk. In case 2 the incubation period was 6-14 days, and she did not
136 have any other risk as she visited monumental areas and also Bangkok without contact
137 with water, flooding or animals. For the third patient the incubation period was 10-13
138 days, and Chiang Mai was the only spot where he swam in fresh water or trekked, he

139 only visited the beach in the south and he denied having contact there with animals,
140 fresh or flooding water.

141 Leptospirosis incidence rate in northern Thailand (Chiang Mai) during 2015 was
142 1.42/100.000 population(10), nevertheless northeastern and southern regions hold the
143 highest incidence rates across the country (median of 5.43 and 5.36/100.000 population
144 respectively) (10). There are recent reports of leptospirosis amongst travelers who
145 visited Thailand(11),(12) , interestingly since 2013 there have been reports of cases in
146 travelers specifically returning from Chiang Mai. In 2013 our group in Spain reported
147 an imported case acquired after canoeing in the Ping River, the main river of Chiang
148 Mai (13); another Japanese traveler acquired leptospira while swimming in his trip at
149 the same area, as reported by a Japanese group (14). Furthermore, during the summer of
150 2015 a Swiss family was diagnosed with leptospirosis after rafting in Mae Taeng
151 (tributary of the Ping River) (15), a zone next to Mae Taman (the spot where one of our
152 travelers swam in the river).

153 Our patients presented with normal or slightly elevated WBC, lymphopenia and low
154 platelets which could suggest leptospirosis undifferentiated fever over other diagnosis
155 such as dengue or typhoid fever which typically present with low WBC, after ruling out
156 them as well as malaria. This is consistent with an observational prospective study, the
157 authors found that thrombocytopenia without leukocytosis or leukopenia were
158 prominent features amongst leptospira infections(16). Moreover, a noticeable
159 leukocytosis is associated with severe forms of the disease and poorer
160 prognosis(16),(17) .

161 These cases highlight the importance of an accurate travel advice to travelers going to

162 North Thailand: avoid swimming or wading in potentially contaminated soils or waters,
163 avoid contact with flooding, avoid contact with rodents(18) and take precautions with
164 other mammals able to shed leptospirens such as elephants (19).

165 Moreover, since leptospirosis may be a common diagnosis among travelers and routine
166 serological tests are not enough sensitive (20), *Leptospira* spp. PCR and MAT should be
167 considered as a reliable tests to diagnose the disease among the suspected cases.

168 Nevertheless, in our report, one patient was PCR negative and one serovar was not
169 identifiable, PCR should be made at correct timing to be positive, from 0-7 days in
170 serum and up to 21 days in urine, and also MAT need to be performed with a
171 representative panel of serovars from the acquisition of leptospira to be positive.(20)

172 In conclusion, since travelers are increasingly visiting Thailand, undifferentiated, non-
173 malaric, fever cases with normal WBC and low platelets, after ruling out diseases such
174 as typhoid fever, scrub typhus or dengue should be considered as possible leptospirosis
175 cases; especially those with a history of contact with water, flooding, animals or moist
176 soils, particularly if they are coming from a tropical country, such as. Thailand after
177 rainy season (from May/June to October/November); even from regions with moderate
178 leptospirosis incidence rate.

179

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