Detection of *Borrelia burgdorferi* by species-specific and broad-range PCR of synovial fluid and synovial tissue of Lyme arthritis patients before and after antibiotic treatment

Sirs,

Lyme disease is caused by the tick-borne spirochete *Borrelia burgdorferi* (1). Due to the variation in clinical presentation and because of the limitations of currently available diagnostic assays it is often difficult to diagnose Lyme arthritis. *B. burgdorferi* could only be cultured from synovial fluid in a minority of patients (2). However, bacterial antigens and DNA of *B. burgdorferi* have been demonstrated in the synovial fluid (SF) and synovial tissue (ST) of patients with Lyme arthritis by immunological methods and the polymerase chain reaction (PCR). After antibiotic treatment PCR usually became negative (3). We therefore studied the sensitivity of species-specific and broad-range PCR in SF and ST samples of Lyme arthritis patients before and two months after treatment.

Samples from 11 Lyme arthritis patients and from 15 controls with the diagnosis of rheumatoid arthritis (n = 10) or calcium pyrophosphate dihydrate deposition disease (n = 5) were analyzed for bacterial DNA by broad-range PCR of synovial ECM before IgM IgG  IgM IgG SF ST PCR PCR and after antibiotic treatment because of the limitations of currently available diagnostic assays it is often difficult to diagnose Lyme arthritis and bacterial meningitis. *B. burgdorferi* DNA was detected in 9 out of 11 patients (82%) with Lyme arthritis by species-specific PCR (SF 82%; ST 18%). Only 1 patient tested positive by broad-range PCR and the *B. burgdorferi* specific sequence was confirmed. PCR results became negative after treatment except for one SF specimen. SF obtained from this patient 6 months after antibiotic treatment demonstrated negative PCR without any additional therapy. ST and SF samples from control patients were negative at all time points. Synovitis resolved in all patients between 2 (n = 3) and 6 months after the diagnosis and initiation of treatment.

Our data support the conclusions of Nocton et al. (3) and Limbach et al. (4) with respect to the high sensitivity of the *B. burgdorferi* specific PCR in Lyme disease. Furthermore, we found a higher sensitivity with SF compared to ST. Conflicting findings by Jaulhac et al. (5) could be due to several reasons (6). Target imbalance as described by Persing et al. (7) seems an unlikely explanation for this phenomenon since the former study used the same PCR target as ours. In agreement with previous studies the sensitivity of broad-range PCR was low (8, 9). Unlike recently reported (10), persistence of *B. burgdorferi* DNA was not observed after antibiotic treatment in our patients. SF seems the preferable specimen to detect *B. burgdorferi* by PCR. Antibiotic therapy with ceftriaxone (2 weeks) or doxycycline (4 weeks) usually resulted in complete cure of Lyme arthritis and species-specific PCR proved a helpful tool in monitoring the treatment success.

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8. Cox C, Kempf K, Gaston J: Detection and characterisation of bacteria in synovial fluid samples from reactive arthritis patients

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Table 1. Clinical data, laboratory parameters and pre-and post-treatment results of species specific PCR for *Borrelia burgdorferi* and treatment modality of 11 patients with Lyme arthritis.

<table>
<thead>
<tr>
<th>Patient with Lyme arthritis</th>
<th>Sex</th>
<th>Age</th>
<th>History of tick bite or ECM</th>
<th>Present. of arthritis</th>
<th>Duration of arthritis before [mos]</th>
<th>Synovitis</th>
<th>CRP [mg/l]</th>
<th>Cell count in SF [x10°/l]</th>
<th>ELISA</th>
<th>Western blot</th>
<th>Culture</th>
<th>Broad-range PCR</th>
<th>PCR results</th>
<th>AB</th>
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<tbody>
<tr>
<td>1 m 26 ECM M 3 + + + - 5</td>
<td>4.5</td>
<td>pos.</td>
<td>pos.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>n.a.</td>
<td>n.a.</td>
<td>DX</td>
<td></td>
<td></td>
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<tr>
<td>2 f 37 ECM M 8 + - + - 4</td>
<td>9.4</td>
<td>neg.</td>
<td>pos.</td>
<td>n.a.</td>
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<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>pos.</td>
<td>pos.</td>
<td>DX</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3 m 62 ECM M 6 + - + - 12</td>
<td>12.7</td>
<td>pos.</td>
<td>pos.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>pos.</td>
<td>neg.</td>
<td>n.a.</td>
<td>n.a.</td>
<td>CF</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4 m 31 - M 2 + + - + 6</td>
<td>9.0</td>
<td>neg.</td>
<td>pos.</td>
<td>n.a.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>n.a.</td>
<td>CF</td>
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<tr>
<td>5 f 48 TB M 36 + + - 18</td>
<td>13.3</td>
<td>neg.</td>
<td>pos.</td>
<td>n.a.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>n.a.</td>
<td>DX</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6 f 53 - M 3 + + + - 100</td>
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<td>neg.</td>
<td>pos.</td>
<td>n.a.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>n.a.</td>
<td>CF</td>
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<td></td>
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<tr>
<td>7 m 63 TB M 0.1 + + + - 129</td>
<td>42.0</td>
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<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>pos.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>CF</td>
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<tr>
<td>8 m 39 ECM M 10 + + + -</td>
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<td>pos.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>pos.</td>
<td>neg.</td>
<td>DX</td>
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<tr>
<td>9 m 24 TB O 15 + + + -</td>
<td>107</td>
<td>pos.</td>
<td>pos.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
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<td>neg.</td>
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<td>n.a.</td>
<td>CF</td>
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<td>10 f 26 - M 11 + + + - 28</td>
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<td>n.a.</td>
<td>n.a.</td>
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<td></td>
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<tr>
<td>11 m 64 TB O 14 + + + - 100</td>
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<td>pos.</td>
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<td>neg.</td>
<td>neg.</td>
<td>neg.</td>
<td>pos.</td>
<td>pos.</td>
<td>neg.</td>
<td>neg.</td>
<td>CF</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

n.a. not available; AB: antibiotics; DX: Doxycyclin (Vibramycin) 2 x 100 g/d po, 4 weeks; CF: Ceftriaxon (Rocephin®) 1 x 2 g/d i.v, 2 weeks.

Presentation of arthritis: M = monoarthritis of the knee; O: oligoarthritis involving the knee; synovitis of the knee: (+) present, (-) absent.
Letters to the Editor


9. WILKINSON N et al.: The detection of DNA from a range of bacterial species in the joints of patients with a variety of arthritides using a nested, broad-range polymerase chain reaction. Rheumatology 1999; 38: 260-6.


Efficacy of thalidomide in refractory adult Still’s disease: A new case report

Sirs,

Adult Still’s disease is a febrile disorder of unknown etiology which affects young adults during the second or third decade, presenting either as a primary disease or as a relapse of childhood onset Still’s disease (1). Clinical features are characterized by an initial pharyngitis with prolonged fever, polyarthralgia and maculopapular rash; liver and pericardium involvement are commonly observed (2). The biological data comprise a major increase in all inflammatory proteins and leukocytosis usually >20,000 cells/mm³. The poor prognosis of Still’s disease is determined by the long time course of its evolution, the frequent relapses, and serious complications [thrombotic thrombocytopenic purpura (3), amyloidosis (4)]. Levels of tumor necrosis factor α (TNFα), like the levels of other inflammatory markers (C reactive protein, ferritin), increase dramatically in patients with Still’s disease. TNFα seems to play a role in the pathogenesis of Still’s disease and therefore some anti-TNFα drugs, such as infliximab (5) and etanercept (6), have been used with success in the treatment of adult Still’s disease. Thalidomide is also known to inhibit TNFα production in human monocytes by stimulating mRNA degradation in a dose-dependent fashion and to reduce the half-life of TNFα mRNA by 50% (11). Stambe et al. (12) reported the efficacy of thalidomide in a 44-year-old woman with adult onset Still’s disease refractory to other treatments. The patient was therefore given thalidomide (200 mg/day, then 100 mg/day). Improvement was observed after 2 weeks of treatment (thalidomide + prednisone) and the disease was in remission during a 6-month follow-up period. During the study, the percentage of monocytes producing TNFα decreased from 42.7% before thalidomide to 1.4% during the treatment.

Our results confirm the efficacy of thalidomide in adult Still’s disease, even at a dosage of 150 mg/day. This efficacy is particularly remarkable when one considers the cost-effectiveness of this drug in comparison to other anti-TNFα drugs. Moreover, no significant side effects were observed.

In conclusion, thalidomide seems to offer a new approach for the treatment inflammatory diseases linked to the overproduction of TNFα. Refractory adult Still’s disease might be a new indication for thalidomide as long as careful monitoring of peripheral nerve conduction with electromyograms every 6 months is conducted to detect asymptomatic neuropathy. In fertile women the use of thalidomide should be authorized only in women taking the birth control pill.

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References


