10.1111/j.1469-0691.2008.02145.x

Concomitant human granulocytic anaplasmosis and Lyme neuroborreliosis

S. Lotric-Furlan[1], E. Ruzic-Sabljic[2] and F. Strle[1]

[1]Department of Infectious Diseases, University Medical Centre Ljubljana and [2]Faculty of Medicine, Institute of Microbiology and Immunology, Ljubljana, Slovenia

INTRODUCTION

Tick-transmitted zoonoses such as human granulocytic anaplasmosis (HGA), Lyme borreliosis and tick-borne encephalitis (TBE) are endemic in several European countries, including Slovenia. *Ixodes ricinus*, the most prevalent species of hard ticks in Slovenia, is the principal vector for *Borrelia burgdorferi* sensu lato, *Anaplasma phagocytophilum* and TBE virus [1].

Information on clinically manifested co-infections with tick-transmitted pathogens is incomplete. Findings from the USA indicate that the frequency of simultaneous diseases caused by infection with more than one tick-borne pathogen is low and varies among geographic regions. Reports from the USA involve cases with concurrent erythema migrans (early localised borrelial skin infection) and HGA [2]. Data on co-infections with different tick-borne pathogens in Europe are limited to the reports on co-infections with TBE virus and *B. burgdorferi* s.l. in patients with acute meningitis [1] and on co-infection with TBE virus and *A. phagocytophilum* [3]. The other possible combinations in European populations, including infections with *A. phagocytophilum* and *B. burgdorferi* s.l., have been indicated mainly but not exclusively by serological findings [3–5].

Residents of Slovenia are often exposed to ticks and thus are at risk of acquiring infection with multiple tick-borne pathogens. Herein we present a patient with acute HGA, established by positive PCR result and seroconversion, in whom borrelial infection was ascertained by the isolation of *Borrelia garinii* from cerebrospinal fluid (CSF).

Corresponding author and reprint requests: Stanka Lotric-Furlan, PhD, MD, Department of Infectious Diseases, University Medical Centre Ljubljana, Japljeva 2, 1525 Ljubljana, Slovenia
E-mail: stanka.lotric-furlan@mf.uni-lj.si

No conflicts of interest declared.

CASE PRESENTATION

On 18 July 2006, a 63-year-old man from the central part of Slovenia was admitted to the Department of Infectious Diseases, University Medical Centre Ljubljana, Slovenia, with a 5-day history of fever up to 40°C, severe headache, chills, malaise, arthralgia and myalgia. He recalled a tick bite without skin lesions 2 weeks prior to the onset of the illness. His previous medical history was unremarkable. With the exception of fever, the physical examination did not show any notable abnormality; rash and meningeal signs were not present.

Routine laboratory tests showed leukopenia (3400/mm^3; normal value 4500–10 000/mm^3), thrombocytopenia (70/mm^3; normal value 140.0–340.0/mm^3), elevated concentration of serum C-reactive protein (112 mg/L; normal <5 mg/L) and elevated concentration of procalcitonin (1.76 μg/L; normal <0.5 μg/L). Lumbar puncture was performed; routine CSF tests revealed no abnormalities but *B. garinii* was isolated from CSF. Duration of fever was 9 days; fever vanished within 24 h after introduction of treatment with doxycycline for 7 days. At subsequent evaluation (after 2 weeks), the patient reported feeling better and laboratory tests were within the normal range. The subsequent clinical course was uneventful.

Microbiological procedures for infection with tick-transmitted microorganisms such as *A. phagocytophilum*, *Ehrlichia chaffeensis*, *B. burgdorferi s.l.* and TBE virus were performed to elucidate the cause of the illness. Giemsa-stained peripheral blood smear examination by light microscopy for the presence of ehrlichial morulae within leukocytes was negative. Serum samples were tested by IFA for the presence of specific IgG antibodies to *A. phagocytophilum* (strain USG3 propagated in HL60 promyelocyte cells), and for IgM and IgG antibodies to *E. chaffeensis* antigens (MRL Diagnostics). To determine serum and CSF IgM and IgG antibodies against *B. burgdorferi* s.l., IFA

without absorption was performed using whole cells of a local isolate of *B. afzelii* as an antigen. At presentation a CSF sample was inoculated directly into a tube with a modified Kelly-Pettenkofer culture medium and incubated for 9 weeks.

Serological testing revealed IgG seroconversion to *A. phagocytophilum* (negative IFA IgG titre at initial examination, >1:1024 2 weeks later). Primers Ehr521 and Ehr790, which amplified the 16S rRNA gene of *A. phagocytophilum*, produced a fragment of the expected size (293 bp) in the acute-phase blood specimen. Serum and CSF borrelial IgM and IgG antibodies determined by IFA were negative but *Borrelia* was isolated from CSF and was typed as *B. garinii* using *Mlu*I restricted DNA, separated by pulse–fluid gel electrophoresis.

Two months after discharge from hospital, when we obtained information on isolation of borrelia from CSF, 14-day treatment with ceftriaxone was instituted.

DISCUSSION

In regions endemic for several tick-transmitted diseases such as Slovenia, persons exposed to ticks may acquire infections with different pathogens, resulting in the presence of more than one tick-borne disease. In our report the diagnosis of confirmed HGA was established by febrile illness after a tick bite, and positive PCR and seroconversion. Borrelial infection was ascertained by isolation of *B. garinii* from CSF without typical clinical signs for Lyme neuroborreliosis. In our patient clinical signs of acute HGA predominated over the signs indicative of Lyme neuroborreliosis. To our knowledge, the patient reported herein represents the first case of concurrent confirmed HGA and confirmed Lyme neuroborreliosis.

The diagnosis of concomitant infection with *A. phagocytophilum* and *B. burgdorferi* s. l. is potentially important because the clinical course of each of the diseases may be modified in the presence of the other and because concomitant infections may affect the choice of antimicrobial therapy.

REFERENCES

1. Lotric-Furlan S, Petrovec M, Avsic-Zupanc T *et al*. Prospective assessment of the etiology of acute febrile illness after a tick bite in Slovenia. *Clin Infect Dis* 2001; **33**: 503–510.
2. Steere AC, McHugh G, Suarez C, Hoitt J, Damie N, Sikand VK. Prospective study of coinfection in patients with erythema migrans. *Clin Infect Dis* 2003; **36**: 1078–1081.
3. Lotric-Furlan S, Petrovec M, Avsic-Zupanc T, Strle F. Concomitant tick-borne encephalitis and human granulocytic ehrlichiosis. *Emerg Infect Dis* 2005; **11**: 485–488.
4. Bjoersdorff A, Wittesjo B, Berglund J, Massung RF, Eliasson I. Human granulocytic ehrlichiosis as a common cause of tick-associated fever in Southeast Sweden: report from a prospective clinical study. *Scand J Infect Dis* 2003; **34**: 187–191.
5. Grzeszezuk A, Ziarko S, Kovalchuk O, Stanczak J. Etiology of tick-borne febrile illnesses in adult residents of North-Eastern Poland: report from a prospective clinical study. *Int J Med Microbiol* 2006; **296** (suppl): 242–249.