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# Severe human infection with *Rickettsia felis* associated with hepatitis in Yucatan, Mexico

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## Abstract

*Rickettsia felis* infection usually is a mild-to-moderate illness characterized by general signs and symptoms. Generally, patients do not require hospitalization. However, we detected 2 severe infections with *R. felis*. Our findings support the importance of *R. felis* infection as a potentially severe illness in humans.

### Keywords

*Rickettsia felis*; Hepatitis; Flea-borne spotted fever; Rickettsioses; Emerging diseases

## Introduction

In 1992, a bacterium was detected in cat fleas (*Ctenocephalides felis*) from a commercial colony (Adams et al., 1990). The bacterium was accepted as a new member of the *Rickettsiaceae* family and named *Rickettsia felis*. The first human case confirmed as an infection with *R. felis* was reported in Corpus Christi, Texas (USA), and since then new human cases were reported in México and worldwide (Pérez-Osorio et al., 2008; Schriefer et al., 1994; Zavala-Velazquez et al., 2000).

Usually, *R. felis* infection is the cause of a mild-to-moderate illness characterized by fever, headache, exanthem, and often neurologic and visceral signs and symptoms (Galvao et al., 2004). Although the patients seldom require hospitalization and the infection is self-limiting, the infection can have more severe manifestations involving pulmonary compromise (Zavala-Velázquez et al., 2006). In this work, we report on 2 patients with a severe illness caused by *R. felis* infection with significant hepatic and pulmonary manifestations.



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Our findings support the importance of *R. felis* infection as a potentially severe disease of humans that should be considered, not only in the differential diagnosis for other febrile illnesses in general, but also in the differential diagnosis of diseases such as hepatitis and other infectious diseases which cause respiratory illness.

## Case reports

### Case 1

The first case was a 29-year-old male patient who lived in close contact with domestic animals (cats and dogs). He worked cleaning septic tanks and also had contact with mice and rats. The patient was hospitalized with high fever (40 °C) of 4 days evolution, headache, myalgia, generalized arthralgia, abdominal pain, nausea, vomiting, and hemorrhagic conjunctivitis. The patient was treated with antipyretics and cephalosporin as a primary treatment without remission of the symptoms. Two days after hospitalization, jaundice was observed in the sclerae and skin, and the patient experienced dyspnea. The dyspnea progressed to severe respiratory insufficiency caused by alveolar hemorrhage; the patient was intubated intratracheally and was ventilated mechanically. Hepatosplenomegaly was detected by abdominal ultrasound, with a hepatic border 2.0 cm below the costal arch. Chest radiographs showed a diffuse micronodular infiltrate with right basal consolidation with interstitial lesions.

Clinical laboratory studies revealed severe anemia (hemoglobin 8g/dl [normal range 12–18 g/dl], hematocrit 27% [normal range 37–51%]), severe thrombocytopenia ($30 \times 10^6$ platelets/L [normal range 140–440 $\times 10^6$ platelets/L]), elevated bilirubin (3.1 mg/dl) with cholestatic pattern, neutrophil leukocytosis (91% [normal range 55–68%]), elevated transaminases (AST 108 u/L [normal range 14–36 u/L], ALT 160 u/L [normal range 9–52 u/L]), and hematuria.

Several diseases with similar signs and symptoms such as dengue fever, malaria, brucellosis, human immunodeficiency virus infection (HIV), tuberculosis (TB), viral hepatitis A (HAV), B (HBV), and C (HVC), leptospirosis, and salmonellosis were excluded (Table 1). The diagnosis of rickettsiosis was established based on a single-step PCR amplification using genus-specific primers for the rickettsial genes *ompA*, *ompB*, and citrate synthase (*gltA*) (Table 2), using as controls DNA of *R. felis* (*Ctenocephalides felis*-LSU strain), *R. rickettsii* (Sheila Smith strain), *R. akari* (Kaplan strain), and *R. typhi* (Wilmington strain). The DNA of the controls and the patient were handled separately, and the PCR reactions were prepared in different PCR chambers to avoid contamination. Identification of *R. felis* as the causative agent was achieved by RFLP analyses of the amplified fragments of *gltA* and the comparison of the DNA sequences of the *ompA*, *ompB* and *gltA* PCR amplicons using the Blast software at the National Center for Biotechnology Information (NCBI) (Fig. 1) (Regnery et al., 1991; Altschul et al., 1997). All the sequences showed 100% identity with the corresponding *R. felis* genes (Table 2).

IFA serology revealed antibodies at titers of 1:128 (IgM) and 1:256 (IgG) against *R. rickettsii* and 1:64 (IgM) and 1:128 (IgG) against *R. akari*. No antibodies against *R. typhi* were delected.

The patient was treated with intravenous chloramphenicol (75 mg/kg daily) for 10 days with symptom reduction within 72 h and a complete recovery after 5 days.

### Case 2

A 33-year-old male who lived in close contact with animals (cats, dogs, goats, opossums), described flea bites 4 days before the onset of the illness. The patient was hospitalized with high fever (38.7 °C) of 5 days duration, headache, myalgia, generalized arthralgia, abdominal pain, nausea, and vomiting. After hospitalization, jaundice was observed in the sclerae and skin, and the patient experienced dyspnea. The patient had hepatosplenomegaly detected by abdominal ultrasound, with a hepatic border 1.5 cm below the costal arch. Chest radiographs showed a diffuse micronodular infiltrate.

Clinical laboratory studies revealed severe anemia (hemoglobin 9 g/dl [normal range 12–18 g/dl], hematocrit 29% [normal range 37–51%]); severe thrombocytopenia ($40.5 \times 10^6$ platelets/L [normal range 140–$440 \times 10^6$ platelets/L]); elevated bilirubin (2.7 mg/ dl), neutrophil leukocytosis (85% [normal range 55–68%]), and elevated transaminases (AST 95 u/L [normal range 14–36 u/L], ALT 120 u/L [normal range 9–52 u/L]).

The diagnoses of dengue fever, malaria, brucellosis, HIV, TB, HAV, HBV, HCV, leptospirosis, and salmonellosis were ruled out (Table 1). The diagnosis of rickettsiosis was established based on a single-step PCR amplification using genus-specific primers for the rickettsial genes *ompA*, *ompB*, and citrate synthase gene (*gltA*) (Table 2), using as controls DNA of *R. felis*, *R. rickettsii* (Sheila Smith strain), *R. akari* (Kaplan strain), and *R. typhi* (Wilmington strain). The DNA of the controls and the patient were handled separately to avoid contamination. Identification of *R. felis* as the causative agent was achieved by RFLP analyses of the amplified fragments of *gltA* and the comparison of the DNA sequences of the *ompA*, *ompB*, and *gltA* PCR amplicons using the Blast software at the National Center for Biotechnology Information (NCBI) (Fig. 1) (Regnery et al., 1991; Altschul et al., 1997). All the sequences showed 100% identity with the corresponding *R. felis* genes (Table 2).

IFA serology demonstrated antibodies at titers of 1:64 for IgM and 1:128 for IgG against *R. rickettsii* and 1:64 for IgM and 1:128 for IgG against *R. akari*. No antibodies reactive to *R. typhi* antigens were detected.

The patient was treated with intravenous chloramphenicol (75 mg/kg daily) for 10 days with symptom reduction within 72 h and a complete recovery after 5 days.

## Discussion

Human rickettsiosis caused by *R. felis* has been documented around the world (Pérez-Osorio et al., 2008). In the last 12 years, rickettsioses caused by *R. felis* and *R. rickettsii* infections have been documented in Yucatan, Mexico (Zavala-Velazquez et al., 2000; Zavala-Castro et al., 2006). Usually, the clinical differences between these 2 infections are based on the severity of the signs and symptoms, with mild clinical manifestations for *R. felis* and moderate to severe clinical manifestations for *R. rickettsii*.

In spite of the moderate symptoms caused by some *Rickettsia* species, visceral and vascular involvement could have serious consequences for some patients, and even cause clinical manifestations usually not associated with the disease. Recently, hepatitis has been associated with rickettsialpox and is a well-documented manifestation of some cases of Rocky Mountain spotted fever but seldom occurs in *R. akari* infection which manifests moderate self-limiting signs and symptoms (Madison et al., 2008). Such an atypical clinical manifestation could also take place in other rickettsial infections and mislead the physician to an incorrect diagnosis.

As in other rickettsial infections, the course of illness caused by *R. felis* includes mild nonspecific signs and symptoms such as fever, arthralgia, myalgia, headache, and macular rush, traditionally confused with other febrile diseases especially with dengue fever, leptospirosis, and any other febrile exanthematous infection caused by a virus or another bacterium. However, other nonspecific but more severe clinical manifestations may occur in *R. felis* infection and other rickettsioses, such as thrombocytopenia, pulmonary compromise, and anemia and should stimulate the consideration of rickettsial infection in the differential diagnosis as well as other severe illnesses such as hepatitis, pneumonia, brucellosis, and malaria.

A grave clinical condition has not been reported in the illness caused by *R. felis*. The severe thrombocytopenia and anemia present in both patients were associated with bleeding represented by hematuria, alveolar hemorrhage, and hemorrhagic conjunctivitis almost certainly caused by the vascular damage that occurs in rickettsial infections. The vascular injury also caused significant hepatic injury in both patients and respiratory compromise in one. The elevated serum bilirubin and transaminases indicated hepatic involvement clinically represented by jaundice that required a workup for viral hepatitis.

Although *R. felis* infection has not been described as a cause of severe disease, some dissimilarity in the clinical features of the *R. felis*-infected patients from different countries has been reported (Galvao et al., 2004). Such reported dissimilarity and our findings suggest a potentially broader range of clinical manifestations in *R. felis* patients.

More investigation has to be done to elucidate the factors involved in the altered pathogenesis of some rickettsial diseases or to determine if the differences in the clinical features of rickettsial infections could be a consequence of genetic differences in the rickettsial strains as well as the result of differences in the genetics and environmental conditions of the host population and/or in rickettsial adaptation to diverse hosts and vectors. Differences in the amino acid composition of the outer membrane protein A of *R. felis* have been reported, and the changes may have implications in the immune recognition and even in the adaptation process to different environments, and could also be involved in the pathogenesis of rickettsial diseases (Pornwiroon et al., 2006: Zavala-Castro et al., 2005).

As in other rickettsial cases in our region, the titer of the immunofluorescence serology does not have a direct correlation with the severity of illness. Both patients had antibodies that were reactive with only spotted fever group rickettsiae, with a higher titer against *R. rickettsii* antigens.

It is important to reconsider the possibility that uncommon signs and symptoms of some rickettsial infections might lead to misdiagnosis and unnecessary deaths without timely and accurate diagnosis.

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## References

Adams JR, Schmidtmann ET, Azad AF. Infection of colonized cat fleas, *Ctenocephalides felis* (Bouche), with a rickettsia-like microorganism. Am J Trop Med Hyg. 1990; 43:400–409. [PubMed: 2240368]

Altscbul SF, Madden TL, Schäffer AA, Zhang J, Zhang Z, Miller W, Lipman DJ. Gapped Blast and PSI-Blast: a new generation of protein database search programs. Nucleic Acids Res. 1997; 25:3389–3402. [PubMed: 9254694]

Galvao MA, Mafra CL, Chamone ChB, Calic SB, Zavala-Velázquez JE, Walker DH. Clinical and laboratorial evidence of *Rickettsia felis* infections in Latin America. Rev Soc Bras Med Trop. 2004; 37:238–240. [PubMed: 15330064]

Madison G, Kim-Schluger L, Braverman S, Nicholson WL, Wormser GP. Hepatitis in association with rickettsialpox. Vector-Borne Zoonotic Dis. 2008; 8:111–115. [PubMed: 18171106]

Pérez-Osorio CE, Zavala-Velázquez JE, Arias JJ, Zavala-Castro JE. *Rickettsia felis* as emergent global threat for humans. Emerg Infect Dis. 2008; 14:1019–1023. [PubMed: 18598619]

Pornwiroon W, Pourciau S, Foil LD, Macaluso KR. *Rickettsia felis* from cat fleas: isolation and culture in a tick-derived cell line. Appl Environ Microbiol. 2006; 72:5589–5595. [PubMed: 16885313]

Regnery RL, Spruill CL, Plikatys BD. Genotypic identification of rickettsiae and estimation of intraspecies sequence divergence for portions of two rickettsial genes. J Bacteriol. 1991; 173:1576–1589. [PubMed: 1671856]

Schriefer ME, Sacci JB Jr, Dumler JS, Bullen MG, Azad AF. Identification of a novel rickettsial infection in a patient diagnosed with murine typhus. J Clin Microbiol. 1994; 32:949–954. [PubMed: 8027348]

Zavala-Castro JE, Small M, Keng C, Bouyer DH, Zavala-Velázquez JE, Walker DH. Transcription of the *Rickettsia felis ompA* gene in naturally infected fleas. Am J Trop Med Hyg. 2005; 73:662–666. [PubMed: 16222005]

Zavala-Castro JE, Zavala-Velázquez JE, Walker DH, Ruiz-Arcila EE, Laviada-Molina H, Olano JP, Ruiz-Sosa JA, Small MA, Dzul-Rosado KR. Fatal human infection with *Rickettsia rickettsii*, Yucatán, Mexico. Emerg Infect Dis. 2006; 12:672–674. [PubMed: 16704818]

Zavala-Velazquez JE, Ruiz-Sosa JA, Sanchez-Elias RA, Becerra-Carmona G, Walker DH. *Rickettsia felis* rickettsiosis in Yucatan. Lancet. 2000; 356:1079–1080. [PubMed: 11009147]

Zavala-Velázquez JE, Laviada-Molina H, Zavala-Castro JE, Pérez-Osorio C, Becerra-Carmona G, Bouyer D, Walker D. *Rickettsia felis*, the agent of an emerging infectious disease: report of a new case in México. Arch Med Res. 2006; 37:419–422. [PubMed: 16513497]

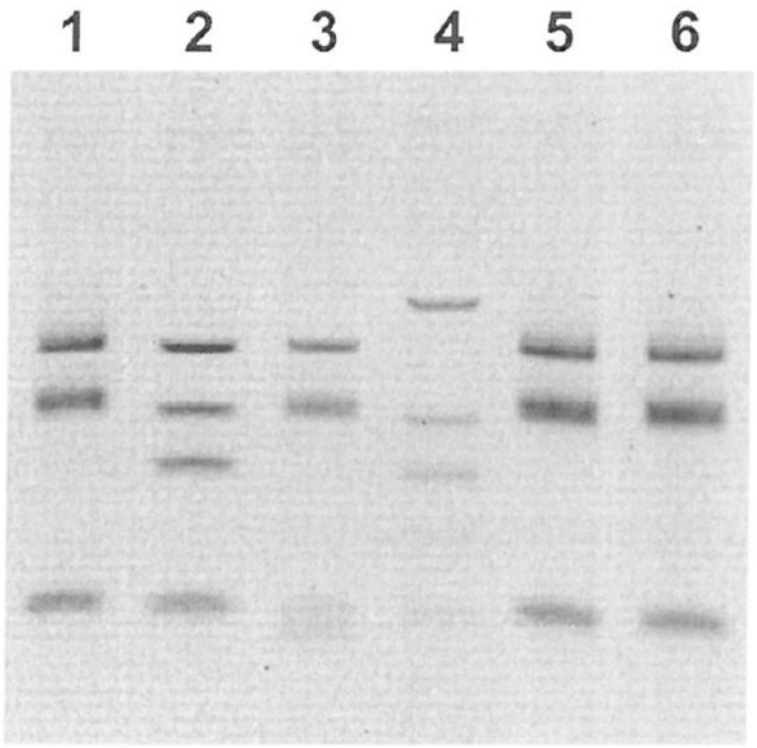


**Fig. 1.**

Restriction fragment length polymorphism (RFLP) of the *gltA* PCR product. Lane 1, *R. felis* positive control; lane 2, *R. akari* positive control; lane 3, *R. rickettsii* positive control; lane 4, *R. typhi* positive control; lane 5, 32-year-old patient; lane 6, 29-year-old patient.

## Table 1

Pathogens excluded by serologic and molecular methods in both patients.

| Pathogen | Diagnostic method | | Results | |
|---|---|---|---|---|
| | Serologic | Molecular | Patient 1 | Patient 2 |
| *Leptospira interrogans* | Microscopic agglutination test (MAT) | | Neg. | Neg. |
| Dengue virus | ELISA[a] | RT-PCR 5′-TCAATATGCTGAAACG CGCGAGAAACCG-3′ 5′-TTGCACCAACAGTCAATGTCTT CAGGTTC-3′ | Neg./Neg. | Neg./Neg. |
| Hepatitis A virus | Anti-HAV[b] | | Neg. | Neg. |
| Hepatitis B virus | HBcAG[b], HBsAg[b] | | Neg. | Neg. |
| Hepatitis C virus | HCV Ag/Ab[b] | | Neg. | Neg. |
| Human immunodeficiency virus | HIV Ag/Ab[b] | | Neg. | Neg. |
| *Mycobacterium tuberculosis* | ELISA[a] | PCR 5′-CACATGCAAGTCGAACGG AAAGG-3′ 5′-GCCCGTATCGCCCGCACGCTCACA-3′ Hybridizing probe 5-GGTGGA AAGCGCTTTAGCGGT-3′ | Neg./Neg. | Neg./Neg. |
| *Brucella abortus* | ELISA[a] | | Neg. | Neg. |
| *Salmonella typhi* (Widal's test) | Detection of antigens O, Vi, and H | | Neg. | Neg. |
| *Plasmodium* falciparum, vivax, ovale, malariae | Malaria Ab[c] | | Neg. | Neg. |

[a] Diagnostic Automation Inc., Calabasas, CA, USA

[b] Abbott Laboratories, Abbott Park, 1L, USA

[c] Bio-Rad Laboratories Inc., Hercules, CA, USA

**Table 2**

Molecular probes and positive controls used in the PCR reactions and sequence identity of the amplicons found in both patients.

| Gene | Primers | Size (bp) | Identity of PCR aimplicon | | | |
|---|---|---|---|---|---|---|
| | | | ***Rickettsia felis, Ctenocephalides felis*-LSU strain (%)** | ***Rickettsia rickettsii* Sheila Smith (%)** | ***Rickettsia akari* Kaplan strain (%)** | ***Rickettsia typhi* Wilmington strain (%)** |
| *ompB* | Rp.330(2) 5′-atggctcaaaaaccaaattttc-3′ | 991 | 100 (AF182279) | 89(CP000848.1) | 89(CP000847.1) | 83 (AE017197.1) |
| | Rp.330(2) 5′-agctctacctgctccattatct-3′ | | 99(CP000053.1) | | | |
| Citrate synthase (*gltA*) | RpCS.1258 5′-attgcaaaaa gtaccgtaaaca-3′ | 382 | 100 (CP000053.1) | 95 (CP000848.1) | 96 (CP000847.1) | 95 (NC_006142) |
| | RpCS.877 5′-gccccgccgtgggcaggccccc-3′ | | | | | |
| *ompA* | Rrl90.70p 5′-atggcgaatatttctccaaaa-3′ | 512 | 100 (AJ563398) | 94 (CP000848.1) | 95 (CP000847.1) | |
| | Rr190.602n 5′-agtgcagcattcgctcccct-3′ | | 99 (CP000053.1) | | | |

AF182279, infected cat fleas, Louisiana Stale University Agricultural Center.

AJ563398, infected domestic dog fleas, Mexico.

CP000053, Marseille-URRWFXCa12 complete genome.