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Journal

Journal of Clinical Microbiology, 53(8)

ISSN

0095-1137

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Publication Date

2015-08-01

DOI

10.1128/jcm.02888-14

Peer reviewed

JCM Journals.ASM.org

Bartonella quintana Aortitis in a Man with AIDS, Diagnosed by Needle Biopsy and 16S rRNA Gene Amplification

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A man with newly diagnosed AIDS presented with months of back pain and fever. Computed tomography (CT) results demonstrated aortitis with periaortic tissue thickening. DNA amplification of biopsy tissue revealed *Bartonella quintana*, and *Bartonella* serologies were subsequently noted to be positive. The patient improved with prolonged doxycycline and rifabutin treatment. This case illustrates how molecular techniques are increasingly important in diagnosing *Bartonella* infections.

CASE REPORT

A 48-year-old heterosexual African male with type II diabetes presented to an emergency room (ER) with several months of abdominal pain, back pain, polydipsia, loss of 30 pounds of body weight, and subjective fevers. He was febrile (38.6°C) and tachycardic, with a glucose level of 298 mg/dl. He was given intravenous fluids and metformin and discharged from the ER. Subsequently, his HIV-1 test returned a positive result; his $CD4^+$ T cell count was 68 cells/mm^3 (7%), and his HIV-1 RNA level was 537,519 copies/ml. He was empirically started on antiretroviral therapy and prophylactic trimethoprim-sulfamethoxazole. Four weeks later, the patient described persistent abdominal and back pain, fever, and chills. The mid-thoracic back pain was sharp, constant, and relieved by leaning forward.

The patient worked as a taxi driver, lived alone in an apartment, and had no pets. He grew up in Ethiopia and moved to the United States in 1991. He reported being heterosexual and denied contact with commercial sex workers or having surgeries or tattoos. He reported no alcohol, tobacco, or illicit drug use. He had last traveled to Ethiopia in 2006, stayed in rural areas with goats, sheep, cows, dogs, and cats, and consumed only store-bought milk and meat.

On examination, the patient had no thrush or lymphadenopathy. His abdomen was soft and mildly tender in response to palpation throughout, without rebound. There was no tenderness in response to palpation along the spine. He had no cutaneous lesions. His laboratory results were notable for a white blood cell count of $2.9 \times 10^3/\mu l$, with 38% polymorphonuclear cells, 36% lymphocytes, 8% monocytes, 15% eosinophils, and a hemoglobin level of 8.9 g/dl. His liver function test results were normal.

Single-phase phase-contrast-enhanced CT results demonstrated abnormal circumferential soft tissue thickening involving the lower abdominal aorta, with additional periaortic soft tissue, inseparable from the aortic wall. Heterogeneous enhancement within the soft tissue suggested active inflammation. A subsequent multiphase CT angiography (CTA) procedure confirmed aortic wall thickening, extending from the superior mesenteric artery to the proximal left common iliac artery (Fig. 1A). Additionally, a wedge-shaped hypodense region in the posterior left kidney was suspicious for a small infarct.

The patient was hospitalized for further evaluation. Routine bacterial, mycobacterial, and fungal blood culture results were negative, as were those of *Coccidioides* complement fixation and immunodiffusion assays, *Cryptococcus* antigen tests, and *Histoplasma* antigen and antibody tests. Results of ovum and parasite stool studies, rapid plasma reagin and *Treponema pallidum* particle agglutination tests, and a *Mycobacterium tuberculosis* gamma interferon release assay were all negative. The patient had two negative sputum results by smear and culture for acid-fast bacilli (AFB) as well as by PCR using GeneXpert (Cepheid, Sunnyvale, CA). The C-reactive protein (CRP) level and erythrocyte sedimentation rate (ESR) were elevated at 25 mm/h (normal range, 0 to 15 mm/h) and 45 mg/liter (normal, <3.1 mg/liter), respectively. A transthoracic echocardiogram (TTE) demonstrated no evidence of endocarditis; the chordae of the mitral valve were redundant, but there was no suggestion of pendant masses or prolapse. Transesophageal echocardiogram (TEE) was recommended because of the potential embolus in the left kidney on CT and the redundant mitral valve chordae on TTE, but the patient declined this evaluation.

Percutaneous fine-needle aspiration and a 20-gauge core biopsy of the inflamed periaortic tissue were performed by an interventional radiologist (Fig. 1B), with a vascular surgeon on call. Histopathological tissue staining for AFB was initially interpreted as showing "occasional rod-like structures" without beading (Fig. 2). Microbiological tests performed on the tissue gave a negative

Received 9 October 2014 **Returned for modification** 3 November 2014
Accepted 28 May 2015

Accepted manuscript posted online 10 June 2015

Citation Lee SA, Plett SK, Luetkemeyer AF, Borgo GM, Ohliger MA, Conrad MB, Cookson BT, Sengupta DJ, Koehler JE. 2015. *Bartonella quintana* aortitis in a man with AIDS, diagnosed by needle biopsy and 16S rRNA gene amplification. J Clin Microbiol 53:2773–2776. doi:10.1128/JCM.02888-14.

Editor: A. B. Onderdonk

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doi:10.1128/JCM.02888-14

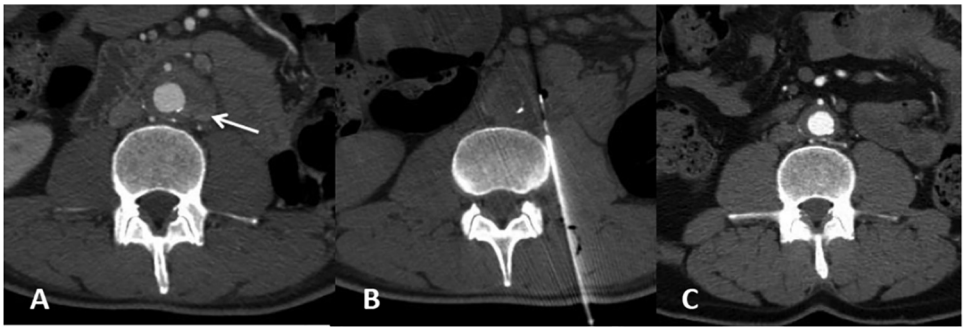

FIG 1 (A) Computed tomography (CT) angiography of the aorta demonstrating circumferential soft tissue thickening of the aorta inferior to the origin of the superior mesenteric artery (arrow), with abnormal periaortic soft tissue indistinguishable from the aortic wall. Heterogeneous enhancement of the soft tissue suggests active inflammation and calcifications present in periaortic tissue. (B) A left paraspinal approach CT-guided percutaneous biopsy specimen demonstrates direct sampling of the inflamed aortic tissue. (C) Follow-up CT angiogram, obtained approximately 7 months after presentation, demonstrating near-complete resolution of circumferential mural thickening and enhancement consistent with treatment response.

AFB smear result and negative culture results for bacteria, fungi, and AFB. The patient was started on empirical *M. tuberculosis* (rifabutin, isoniazid, pyrazinamide, and ethambutol) and *M. avium-M. intracellulare* (clarithromycin) therapy because of the histopathological tissue AFB stain result (even though the bacilli had an atypical appearance), the severe immunosuppression of the patient due to AIDS, the severity of the illness and location of the lesion, and the patient's long-term potential exposure in a country where *M. tuberculosis* is endemic.

The patient's pain significantly improved over the next 2 weeks on therapy. Because no microorganisms were isolated from the biopsy tissue and because of the high-risk location of the lesion and the potential long-term therapy required, the formalin-fixed, paraffin-embedded pathology specimen was sent to the University of Washington Molecular Diagnostics Laboratory (UW-MDL) for identification of the rod-like organisms using PCR DNA amplification. PCR tests were negative for *M. tuberculosis* and *M. avium-M. intracellulare*, but PCR using broad-range, bacterial 16S rRNA gene primers unequivocally identified *Bartonella quintana* (forward primer 27F sequence, 5′-AGAGTTTGATCCTGGCTCAG-3′; reverse primer 357ml sequence, 5′-CTGCTGCCICCCGTAGGAG-3′). The amplified product included 263 nucleotides of the 16S rRNA gene (GenBank accession number KR866081). Basic Local Alignment Search Tool (BLAST) analysis revealed a 100% match to three different strains of *B. quintana* (RM-11, Toulouse, and Fuller) in the database. The closest match with any other known *Bartonella* spp. was 98% nucleotide identity with *B. grahamii* and *B. henselae*. Notably, the last case of *B. quintana* identification by PCR at UW-MDL had occurred more than 1 year earlier, making laboratory contamination unlikely. PCR using broad-range fungal primers detected *Malassezia restricta*, a common skin commensal which, according to UW-MDL researchers,

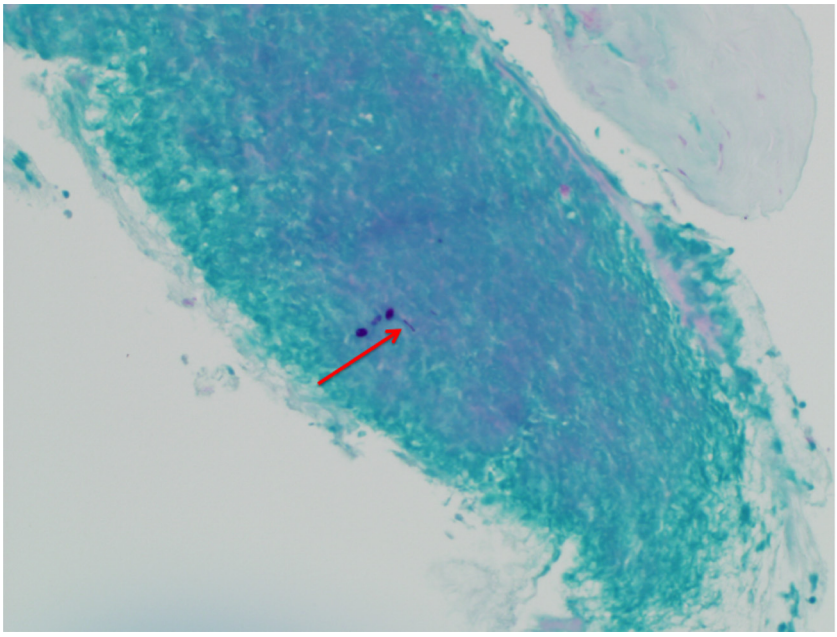

FIG 2 On histopathological examination, the core biopsy tissue was sparsely cellular, showing mostly acellular fibrous tissue with occasional spindle cells. Initially, the AFB tissue staining was interpreted as showing occasional rod-like structures (red arrow). Ultimately, these rod-like structures were considered to lack the characteristic beading typically seen with mycobacterial organisms, and therefore the rod-like structures were considered to represent artifacts.

TABLE 1 *Bartonella quintana* and *Bartonella henselae* serologies by indirect fluorescence assay, $CD4^+$ T cell counts, and *Bartonella* treatment[a]

Date of sample collection	*Bartonella quintana*		*Bartonella henselae*		CD4 count (in cells/mm^3) (%)	*Bartonella* treatment[b]
	IgM titer	IgG titer	IgM titer	IgG titer		
13 February 2013	<1:16	**1:1,024**	<1:16	**>1:1,024**	114 (8)	C, R, I, P, E
13 March 2013	<1:16	**1:512**	<1:16	**>1:1,024**	127 (9)	C, R, I, P, E
13 May 2013	<1:16	**1:512**	<1:16	**>1:1,024**	144 (10)	D, R
13 June 2013	<1:16	**1:512**	<1:16	**>1:1,024**		D, R
13 July 2013	<1:16	Indet[c]	<1:16	**>1:1,024**		D, R
15 August 2013	<1:16	**1:128**	<1:16	**1:512**	165 (10)	D, R
12 September 2013	<1:16	<1:64	<1:16	**1:1,024**		D, R
31 October 2013	<1:16	<1:64	<1:16	**1:512**	203 (11)	D, R
16 January 2014	<1:16	<1:64	<1:16	**1:256**	306 (14)	D
11 July 2014	<1:16	<1:64	<1:16	**1:128**	242 (14)	none

[a] Bold font indicates positive indirect fluorescence assay (IFA) test results.
[b] Treatment drugs: C, clarithromycin; R, rifabutin; I, isoniazid; P, pyrazinamide; E, ethambutol; D, doxycycline.
[c] Indet, indeterminate; i.e., a high degree of nonspecific fluorescence was observed.

is a common cause of contamination in tissue blocks. Fungal blood and biopsy culture results were negative.

Doxycycline (100 mg administered orally [p.o.] twice a day [b.i.d.]) therapy was started after the *B. quintana* PCR results were obtained. The patient reported no direct risk factors for *B. quintana* infection, including prior homelessness or infestation with body lice, and no pruritic rash. He did occasionally provide rides to homeless passengers in his taxicab, but never saw any lice in his cab or on his body. Subsequent serological tests for *Bartonella* were performed, and an indirect fluorescence antibody (IFA) test for *Bartonella* (ARUP Laboratories) demonstrated a *B. quintana* IgG of 1:1,024 (negative, <1:64) and IgM of <1:16 (negative, <1:16) and a *B. henselae* IgG of >1:1,024 and IgM of <1:16 (Table 1). After PCR results were obtained, we attempted unsuccessfully to culture blood for *Bartonella*. Unfortunately, the sole blood specimen available that was drawn before the patient received antibiotics was only 1 ml, the specimen had been refrigerated for 3 weeks, and the blood was in a heparin tube (instead of the ideal EDTA tube), all of which are suboptimal for *Bartonella* isolation. The result of Warthin-Starry silver staining of the core biopsy tissue sample was negative, and histopathological examination revealed no specific characteristics of *Bartonella* infection.

A repeat CTA procedure performed 3 weeks after hospital discharge demonstrated no change, but the ESR had decreased to 29 and the CRP level to 1.0 and his eosinophilia had resolved. The patient was maintained on doxycycline (100 mg p.o. b.i.d.) and rifabutin (150 mg p.o. daily) treatment for an additional 6 months and then on doxycycline treatment alone until his $CD4^+$ T cell count remained consistently above 200 cells/mm^3 (approximately 14 months after the initial diagnosis of *Bartonella* aortitis) (Table 1) (1). Eighteen months after the diagnosis, his *B. quintana* IgG level was <1:64 and his *B. henselae* IgG level was 1:128 (Table 1). Serial abdominal CT images continued to show improvement, and a repeat CTA (Fig. 1C) procedure performed 8 months after the initial imaging demonstrated significantly decreased aortic inflammation, with mild residual mural thickening extending from the origin of the superior mesenteric artery to the left common iliac artery, without significant enhancement or aneurysm.

We report a case of *B. quintana* aortitis in a man with AIDS. No other *B. quintana* aortitis cases in AIDS patients were identified in a review of the literature. Most causes of aortitis are autoimmune (e.g., giant cell arteritis, Takayasu's arteritis, rheumatoid arthritis, *HLA-B27* spondyloarthropathies, and antineutrophil cytoplasmic antibody [ANCA]-associated arthropathies) or idiopathic (2). Among the infectious causes, bacterial organisms such as *Salmonella*, *Staphylococcus*, and *Streptococcus* spp. are the most common (3). Luetic aortitis caused by *Treponema pallidum* is rare and usually involves the ascending aorta (4). Mycobacterial aortitis is uncommon in the developing world and usually involves erosion of the aortic wall by a contiguous lesion (5). There has been one case report of an immunocompromised elderly woman in whom serology confirmed the presence of *B. henselae* infrarenal abdominal aortic aneurysm and endocarditis (6). Another case report describes detection of *B. quintana* by 16S rRNA gene amplification in biopsied aortic tissue from a non-HIV-infected man who underwent abdominal aortic aneurysmal repair (7), but he had no risk factors for body lice exposure, exhibited no signs of infection, and had negative *Bartonella* serologies and Warthin-Starry stain results.

Three *Bartonella* species are the most common causes of human infection: *B. quintana*, *B. henselae*, and *B. bacilliformis* (8). Immunocompromised, HIV-infected patients develop *B. quintana* and *B. henselae* infections, with symptoms that include relapsing bacteremia, fever of unknown origin, endocarditis, and bacillary angiomatosis (BA). Although those two species have equal predilections to cause cutaneous BA in HIV-infected patients, *B. quintana* has a unique tropism for bone and heart valves, whereas *B. henselae* is more likely to infect liver, spleen, and lymph nodes (9). The origin of the aortic lesion in this patient is uncertain; without TEE, concomitant endocarditis was not completely eliminated. Another possible origin is extension of a contiguous vascular BA lesion into the aortic wall. One case report described *B. quintana* in a patient with AIDS manifesting as a single, large intra-abdominal BA lesion which eroded into the mesenteric vasculature (10). $CD4^+$ T cell counts were in the very low range at which BA occurs (9) for both the latter patient (10 cells/mm^3) and our patient (68 cells/mm^3).

Bartonella infections can be diagnosed (8) by histopathological examination of tissue with Warthin-Starry silver stain, by special *Bartonella* blood culture analysis (9) (unavailable in most hospital laboratories), by serological assays, and increasingly, by PCR amplification of DNA extracted from biopsy tissue, using specific or

broad-range primers (as in this case). IFA is currently the most accessible method of diagnosis, but there are a number of caveats with existing IFA tests. First, the *Bartonella* IgM test result is almost always negative even in acute cases in immunocompetent patients. Second, AIDS patients with profound immunocompromise often do not develop anti-*Bartonella* antibodies (11). Third, as seen in this case, there often is IFA cross-reactivity between *B. quintana* and *B. henselae* (11). Our patient had *B. quintana*, but his *B. henselae* titers were consistently higher than his *B. quintana* titers (Table 1). *Bartonella* titers of >1:800 have been shown to have a high positive predictive value for endocarditis (12); presumably, such titers would be predictive of other endovascular infections. Had *Bartonella* been considered a causative agent at clinical presentation (which it was not, based on patient history), serological testing might have obviated an invasive procedure for diagnosis.

There are no prospective studies of *Bartonella* treatment in immunocompromised hosts. Because of the high incidence of relapse after short courses of antibiotics, erythromycin or doxycycline treatment is recommended for a minimum duration of 3 to 6 months for severe infections (1). Of note, the initial clinical response to antimycobacterial treatment was most likely due to the administration of rifabutin and clarithromycin; both drugs are active against *Bartonella* species (8). Following IFA titers has been useful to ensure decrease with treatment; a subsequent severalfold titer increase can herald relapse (1).

Nucleotide sequence accession number. The amplified product including 263 nucleotides of the 16S rRNA gene of *B. quintana* was deposited in GenBank under accession number KR866081.

ACKNOWLEDGMENTS

We declare that we have no conflicts of interest. We have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Potential conflicts of interest that the editors considered relevant to the content of the manuscript have been disclosed.

J.E.K. received funding support from NIH grants R01AI52813 and R01AI103299 and a California HIV Research Program Award.

A.F.L. and S.A.L. provided direct patient care. M.B.C. and S.K.P. performed the interventional radiology (IR)-guided biopsy. M.A.O. performed cross-sectional imaging. G.M.B. and J.E.K. performed the *Bartonella* culture experiments. S.A.L. and J.E.K. wrote the manuscript.

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