Chapter 6

Prevalence and clinical presentation of Lyme arthritis in a large cohort of patients with recent-onset arthritis

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Abstract

As the value of performing *Borrelia burgdorferi* serology in recent-onset arthritis patients is unknown, we aimed to evaluate the prevalence of positive Lyme serology among patients with recent-onset arthritis in relation to their long-term disease outcome.
Serologic testing for *B. burgdorferi* was performed in 1180 consecutive patients presenting with recent-onset arthritis. Serology was performed using an Enzyme Immune Essay (EIA) and confirmation was done by an immunoblot. The clinical diagnosis at presentation and follow up during the disease course was studied in medical files of the patients with positive or unequivocal lab results. Subsequently, the proportion of patients with Lyme arthritis was determined, as well as the diagnostic accuracy of *B. burgdorferi* serology.

Of 1180 patients with recent-onset arthritis, 53 patients had positive EIA which was confirmed by a positive or equivocal immunoblot, indicating a seroprevalence of of 2.0- 4.5%. Nine cases of definite LA were identified, indicating a prevalence of 0.8% within a recent-onset arthritis cohort. These patients were characterized by younger age, and typically presented with involvement of at least one knee. Four patients were *B. burgdorferi* seropositive and did not have a clear other diagnosis. However, there was no indication that LA presented with other primary clinical manifestations than arthritis with involvement of large joints. The remaining *B. burgdorferi* seropositive patients had a clear different clinical diagnosis. Based on the low prevalence of LA, positive predictive value (PPV) of serologic testing was low; however by selecting patients with a high clinical suspicion, eg in patients presenting with an oligo- or monoarthritis of large joints, the PPV increased to 67%.
The present data advocates performing *B. burgdorferi* serology testing only in patients with an increased clinical suspicion, and not in all patients presenting with recent-onset arthritis.

Introduction

Early identification and prognostication of early arthritis patients has been shown relevant and is associated with an improved disease outcome as treatment strategies can be initiated early. Lyme arthritis is an uncommon form of early arthritis that is curable.

Lyme arthritis is one of the clinical manifestations of Lyme Borreliosis (LB), caused by *Borrelia burgdorferi* sensu lato (sl). There are several species of *Borrelia burgdorferi* sl that can cause disease in humans: *B. burgdorferi* ss, *B. afzelli*, *B. garinii*, *B. bavariensis* and *B. spielmanii* [568]. *B. burgdorferi* ss is the only species prevalent in North America, while in Eurasia all abovementioned species can be prevalent [35]. Different species are associated with different clinical manifestations: *B. burgdorferi* ss in particular is associated with Lyme arthritis (LA), but the species prevalent in Eurasia can also cause an arthritis. A report on 1471 patients in Sweden with a definite diagnose of Lyme disease showed that 7% of all patients had arthritis with arthritis being the sole manifestation in 4.4%, [430] while data from North-America indicate that up to 60% of untreated infected patients develop arthritis [150]. LA can present 12-50 weeks after a recorded tick bite and is considered a late manifestation of LB. The classical clinical manifestation of LA comprises mono- or oligo-artritis of the large joints, most often the knee, sometimes typically preceded by an erythema migrans (EM). Nevertheless, also elbows, ankles and hips can be affected and polyarthritis is described in up to10 % of cases complicating the differentiation from other forms of recent-onset arthritis [569].

Cultivation of *B. burgdorferi* from a patient's skin or blood is the gold standard for demonstration of active infection, but it is time consuming and lacks clinical sensitivity [235]. Serological testing is the most commonly applied technique to support of the diagnosis of LB. However serological tests for *B. burgdorferi* can not distinguish past from present infection. For diagnosis of Lyme arthritis guidelines recommend a 2-tier testing approach starting with IgG enzyme immune assay (EIA), followed by IgG immunoblot to confirm the result [109, 416, 570]. As LA is a late manifestation and a symptom of disseminated disease, sensitivity of serologic testing for anti-borrelia IgG is generally very high in patients with LA. However, in areas endemic for Lyme disease the combination of C6 EIA and immunoblot can be positive as well in 64-79% of patients with previous Lyme disease and in 1-3% of healthy subjects. Experts underline the importance of taking into account the a priori chance of active *B. burgdorferi* infection and the estimated specificity of *B. burgdorferi* serology in the subject of interest [234, 571].

It is unclear whether *B. burgdorferi* serological testing should be applied to all patients with an recent-onset arthritis. A Swedish study indicated that positive serological results, though helpful in diagnosing Lyme arthritis, can also be found in patients with definite other rheumatological diagnoses and thus can be false positive. Furthermore, we hypothesized that the clinical presentation of Lyme arthritis in Europe may be less characteristic as several species other than *B. burgdorferi* ss are prevalent [35]. Previous studies generally diagnosed Lyme based on the classical presentation in combination with positive Lyme serology [112, 116, 117]. By this approach patients with an atypical presentation of LA may have been missed.

With the current study we aimed to evaluate the prevalence of positive *B. burgdorferi* serology among patients with recent- arthritis and to appreciate the value of serological testing in these patients by studying the relation between the test results and the disease course over a follow-up of several years to determine the final diagnosis.

Our second aim was to evaluate the contribution of serologic testing to the clinical diagnosis of LA. We studied the Leiden Early Arthritis Clinic (EAC), a large inception cohort enrolling all consecutive patients presenting with recent-onset arthritis. In 1180 early arthritis patients' clinical evaluation and serologic testing for *B. burgdorferi* was performed.

Patients and methods

Study population

Patients were included in the Leiden Early Arthritis Clinic cohort, a large inception cohort initiated in 1993 that enrolled consecutive patients presenting with arthritis to the Leiden Rheumatologic outpatient clinic and that had symptom duration less than 2 years. A detailed description is provided in previous reports [572]. At the first visit, the rheumatologist completed a questionnaire regarding the presenting symptoms, symptoms, this included information on skin manifestations and tick bites. Physical examination was performed and blood samples were taken for routine diagnostic laboratory screening. Follow-up visits were performed on a yearly basis. Written informed consent was obtained from all participants. This study was approved by the local Medical Ethics Committee.

The present study included all patients who presented to the EAC during the period of February 1993 to April 1997 (cohort 1) and the period July 2003 to June 2008 (cohort 2). For patients of cohort 1 serology for common infectious causes of arthritis including *B. burgdorferi* was performed routinely at inclusion;

for patients in cohort 2 *B. burgdorferi* serology was performed on prospectively stored serum samples.

B. burgdorferi serology

In sera from cohort 1, antibodies against *Borreliae* were detected by the IgG and IgM flagellin-EIA (Dako Cytomation, Glostrup, Denmark), using the procedure recommended by the manufacturer. All available positive samples were retrospectively retested with the IgG/IgM C6 Lyme EIA Kit (Immunetics, Boston, USA) according to manufacturers' protocol. In cohort 2, the C6 Lyme EIA Kit was used on prospectively stored serum samples taken for the EAC cohort. All positive enzyme immunoassay (EIA) results from both cohorts were confirmed by the *Borrelia* Europe LINE blot (Virotech GmbH, Rüsselsheim, Germany) and a second EIA, the Liaison® *Borrelia burgdorferi* IgG (VlsE) and IgM (VlsE+OspC) (Diasorin, Italy), both according to the manufacturers' protocols.

Evaluation of serologic test results

The results of current *B. burgdorferi* serologic testing strategies among recent-onset arthritis patients were evaluated in cohort 2. First, serologic results were interpreted by an experienced microbiologist, unaware of clinical presentation and final diagnosis. Serologic test results were classified as: A. Positive, very suspect for active or recent infection, defined as a C6 Lyme index (IgG/IgM) > 5 and a Liaison VlsE IgG value > 40, combined with a positive IgG immunoblot, B: Positive, all other confirmed immunoblot positive serologic results not fitting the abovementioned criteria most likely fitting an old, cleared infection, C: Inconclusive/equivocal immunoblot, combined with a positive/equivocal EIA; a consecutive serum would be necessary to make an adequate serological diagnosis. However, as LA is a late manifestation this serologic classification is most likely not compatible with LA and category D: Negative EIA screening serology.

Clinical diagnosis of Lyme arthritis

Clinical records of all patients with positive or inconclusive serology were traced and the data obtained at the time of the initial diagnosis and during the disease course were thoroughly studied by two experienced rheumatologists (JdVB, AHM). The final diagnosis was classified in two groups; if another clinical diagnosis was certain this was classified as "No LA" and all other cases were classified as "Possible LA".

Based on the combination of clinical data and serologic results patients were reclassified into three LA categories. The first category were patients with a

"definite LA", the second category of patients were patients with a "possible LA", and the third category were patients with "No LA".
For further analyses on the prevalence and clinical characteristics of Lyme arthritis only patients with positive C6-peptide EIA confirmed by blot and a clinical diagnosis of Lyme arthritis were considered.

Statistical analysis

Statistical analysis was performed using a statistical software package (SPSS for windows, version 17.0). Baseline characteristics were compared between patients with and those without definite diagnosis of LA as determined prospectively by the treating physician. Positive predictive value and negative predictive value of *B. burgdorferi* serology was determined for patients in the EAC.

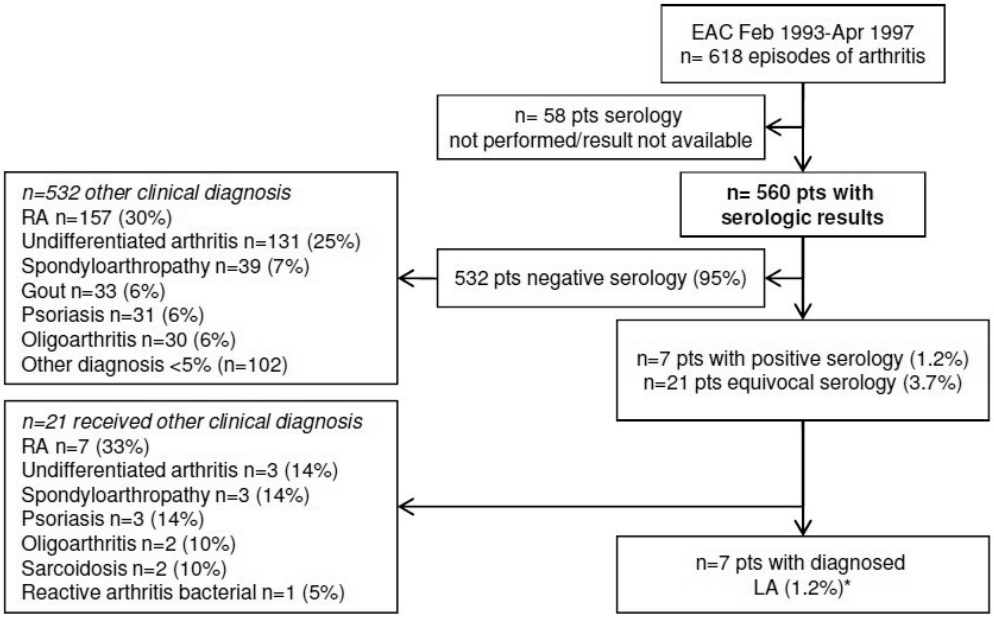

Figure 1: Serological results from cohort 1. * 6 pts could be confirmed with the C6-peptide EIA, the C6-negative patient had an equivocal immunoblot.

Results

Prevalence of positive *B. burgdorferi* serology in early arthritis patients

Cohort 1 consisted of 618 patients consecutively included between 1993 and 1997. Serology results were available for 560 patients. Seven patients had a positive EIA confirmed by a positive immunoblot (1.2%), twenty-one patients

had a positive flagellin EIA with a inconclusive immunoblot (3.7%). Seven of the 28 serologically positive or equivocal patients were diagnosed as Lyme arthritis by their treating rheumatologist (Figure 1). Twenty-three of the 28 serologically positive or equivocal patients could be retrospectively retested in the C6-peptide EIA. In six patients (1.1%) the C6-peptide EIA was also positive; these patients were all diagnosed with Lyme arthritis with a positive immunoblot. The one patient that was diagnosed as LA but had a negative C6-peptide EIA had an equivocal immunoblot. She presented with arthritis of the elbow that resolved after antibiotic treatment. This could have been a LA, but because of the absence of sufficient IgG in the immunoblot it is unlikely, after treatment there was no serological follow up. One patient had a positive immunoblot, but a negative C6-peptide EIA, clinically she was diagnosed as an spondylarthopathy. Serology for this patient can be consistent with a previously cleared infection [591].

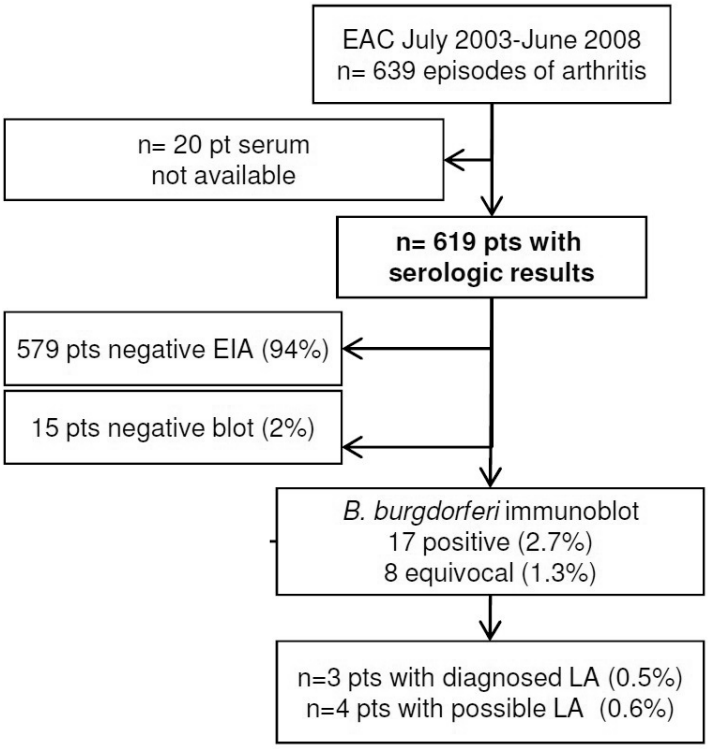

Figure 2: Serological results from cohort 2.

Cohort 2 consisted of 639 patients included between 2003 and 2008. Serum was available of 619 patients. Of these, 40 were reactive in the C6-peptide EIA, which was confirmed by a positive immunoblot in 17 patients, yielding a

seroprevalence of 2.7 %. (Figure 2). The immunoblot was equivocal in 8 patients (1.3%).

Prevalence of Lyme arthritis

The prevalence of definite LA in cohort 1 was 6 of 560 included patients (1.1%). Characteristics and outcome of the 25 patients from cohort 2 with positive C6-peptide EIA and positive or equivocal immunoblot are shown in table 1. Three patients (12%) were diagnosed at presentation with LA and treated with antibiotics, which had resulted in resolution of arthritis. One patient with positive serology was treated with antibiotics without any improvement of the MCP's; afterwards the clinical diagnosis was set at RA and he achieved remission on DMARD therapy. In the remaining 21 patients the diagnoses were recorded by the treating rheumatologist (See table 1). In four patients with other recorded diagnosis and positive or equivocal serology for Lyme, LA could have been considered. These patients were RF/ACPA seronegative and did not have any other typical symptoms like psoriasis, dactylitis or gut disease. One (case 4) of four also had erosive damage on X-ray. These four patients achieved remission and were discharged without any antibiotic treatment. In the natural course of LA natural remission, but also progression to erosive disease has been described [155]. Case 7 responded to treatment with prednisone for arthritis of the hands. There was no progression of disease, making LA unlikely. The prevalence of LA in cohort 2 was 0.5% of definite LA to 1.1% when combined with the possible LA cases.

Diagnostic performance of B. burgdorferi serology

Diagnostic performance of third generation EIA and immunoblot was only studied in cohort 2 because in this cohort all patients were tested with a C6-peptide EIA and immunoblot. Taking into account that in 3 cases a definite diagnosis of LA and 4 cases a possible diagnosis of LA was made, overall specificity of *B. burgdorferi* serology for the clinical diagnosis was 96-97%, due to the low prevalence of Lyme disease. LA is considered a late manifestation of an infection with *B. burgdorferi* and serology should be unequivocally positive. If patients with equivocal blood results (serologic category C) were regarded as negative for active *B. burgdorferi* infection, then serologic specificity increased to 98%. Given the prevalence of LA among this second cohort (0.5-1.1%), the positive predictive value of positive *B. burgdorferi* serology was very low (12-28%).

Pt	Clinical category	Serol. categ.	Lyme category	Diag-nosis	Tick bite	Month started	RF pos.	ACPA pos.	#swollen joints	Involved joints	Current status
1	Possible	A	Definite	LA	no	okt	no	NA	1	knee	minimal hydrops after AB po and iv
2	Possible	A	Definite	LA	no	okt	no	no	1	knees, alternating	remission after AB
3	No LA*	A	Definite	RA	yes	okt	yes	yes	9	knee, MCPs	remission after AB of knee; treatment for RA
4	Possible	A	Possible	OA	no	jan	yes	NA	1	knee, ankle	persistent OA hands and hydrops knee; after first visit no ankle complaints anymore
5	Possible	B	Possible	OA	NA	okt	no	NA	1	knee	OA with hydrops one knee
6	Possible	C	Possible	OA	NA	jan	no	NA	1	knee	OA one knee
7	Possible	A	Possible	UA	no	jul	no	no	9	MCP, PIP,MTP	remission after prednisone (SAVE-trial)
8	No LA	A	No LA	UA	no	jun	yes	no	10	MCPs PIPs	no FU after 2005
9	No LA	A	No LA	RA	no	UK	yes	no	3	MCP,sc	remission with methotrexate
10	No LA	A	No LA	PA	no	sep	no	no	2	MCP, PIP	remission without medication
11	No LA	A	No LA	Gout	NA	okt	yes	no	14	MCP, PIP	gout; remission with allopurinol
12	No LA	A	No LA	RA	no	apr	no	no	12	wrists, MCP, PIP	remission
13	No LA	A	No LA	UA	NA	UK	no	no	6	PIP, MCP	Psoriasis
14	No LA	B	No LA	PA	UK	okt	yes	NA	6	MCP	Psoriasis
15	No LA	B	No LA	SLE	no	jan	no	NA	3	MCP, MTP	SLE with nefritis; currently in remission
16	No LA	B	No LA	RA	no	jan	yes	yes	14	MCP, wrist MTP	RA with methotrexate
17	No LA	B	No LA	RA	NA	dec	yes	NA	10	PIP,MTP,MCP	active RA
18	No LA	B	No LA	RA	yes	jan	no	no	6	PIPs	UCTD/SLE overlap with leucopenia, elevated liverenzymes, livido reticularis and ANF+
19	No LA	C	No LA	SA	NA	mar	no	NA	7	DIP, PIP, MTP	IBD
20	No LA	C	No LA	RA	NA	may	yes	yes			remission with methotrexate
21	No LA	C	No LA	RA	NA	sept	yes	no	8	PIP, MCP	RA with methotrexate
22	No LA	C	No LA	RA	NA	UK	no	NA	14	elbows, wrists, knees, MCP, MTP	remission
23	No LA	C	No LA	OA	NA	jan	no	NA	1	PIP	no FU
24	No LA	C	No LA	RA	NA	mar	yes	yes	4	MCP, PIP	moderately active RA
25	No LA	C	No LA	RA	NA	sep	yes	yes	10	MCP, MTP	no FU after 2007

Table 1: Clinical characteristics of *B. burgdorferi* serologically positive/equivocal patients. Lyme arthritis (LA) rheumatoid arthritis (RA), oligoarthritis (OA), undifferentiated arthritis (UA), psoriatic arthritis (PA), spondylarthropathy (SA), undifferentiated connective tissue disease (UCTD), inflammatory bowel disease (IBD). UK: unknown, NA: not applicable, FU; follow-up. * patient presented with arthritis of the knee and hands and was treated as LA accordingly; the symptoms of the knee disappeared but the second diagnosis RA remained.

Lyme arthritis: clinical presentation in an early arthritis cohort

Among the total number of 1180 patients studied, a definite diagnosis of Lyme arthritis was made in 9 patients (0.8%). Of 1180 patients with recent-onset arthritis, 53 patients had a positive EIA which was confirmed by a positive (24pts) or equivocal (29pts) blot, indicating a seroprevalence of 2.0- 4.5%. Characteristics of the patients with and without Lyme arthritis are presented in Table 2. As compared to other arthritis patients, patients with Lyme arthritis were younger, and had lower swollen joint count. The distribution of inflamed joints was more often asymmetric including almost always one (n=8) or both (n=1) knee(s). In one patient both wrists, MCPs and PIPs of both hands were also affected in addition to arthritis of one knee. Interestingly, this patient was initially diagnosed as Lyme arthritis and treated with antibiotics which resulted in improvement of the knee arthritis, but not of that of the other joints. Because of persisting arthritis of the wrists and small hand joints during several months, an additional diagnosis of rheumatoid arthritis was made and currently he is still being treated with DMARDs with good clinical response.

	Patients with Lyme arthritis (n=9)	**Patients with other diagnoses (n= 1170)**	**P-value**
Female, n (%)	5 (56)	691 (59)	0.834
Age, median (IQR)	26 (22-48)	52 (38-65)	**0.002**
Rheumatoid factor positive, n (%)•	1 (11)	299 (27)	0.274
ACPA positive, n (%)	1 (13)	235 (28)	
Additional symptoms			
none, n (%)	5 (56)	475 (43)	0.441
systemic, n (%)	3 (33)	417 (38)	0.794
Swollen joint count, median (IQR)	1 (1-2)	4 (1-8)	0.066
Affected joints at start of symptoms#			
Large joints only	8 (89)	365 (32)	**0.004**
Symmetric distribution	1 (11)	552 (50)	0.115
Lower extremities only ⁑	7 (78)	303 (27)	**0.015**
Upper extremities only⁑	0 (0)	403 (36)	
CRP, median (IQR)β	12 (5-25)	13 (4-33)	0.374
ESR, median (IQR)	18(14-30)	26 (11-46)	0.198
Duration of symptoms in weeks, median (IQR)	9 (4-21)	13(5-29)	0.637
VAS morning stiffness, median (IQR)	10 (4-31)	50 (20-73)	0.009
VAS pain, median (IQR)	12 (7-52)	48 (28-64)	0.018

Table 2: Baseline characteristics of patients with definite diagnosis of Lyme arthritis (n=9) compared to patients with other diagnosis

By selecting only patients with high clinical suspicion for LA (oligoarthritis including a knee and < 50 years) from all tested patients form cohort 1 and 2, the a priori chance of LA increased from 0.8% to 6.4% (n = 125 out of n = 1180) with improvement of PPV (42-85%). Of all patients with definite LA (n=9), two reported skin abnormalities: one an erythema, possibly erythema migrans (not confirmed by a physician) and one nodulosis. Of all patients included in the study 35 (3%) reported a tick bite in the past, in two of these patients a clinical diagnosis of Lyme arthritis was made. Hence the majority of patients with definite diagnosis of LA (n=7 of 9 pts) did not remember any tick bite. Selecting patients with a tick bite in the past and oligoarthritis increased a priori chance of LA to 10%; however, the majority of patients with a diagnosis of LA did not remember any tick bite, therefore this is not a sensitive marker.

Discussion

Lyme arthritis (LA) is relevant to identify because the disease should be treated with antibiotic therapy. *B. burgdorferi* serologic testing is available to aid in the diagnosis, but interpretation of serology can be difficult as also patients with previous Lyme disease can have positive serology. Among this large cohort of patients with recent-onset arthritis seroprevalence of Lyme antibodies was 2.0-4.5%; incidence of definite Lyme arthritis was 0.8%. The positive predictive value for LA of *B. burgdorferi* seropositivity was very low in the recent-onset arthritis population (≤28%). Due to the low prevalence of Lyme arthritis serologic test performance improves markedly by preselecting patients with high clinical suspicion.

Most studies concerning Lyme disease have focused on describing the full clinical spectrum of the disease by selecting all patients with typical presentation and positive serology. By this approach, theoretically, patients with atypical presentation could have been overlooked. In our study, serology for *B. burgdorferi* antibodies was performed on all patients presenting with recent-onset arthritis. Patients with definite diagnosis of LA, defined by positive serology and clinical diagnosis of LA, were characterized by younger age, and typically had arthritis of one or both knees. Four *B. burgdorferi* seropositive patients did not have a clear other diagnosis. The clinician did not perform serology at the first visit of the outpatient clinic. All patients were discharged without antibiotic treatment and complaints resolved spontaneously. In the natural course of LA natural remission, but also progression to erosive disease has been described [155]. It is also possible that these patients recently suffered from LA that had naturally resolved when the visit to the outpatient clinic took place. Two patients had hydrops of the knee with a low cell count that resolved

over time. From this cohort of 1180 patients of early onset arthritis we conclude that it is unlikely that LA presents with other symptoms than arthritis with involvement of the large joints. Thorough examination of clinical charts of patients with positive serology but no other clinical diagnosis did not give arguments for atypical patterns of Lyme arthritis. Only one patient in the possible LA group did not have arthritis of the large joints, and this patient responded well to prednisone treatment, without progression of disease making a LA less likely.
Main drawback of this study is the lack of a gold standard to confirm the diagnosis Lyme arthritis. However, this problem is inherent to the diagnostic testing of Lyme in general as also culturing of *B. burgdorferi* species and PCR on synovial fluid lack sufficient sensitivity and specificity respectively. In fact, this advocates evaluation of performance of serologic testing in patients with recent-onset arthritis. By using the Leiden EAC cohort we had the possibility to check clinical records for a long follow-up period, confirming our data on final clinical diagnosis.

The frequency of LA is comparable between the first cohort (1993-1997; 1.1%) and the second cohort (2003-2008; 0.5-1.1%). It is striking though that the incidence of LB in Europe seems to be gradually increasing the last decades, while we find a stabile of decreasing amount of LA presenting to rheumatologists over the years [118-120]. The higher seroprevalence in the absence of LA in the second cohort can be due to more old *B. burgdorferi* infections. Possibly, growing awareness of Lyme disease resulted in more frequent antibiotic treatment in case of tick bites or reported skin abnormalities.
For the Netherlands, no exact data on seroprevalence among the population are known. The CBO guidelines refer to data from personal communication indicating that seroprevalence varies between 4 and 8%, but seroprevalence is highly influenced by geographic area and population of interest. For example, among Dutch foresters seroprevalence was as high as 20% [322]. In our population the seroprevalence was 2.0-4.5%.
Frequency of LA was low in our cohorts. As Lyme arthritis is considered a manifestation of late, disseminated Lyme disease sensitivity of a serologic test approaches 100%. Based on the low frequency of Lyme arthritis, negative predictive value (NPV) is consistently high, but PPV only contributes significantly for patients with high clinical suspicion of Lyme disease. Most patients in whom Lyme arthritis was confirmed did not remember any tick bite nor a skin lesion, making these characteristics less useful for determination of a priori chance of Lyme disease. The classical clinical manifestation of LA comprises mono- or oligo-artritis of the large joints, most often the knee, but also elbows, ankles and hips can be affected and polyarthritis is described in up

to10 % of cases of recent-onset arthritis [569]. Selecting patients with oligoarthritis including a knee resulted in 226 patients out of 1180, including 8 out of 9 definite LA cases. The one case missed by this selection was the patient who was additionally diagnosed with RA and presented with polyarthritis. Of n=226 patients, n=12 had positive *B. burgdorferi* serology making antibiotic treatment a worthwhile consideration for he benefit of these patients. Based on these data serologic testing for Lyme disease should always be considered in patients with oligoarthritis including a knee, even in the absence of a tick bite or erythema migrans.

In conclusion, our data show that amongst patients presenting with early-onset arthritis LA is diagnosed in 0.8%. Given the very low prevalence of LA, preselection of patients with a suspect clinical presentation is necessary to increase the positive predictive value to acceptable levels.

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