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Bartonella alsatica endocarditis in a French patient in close contact with rabbits

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INTRODUCTION

Bartonella species are Gram-negative bacilli that belong to the alpha 2 subgroup of Proteobacteria and are agents of blood culture-negative endocarditis (BCNE) [1]. Among the *Bartonella* species involved in BCNE, *B. henselae* and *B. quintana* are the main causative agents [1]. However, endocarditis due to other rare *Bartonella* spp. have been reported, including *B. elizabethae*, *B. vinsonii* subsp. *berkhoffii*, *B. vinsonii* subsp. *arupensis*, *B. koehlerae* and recently *B. alsatica* [2,3]. *B. alsatica* is a recently identified agent that causes bacteraemia in healthy wild rabbits in Alsace, France [4]. To the best of our knowledge there is only one case of infection with this species in humans, i.e, an endocarditis in a 74-year-old man who was in close contact with rabbits [2]. In the present study, we report the second human case of *B. alsatica* endocarditis in a French patient in close contact with rabbits, diagnosed using specific serogical methods, including microimmunofluorescence and Western blot with cross-adsorption studies.

METHODS

Methods used for serological diagnosis were microimmunofluorescence (MIF), as well as Western-blot and cross-adsorption performed as previously described [2,5]. For serological testing by MIF, a titre of IgG ≥1:400 to any *Bartonella* species was found to be highly predictive of *Bartonella* endocarditis. PCR from blood was performed targeting a portion of the *Bartonella* internal transcribed spacer (ITS) region and the *ftsZ* gene in a Lightcycler (Roche Diagnostics, Meylan, France) apparatus using primers and TaqMan probes (Applied Biosystems, Coignienes, France). Culture was carried out on blood-enriched media in a moist atmosphere with 5% CO_2 as well as in HEL cells.

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RESULTS

A 77-year-old woman, rabbit breeder, was hospitalised in December 2006 for fever, and systolic murmur suggestive of a cardiac disorder. Echocardiography revealed a mitral vegetation that confirmed the diagnosis of endocarditis. The causative role of *B. alsatica* was proven by serology using MIF and Western-blot following cross-adsorption (Fig. 1). The serum showed a titre of 1:400 for all *Bartonella* tested, including *B. alsatica* (Fig. 1). We performed Western blotting using *Bartonella* sp. antigens and after adsorption only *B. alsatica* antigens retained all antibodies. Regular blood culture and PCR were negative.

The patient was successfully cured with a course of gentamicin for 15 days and amoxicillin for 6 weeks. On follow-up, 1 year later, the patient was well but reported that all rabbits in her farm had died. Retrospective analysis from January 2006 to December 2007 revealed that this case was the only one due to *B. alsatica* among 17 cases (5.9%) of *Bartonella* endocarditis, including five due to *B. henselae* and 11 due to *B. quintana*.

CONCLUSIONS

To the best of our knowledge, this is the second human case of *B. alsatica* endocarditis. The diagnosis was made using serological methods, including MIF and Western blot with cross-adsorption studies. These latter methods have been successfully used for the diagnosis of *Bartonella* endocarditis [5]. The association of an IgG titre ≥1:400 and positive Western blot and cross-adsorption in patients with PCR proven *Bartonella* endocarditis were demonstrated to identify efficiently the causative species in all cases [5]. When applied to patients diagnosed using serological tests only, this technique allowed identification of the causative species in 20 of 22 cases [5]. In the present study, although culture and PCR from

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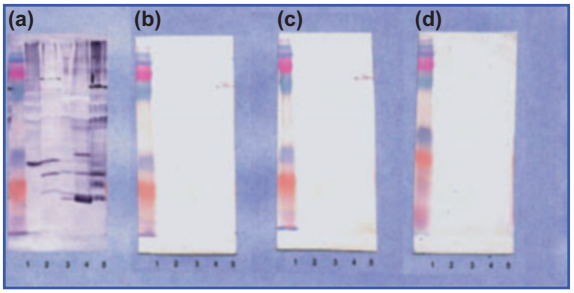

Fig. 1. Western blot before (a) and after cross-adsorption with *Bartonella quintana* (b), *Bartonella henselae* (c), and *Bartonella alsatica* (d). Line 1, *Bartonella quintana*; line 2, *Bartonella henselae*; line 3, *Bartonella elizabethae*; line 4, *Bartonella vinsonii* subsp. *berkhoffii*; line 5, *Bartonella alsatica*.

blood were negative, it is well known that these methods lack sensitivity in these samples. Cardiac valves remain the best samples for PCR and isolation but such a specimen was not available in our case. However, the patient was successfully cured using the current recommended treatment, i.e., an association of aminoglycoside and amoxicillin [1]. Our finding is interesting because the only known reservoir of *B. alsatica* to date is rabbits and the two human cases support the hypothesis of a link between rabbits and infection in these patients. Although rabbits have been reported to be asymptomatic in the previous studies, in the present study all of them died. Nevertheless, blood samples from these rabbits were not available. This current case confirms that *B. alsatica* may be an agent at least of BCNE and infected wild rabbits could be a potential source of human contamination when hunted and skinned. Further studies are warranted to better understand the relationships between rabbits and *B. alsatica* infections in humans.

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