

CONCISE REPORTS

Intra-articular co-infection by *Borrelia burgdorferi* and *Chlamydia trachomatis*

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Abstract

Objective—*Chlamydia trachomatis* and *Borrelia burgdorferi* infections are frequently the cause of unexplained oligoarthritis, as shown by identification of bacteria specific DNA in joint material from patients with reactive arthritis, Lyme arthritis, and undifferentiated oligoarthritis. The aim of this study was to determine whether the two organisms occur simultaneously in joint material from patients with arthritis.

Methods—Seventy six patients with unexplained arthritis were prospectively studied. Synovial fluid was obtained from all patients and examined for DNA from *C trachomatis* and *B burgdorferi* using specific polymerase chain reaction (PCR) protocols. Data concerning prior genitourinary infection or a history of tick bite were recorded and serum antibodies to *C trachomatis* and *B burgdorferi* were determined.

Results—Six patients (8%) had DNA from both *C trachomatis* and *B burgdorferi* in the same synovial fluid specimen (mean leucocyte count 11.925/mm³, 65% granulocytes). These patients (four men, two women; mean age 33.7 years) all had oligoarthritis of the knee, ankle, or both (mean disease duration 11.3 months). From the history and serological examination, four patients had some evidence of actual or previous infection with one or other of the bacteria, while the other two patients had a positive serological test for *Chlamydia* only.

Conclusions—DNA from two different microorganisms which are known to be triggering agents for arthritis may be present simultaneously in joint material from patients with unexplained oligoarthritis. This finding raises the question as to whether, in such cases, one or both bacteria contribute to the pathogenesis of the disease or whether they are only innocent bystanders.

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Chlamydia trachomatis and *Borrelia burgdorferi* are thought to cause arthritis by persisting intra-articularly in a metabolically active though usually a non-culturable state.¹

Polymerase chain reaction (PCR) is the method of choice for detecting persisting bacteria in joints.¹ Recently, universal PCRs have been developed for use in the diagnosis of undifferentiated arthritis. However, their sensitivity is lower than species specific PCRs and, to date, the universal PCR has not detected *B burgdorferi* in cases of undifferentiated arthritis.^{2,3} Nevertheless, universal PCR has detected several organisms simultaneously in one inflamed joint which have not been related to arthritis.^{2,3}

This study was undertaken to determine whether *C trachomatis* and *B burgdorferi*, the two most important arthritis triggering organisms in western countries,^{1,4} can be simultaneously detected by species specific PCR in unexplained oligoarthritis, or whether the detection of one of these bacteria excludes the presence of the other.

Methods

As part of a comprehensive study on early arthritis we prospectively studied 76 patients with unexplained oligoarthritis visiting a tertiary care outpatient clinic for the first time. The results of an extended rheumatological diagnostic programme in patients who fulfilled the criteria for unclassified arthritis will be published elsewhere. In this study patients with unexplained oligoarthritis are analysed.

Synovial fluid was obtained from all patients and examined for DNA from *C trachomatis* and *B burgdorferi* using species specific PCR protocols:⁵ *C trachomatis* nested PCR was used to target *C trachomatis* major outer membrane protein (MOMP) with 152 base pairs using the primer CT05/CT06//CT03/CT04⁶ and *Borrelia burgdorferi sensu lato* nested PCR was used to target *B burgdorferi* outer surface protein A (OspA) with 146 base pairs using the primer BBSL1/BBSL2//MRL7/MRL11a.^{7,8} Both tests were able to detect the specific bacteria with a sensitivity of one organism/ml synovial fluid. Numerous negative controls were included; PCRs were defined as positive only when the results were reproducible twice and when all negative controls remained negative. The correct identity of the PCR products was confirmed by restriction analysis or by direct sequencing of the PCR product.⁵

Data concerning prior infection of the genitourinary tract and a history of tick bite were recorded and serum antibodies for *C trachomatis* IgA/IgG (ELISA test; Medac, Wedel,

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Table 1 Characteristics of six patients with unexplained oligoarthritis PCR positive for both *Borrelia burgdorferi* and *Chlamydia trachomatis* DNA in the synovial fluid

	Patient no					
	1	2	3	4	5	6
Sex (male/female)	M	M	M	F	M	F
Age (years)	25	27	48	21	36	45
Affected joints (knee/ankle)	k	k/k/a	k/k	k	a/a	k/k
Duration (months)	9	18	30	2	1	8
Inflammatory low back pain ⁹	+	-	+	-	+	-
HLA-B27	+	-	-	-	+	-
Leucocytes in SF (n/mm ³)	23250	6450	10400	13150	14700	3600
Neutrophils in SF (%)	88	33	49	85	67	70
History of tick bite/ECM	-/-	+/-	-/-	-/-	+/+	-/-
<i>B burgdorferi</i> serology	+	+	-	-	-	+
History of urogenital infection	-	-	-	-	+	-
<i>C trachomatis</i> in urogenital smears*	-	+	-	-	-	+
<i>Chlamydia</i> serology IgA/IgG (ELISA)	-/+	+/+	-/+	-/+	-/-	-/-

SF = synovial fluid; ECM = erythema chronicum migrans.

*Detected by immunofluorescence or culture.

Germany) and *B burgdorferi* (haemagglutination test; Labor Diagnostik, Heiden, Germany) were determined. For detection of active urogenital chlamydial infection urethral smears were taken and examined by immunofluorescence or culture.

Results

DNA from either *C trachomatis* or *B burgdorferi* was present in synovial fluid from 22 of the 76 patients (29%)⁵; the results of these patients will be published elsewhere (Schnarr *et al*, submitted). Surprisingly, in a further six patients (8%), DNA from both *C trachomatis* and *B burgdorferi* was reproducibly detected in the same synovial fluid specimen. Table 1 shows the individual characteristics of the "double positive patients" (four men, two women; mean age 33.7 years), all of whom had oligoarthritis of the knee, ankle, or both (mean disease duration 11.3 months). Three of the six patients (two positive, one negative for HLA-B27) also complained of inflammatory low back pain.⁹ With regard to their history and the results of serological tests, two patients (nos 3 and 4) had positive serological tests for *Chlamydia* only. Four patients (nos 1, 2, 5, and 6) had some evidence of ongoing or previous infection with both organisms: patient 5 had had erythema chronicum migrans and a urogenital infection in the past, and patients 2 and 6 had active asymptomatic urogenital infection with *C trachomatis* at the time of the investigation.

Discussion

We describe for the first time the simultaneous detection in the synovial fluid from inflamed joints of DNA from *C trachomatis* and *B burgdorferi*, two bacterial species which have been typically associated with the pathogenetic process leading to arthritis.

Molecular biological techniques such as PCR are increasingly being used to identify a potential bacterial aetiology in arthritic diseases.¹ In general, detection of specific bacterial DNA or RNA in the inflamed joint (synovial fluid or synovial tissue) is considered evidence for a bacterial aetiology. However, recent publications using broad range or specific PCRs²⁻³ have identified DNA from organisms not previously related to arthritis. It was speculated

that monocytes that have phagocytosed bacteria elsewhere in the body may have been disseminated into the inflamed joint. Therefore, because of its extreme sensitivity, a positive PCR result may reveal a previously unexpected degree of intrinsic microbial presence at many anatomical sites.² Recently, Schumacher *et al* reported the detection of *C trachomatis* in healthy joints¹⁰ and in the joints of patients with diseases not related to *C trachomatis* such as osteoarthritis,¹¹ which suggests that the presence of *C trachomatis* in joints does not necessarily indicate that *C trachomatis* is the causative factor. Furthermore, they noted several patients with different forms of arthritis in whom DNA from *C trachomatis* as well as from *C pneumoniae* was found in the synovial tissue of the same individual.¹²

The detection of two known arthritis causing microorganisms in one inflamed joint sets the scene for several possible pathogenetic models. Either both organisms are causing inflammation, or one organism is triggering the disease and the second is an innocent bystander disseminated into the inflamed joint by increased vascularisation, or none of them is responsible for the disease. The finding of two pathogenic organisms at the disease site does not primarily indicate that they are both innocent bystanders; numerous examples of co-infection, either bacterial or viral, are known in medicine.¹³ In four of the six patients reported here, additional evidence such as coincidental history, serological tests, and urogenital smears for *C trachomatis* indicated co-infection by both *B burgdorferi* and *C trachomatis*. Some patients were seronegative for *B burgdorferi* or *C trachomatis*, or both. This may be because *C trachomatis* is an intracellular agent or because of the relatively low sensitivity of the serological test used for *B burgdorferi*. Furthermore, most patients (nos 1, 2, 3, and 6) had already been treated with steroids before serological testing. To examine the pathogenesis and define the precise causative role of these organisms, further analysis of the host-bacteria interaction such as specific T cell responses, gene induction, and cytokine profiles need to be studied.

Antibiotic treatment should be reserved for patients with chlamydial infections of the urogenital tract, those with arthritis in whom *B burgdorferi* DNA is detected in the inflamed joint, or those with unambiguously positive *B burgdorferi* serological tests. No benefit of antibiotic treatment has so far been reported for patients in whom *C trachomatis* DNA has been detected in the joints.¹⁴

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