

Lyme Meningitis in Children with Acute Serous Meningitis with no Clinical Signs of Lyme Borreliosis at Presentation

Mojca Rozic¹, Andra Leskovec², Eva Ruzic-Sabljić³, Maja Arnez^{1,*}

¹University Medical Center Ljubljana, Faculty of Medicine University of Ljubljana, Ljubljana, Slovenia

²Community Health Center Idrija, Idrija, Slovenia

³Institute of Microbiology and Immunology, Faculty of Medicine University of Ljubljana, Ljubljana, Slovenia

Email address:

mojca.rozic@kclj.si (M. Rozic), andra.leskovec@gmail.com (A. Leskovec), eva.ruzic-sablji@mf.uni-lj.si (E. Ruzic-Sabljić),

maja.arnez@kclj.si (M. Arnez)

*Corresponding author

To cite this article:

Mojca Rozic, Andra Leskovec, Eva Ruzic-Sabljić, Maja Arnez. Lyme Meningitis in Children with Acute Serous Meningitis with no Clinical Signs of Lyme Borreliosis at Presentation. *American Journal of Pediatrics*. Vol. 5, No. 4, 2019, pp. 246-253.

doi: 10.11648/j.ajp.20190504.24

Received: October 18, 2019; **Accepted:** November 5, 2019; **Published:** November 11, 2019

Abstract: We evaluate the incidence of Lyme meningitis (LM) in children with acute serous meningitis and compare demographic, clinical and laboratory findings in children with LM and non-LM. During 2004-2005, 122 children fulfilled the inclusion criteria for this prospective clinical study (age < 15 years, meningitis, without typical clinical sign for Lyme borreliosis on admission). Antibodies to *B. burgdorferi* sensu lato were determined in blood and cerebrospinal fluid (CSF) and isolation of *B. burgdorferi* sensu lato was performed. LM was confirmed by isolation of *B. burgdorferi* sensu lato from blood and/or CSF and/or seroconversion to borrelial antigens and/or demonstration of borrelial intrathecal antibody production and/or history of erythema migrans. LM was probable in patients with positive but unchanging borrelial serum antibody titers. LM (83% confirmed, 17% probable) was established in 41 (34%) patients. Demographic, clinical and neurologic findings were comparable between the two groups. Fever and peripheral leukocytosis were more common in non-LM and inappetence and lymphocytic pleocytosis in LM. Borrelial serum IgM and/or IgG was found in 25%, seroconversion in 39%, IgG intrathecal antibody production in 5% and isolation of *B. burgdorferi* sensu lato from CSF and blood in 41% and 22% of patients, respectively. LM was found in 34% of children with acute serous meningitis. It is impossible to distinguish LM from non-LM only from medical history, clinical examination and basic blood and CSF investigations. For this reason, other signs of Lyme borreliosis and microbiological studies on Lyme borreliosis are compulsory.

Keywords: Lyme Meningitis, Children, Lyme Borreliosis, Diagnosis

1. Introduction

Lyme borreliosis (LB) is a tick-borne infectious disease caused by *Borrelia burgdorferi* sensu lato. The principal vectors of LB are *Ixodes scapularis* in the United States of America (USA) and *Ixodes ricinus* in Europe. In USA, the only species known to cause LB is *B. burgdorferi* sensu stricto (*B. burgdorferi*) while in Europe at least five species can cause human disease from which *B. afzelii* and *B. garinii* are predominant. *B. afzelii* is mostly associated with a skin lesion known as erythema migrans (EM), which is pathognomonic clinical sign of LB, while *B. garinii* seems to be most neurotropic [1–3].

LB appears in three stages: early localized, early disseminated and late LB [1–3]. Acute serous meningitis is a part of early, disseminated stage of the disease [4, 5]. Diagnosis of LM can be simple if associated clinical signs of LB (EM, cranial nerve involvement, borrelial lymphocytoma, radiculitis, etc.) are clearly evident. Differentiating Lyme meningitis (LM) from other forms of acute serous meningitis in children is a common diagnostic dilemma in LB endemic regions [6–9]. The vast majority of cases in children are due to enteroviral infection [10]. Parenteral antibiotics are indicated in children for LM but not when causes of acute serous meningitis are viral [11].

This prospective clinical study was performed to evaluate

the incidence of LM in children with acute serous meningitis and to compare demographic, clinical and laboratory findings in children with LM and non-LM.

2. Patients and Methods

2.1. Patients and Study Design

The prospective clinical study was conducted at the Department of Infectious Diseases, University medical Centre Ljubljana, Slovenia in 2004 and 2005. The study was approved by the Medical Ethics Committee of the Ministry of Health of the Republic of Slovenia. Informed consent was obtained from parents of all patients. Our patients were consecutive children younger than 15 years, hospitalized because of acute serous meningitis, without EM or other highly suggestive clinical sign of LB (cranial nerve involvement, borreliar lymphocytoma, radiculitis etc.). The patients were followed-up for at least 12 months.

2.2. Methods

Medical history, physical examination, basic hematologic, biochemical and microbiologic investigations, and lumbar puncture with cerebrospinal fluid (CSF) investigation were performed at the time of admission. Abnormal CSF findings were defined as reported previously in our study of patients with multiple EM and central nervous system (CNS) involvement [12]. Briefly, a white blood cell count (WBC) $\geq 5 \times 10^6/l$ in CSF was considered as CSF pleocytosis, elevated protein level ≥ 0.45 g/l, albumin ≥ 300 mg/l, IgG ≥ 40 mg/l, IgM ≥ 0.7 mg/l, IgA ≥ 5 mg/l and decreased glucose concentrations $< 50\%$ of blood glucose. CSF flow rate and the presence of intrathecal antibody production were defined according to the criteria reported by Reiber *et al* [13]. In each patient venous blood and CSF samples were simultaneously taken for determination of CSF flow rate. In blood and CSF concentrations of albumin and immunoglobulins: G (IgG), A (IgA) and M (IgM) were determined.

2.3. Microbiologic Investigations

Borreliar immunofluorescent assay (IFA) of IgM and IgG antibody titres without preabsorption were determined in serum and CSF as reported previously [14]. Titers ≥ 256 in serum and ≥ 16 in CSF were considered positive. Intrathecal

specific antibody synthesis was determined by calculation of antibody index (AI). AI > 1.4 was considered as increased [13]. Borreliar serum antibodies measurements were repeated 1, 3, 6 and 12 months after the enrolment into the study.

Blood and CSF specimens were cultured in modified Kelly-Pettenkofer (MKP) medium. Specimens were incubated at 33°C and examined by dark-field microscopy. Isolated strains of *B. burgdorferi* sensu lato were identified by polymerase chain reaction (PCR) and/or by pulsed-field gel electrophoresis (PFGE) as reported previously [15–17]. DNA was restricted by *Mlu*I restriction enzyme.

The diagnosis of LB was established according to the case definitions used in our previous studies [18]. Briefly, LB was considered confirmed by isolation of *B. burgdorferi* sensu lato from blood and/or CSF and/or by seroconversion to borreliar antigens and/or by demonstration of borreliar intrathecal antibody production and/or EM four months prior to the onset of meningitis. LB was considered probable in patients with positive but unchanging borreliar serum antibody titers.

2.4. Treatment

Patients with LM were treated with intravenous ceftriaxone for 14 days.

2.5. Statistical Analysis

Differences in categorical data were analyzed by the Yates corrected chi square test or Fisher's exact test, whereas differences in continuous data were assessed by Kruskal-Wallis test and Wilcoxon rank sum test. All P values were two tailed; $P < 0.05$ was considered statistically significant.

3. Results

During the two-year period 122 patients (36 girls, 86 boys) fulfilled the inclusion criteria. The median age of the patients was 7.0 years (range: 2.5 to 14.5). There was no difference in age regarding sex of the patients: median age in girls was 6.75 (3 – 14.5) years and in boys 7.5 (2.5 – 14.5) years ($p = 0.1290$). Median duration of hospitalization was 4 (2 – 18) days.

According to the case definitions, the diagnosis of LM was established in 41 (34%) out of 122 patients with acute serous meningitis. LM was confirmed in 34 (83%) and probable in 7 (17%) patients (Table 1).

Table 1. Case definitions for the diagnosis of Lyme meningitis in 41 patients with acute serous meningitis.

Case definition for Lyme meningitis ¹	Confirmed	Probable
Number of patients (%)	34 (83)	7 (17)
Isolation of <i>B. burgdorferi</i> sensu lato from blood	3	
Isolation of <i>B. burgdorferi</i> sensu lato from blood + IFA-IgM seroconversion to borreliar antigens	1	
Isolation of <i>B. burgdorferi</i> sensu lato from blood + IFA-IgG seroconversion to borreliar antigens	1	
Isolation of <i>B. burgdorferi</i> sensu lato from blood + Isolation of <i>B. burgdorferi</i> sensu lato from CSF	2	
Isolation of <i>B. burgdorferi</i> sensu lato from blood + Isolation of <i>B. burgdorferi</i> sensu lato from CSF + IFA-IgG seroconversion to borreliar antigens + history of recent solitary EM	1	
Isolation of <i>B. burgdorferi</i> sensu lato from CSF	8	
Isolation of <i>B. burgdorferi</i> sensu lato from CSF + IFA-IgG seroconversion to borreliar antigens	2	
Isolation of <i>B. burgdorferi</i> sensu lato from CSF + history of recent solitary EM	1	
Isolation of <i>B. burgdorferi</i> sensu lato from CSF + positive borreliar serum IFA-IgG antibodies	1	
IFA-IgG seroconversion to borreliar antigens	8	

Case definition for Lyme meningitis ¹	Confirmed	Probable
IFA-IgM seroconversion to borrelial antigens	2	
IFA-IgG seroconversion to borrelial antigens + history of recent solitary EM	1	
History of recent solitary EM	1	
Positive borrelial serum IFA-IgG antibodies + borrelial intrathecal IFA-IgG antibody production (AI = 7.6) + history of recent multiple EM	1	
Positive borrelial serum IFA-IgG antibodies + borrelial intrathecal IFA-IgG antibody production (AI = 29.79)	1	
Positive borrelial serum IFA-IgG antibodies		5
Positive borrelial serum IFA-IgM antibodies		2

¹= reference 18

IFA = Immunofluorescent assay

CSF = cerebrospinal fluid

EM = erythema migrans

AI = antibody index (≤ 1.4 normal value)

The month of the hospitalization in patients with LM and non-LM is shown in figure 1.

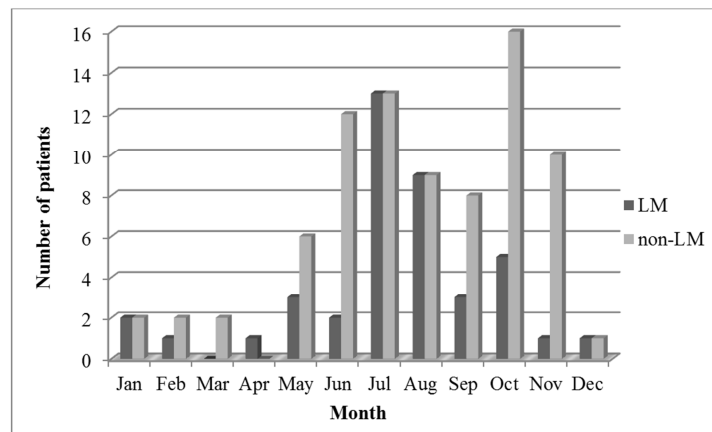


Figure 1. Month of hospitalization in 41 patients with Lyme meningitis (LM) and 81 patients with non-Lyme meningitis (non-LM).

Demographic features and clinical characteristics in the two groups are shown in table 2. The duration of illness before the hospitalization was longer in patients with LM but the difference was not statistically significant.

Table 2. Demographic and clinical features in 41 patients with Lyme meningitis (LM) and 81 with non-Lyme meningitis (non-LM).

Variable	LM	Non-LM	P
Female ¹	16 (39)	20 (25)	0.1529
Male ¹	25 (61)	61 (75)	
Age (years):			0.4787
Mean \pm SD	7.3 \pm 3	7.9 \pm 3.6	
Median (range)	7 (2.5 – 14.5)	7.5 (3 – 14.5)	
Tick bite ¹	22 (54)	35 (43)	0.3678
Incubation (days) ²			0.6610
Mean \pm SD	23.9 \pm 21.8	24.9 \pm 17.5	
Median (range)	16 (6 – 79)	19 (3 – 60)	
Biphasic course of the illness ¹	16 (39)	35 (43)	0.8038
Duration of the illness ³			0.0742
Mean \pm SD	16.3 \pm 34.9	6.4 \pm 6.4	
Median (range)	7 (1 – 180)	4 (0 – 25)	
Duration from the second phase ³			0.6319
Mean \pm SD	10.8 \pm 37.2	1.9 \pm 1.8	
Median (range)	1 (0 – 150)	1 (0 – 8)	
Interval between the onset of the initial and the second phase of the illness (days)			0.0270
Mean \pm SD	7.1 \pm 6.8	9.8 \pm 5.2	
Median (range)	6 (2 – 30)	9 (2 – 22)	
Antibiotics used prior the hospitalization ¹	9 (22)	18 (22)	0.8440
Duration of hospitalization (days)			0.8682
Mean \pm SD	4.8 \pm 1.7	5.2 \pm 2.8	
Median (range)	4 (3 – 9)	4 (2 – 18)	

¹Number of patients (%)

²14 patients with LM, 22 patients with non-LM

³Days prior to the hospitalization

At the time of admission to the hospital fever was more common in non-LM ($p = 0.0030$) and inappetence in LM ($p = 0.0073$). Five (12%) patients with LM had had history of untreated EM from seven days to two months prior to the

inclusion into the study. On physical examination, no difference in clinical and neurologic signs between the patients with LM and non-LM were found. Patients with LM had significantly milder illness than patients with non-LM (Table 3).

Table 3. Clinical and neurologic symptoms and signs in 41 patients with Lyme meningitis (LM) and 81 with non-Lyme meningitis (non-LM).

Symptom ¹	LM	Non-LM	P
Headache	36 (88)	72 (89)	1.0000
Fever	25 (61)	70 (86)	0.0030
Vomiting	25 (61)	58 (72)	0.3253
Fatigue	11 (27)	22 (27)	0.8596
Photophobia	8 (20)	16 (20)	0.8341
Inappetence	10 (24)	5 (6)	0.0093
Nausea	6 (15)	13 (16)	0.9516
Abdominal pain	3 (7)	18 (22)	0.0708
Neck pain	7 (17)	8 (10)	0.3944
Myalgia	5 (12)	5 (6)	0.3012
Arthralgia	5 (12)	7 (9)	0.5348
Skeletal pain	2 (5)	1 (1)	0.2611
Rigor	0	4 (5)	0.2993
Diarrhoea	3 (7)	6 (7)	1.0000
Sore throat	0	5 (6)	0.1668
Common cold	4 (10)	6 (7)	0.7311
Cough	4 (10)	8 (10)	1.0000
Earache	1 (2)	2 (2)	1.0000
Dizziness	1 (2)	2 (2)	1.0000
Irritability	1 (2)	1 (1)	1.0000
Skin rash	1 (2)	2 (2)	1.0000
Consciousness disturbance	1 (2)	2 (2)	1.0000
Convulsions	0	2 (2)	0.5501
Collapse	0	2 (2)	0.5501
Aphasia	0	1 (1)	1.0000
Palpitation	1 (2)	0	0.3361
Sternal pain	0	1 (1)	1.0000
Clinical sign ¹			
Temperature $\geq 38^{\circ}\text{C}$	13 (32)	37 (46)	0.1980
Erythematous throat	11 (27)	20 (25)	0.9712
Dehydration	4 (10)	20 (25)	0.0856
Abdominal tenderness	5 (12)	5 (6)	0.3012
Skin rash	2 (5)	8 (10)	0.4925
Cardiac murmur	5 (12)	4 (5)	0.1617
Conjunctivitis	4 (10)	3 (4)	0.2229
Otitis	2 (5)	2 (2)	0.6017
Rhinitis	1 (2)	2 (2)	1.0000
Bronchitis	1 (2)	2 (2)	1.0000
Enlarged liver	0	6 (7)	0.0964
Neurologic sign ¹			
Positive meningeal signs	35 (85)	70 (86)	0.9061
Tremor	5 (12)	10 (12)	0.7888
Ataxia	1 (2)	1 (1)	1.0000
Conscious disturbance	0	4 (5)	0.2993
Reduced muscle strength	0	1 (1)	1.0000
Elevated muscle tone	0	2 (2)	0.5501
Hyperreflexia	0	2 (2)	0.5501
Babinski's sign	0	2 (2)	0.5501
Severity of initial illness ²			
Mild	34 (83)	51 (63)	0.0396
Moderate	7 (17)	29 (36)	0.0533
Severe	0	1 (1)	1.0000

¹Number of patients (%)

²Mild = meningitis, Moderate = meningoencephalitis, Severe = treatment in intensive care unit

Higher frequency of leukocytosis [57/81 (70%) versus 20/41 (49%); $p = 0.0327$] with neutrophil predominance [73/81 (91%) versus 30/41 (73%); $p = 0.0175$] was detected

in patients with non-LM. No other significant difference regarding basic hematologic and biochemical investigations were found between the two groups.

Blood for culturing for *B. burgdorferi* sensu lato was taken from 110/122 (90%) patients with acute serous meningitis: in 37 (90%) with LM and 73 (90%) with non-LM ($p = 1.0000$). The overall isolation rate of *B. burgdorferi* sensu lato (*B. afzelii* 3, *B. garinii* 1, *Borrelia* did not grow well enough to enable identification of the genospecies 4) from blood in patients with LM was 22% (8/37).

At the time of admission to the hospital, borreliar serum IFA IgM and IgG were determined in all 41 patients with LM and 79 (98%) with non-LM ($p = 1.0000$). Positive IgM and IgG antibodies against *B. burgdorferi* sensu lato were found in 2 (5%) and 8 (20%) patients with LM, respectively. We

couldn't find any patient with simultaneous presence of specific serum IgM and IgG. Seroconversion to borreliar antigens was detected in 16/41 (39%) patients.

At the time of inclusion into the study lumbar puncture and CSF investigation were performed in all patients. Time interval between the beginning of illness and lumbar puncture was comparable between the two groups [7 (1 – 180) days in LM versus 4 (0 – 26) in non-LM, $p = 0.0674$].

CSF findings in 122 patients with acute serous meningitis are shown in table 4. Lymphocytic predominance was more frequently found in patients with LM.

Table 4. Cerebrospinal fluid findings in 41 patients with Lyme meningitis (LM) and 81 with non-Lyme meningitis (non-LM).

Variable	LM	Non-LM	P
White blood cell count $\times 10^6/l$			
Mean \pm SD	296.7 \pm 445.8	235 \pm 329.4	0.8901
Median (range)	123 (8 – 1877)	133 (6 – 2048)	
Lymphocytic predominance ¹	29 (71)	35 (43)	0.0073
Number of abnormal findings/number of examined patients (%)			
Decreased glucose concentration ²	10/40 (25)	9/81 (11)	0.0873
Elevated total protein concentration (≥ 0.45 mg/L)	15/40 (38)	39/81 (48)	0.3607
Elevated albumin concentration (≥ 300 mg/L)	12/37 (32)	22/68 (32)	0.8337
Elevated total IgG concentration (≥ 40 mg/L)	11/37 (30)	18/68 (26)	0.8979
Elevated total IgA concentration (≥ 5 mg/L)	7/37 (19)	10/68 (15)	0.7775
Elevated total IgM concentration (≥ 0.7 mg/L)	28/37 (76)	51/66 (77)	0.9530

¹Number of patients (%)

²< 50% of blood glucose

Ig = Immunoglobulins

In 8/122 (7%) patients (all were boys) more than $1000 \times 10^6/l$ WBC were found in CSF: in 4 (10%) with LM and 4 (5%) with non-LM ($p = 0.4310$).

In 109/122 (89%) patients CSF was taken for culturing for *B. burgdorferi* sensu lato: in 37 (90%) patients with LM and 72 (88%) in non-LM. The overall isolation rate of *B. burgdorferi* sensu lato (*B. afzelii* 6, *B. garinii* 6, *Borrelia* did not grow well enough to enable identification of the genospecies 3) from CSF in patients with LM was 41% (15/37). In 8% (3/37) *B. burgdorferi* sensu lato was isolated simultaneously from blood and CSF.

At the time of inclusion into the study borreliar CSF IFA IgM and IgG were determined in 109/122 (89%) patients: 39 (95%) in LM and 70 (86%) in non-LM ($p = 1.0000$). Positive CSF IgG antibodies against *B. burgdorferi* sensu lato were found in 2 (5%) patients with LM. In these patients intrathecal borreliar IgG synthesis was also established. We couldn't find any patients with LM and positive borreliar IgM in CSF.

4. Discussion

In the present study we were interested in the incidence of LM in Slovenian children with acute serous meningitis without EM or other highly suggestive clinical sign of LB. To our knowledge this is the first prospective study about this problem reported in the literature. In two years, we treated 122 children for acute serous meningitis. Using case definitions for the diagnosis of LB, LM was established in 41

(34%) of patients (Table 1). Acute serous meningitis in children is mainly of enteroviral origin [10]. Because LM and enteroviral meningitis are common in LB-endemic regions and both occur mostly in the summer and fall, it is essential to differentiate clinical and laboratory features when EM is absent [19, 20]. The treatment of viral meningitis is supportive only, whereas the treatment of LM with parenteral antibiotics significantly decreases both the acute and the long-term symptoms [11, 21].

In Norway, most children with neuroborreliosis are diagnosed between June and November, whereas no children are diagnosed between January and March [22]. The results of our study partially support this finding. The majority of our patients with LM were registered during the warm months. The numbers of hospitalized children with LM and non-LM were comparable, however patients with LM and non-LM were seen throughout the whole year (Figure 1).

The nervous system is involved in 10% to 15% of patients with untreated *B. burgdorferi* sensu lato infection [20]. Neurologic involvement in LB in children is most commonly seen as lymphocytic meningitis, with or without cranial nerve palsy [20, 23, 24].

Retrospective studies from Europe and the USA report less pronounced signs and symptoms (fever, headache, neck stiffness) of meningitis, longer duration of symptoms before evaluation, lymphocytic pleocytosis and elevated CSF protein levels more often in LM than in non-LM patients [6, 11, 25, 26]. However, reported patients also exhibit papilledema, EM rash or cranial neuropathy that are typical

or highly suggestive clinical signs for LB in up to 88% [6, 11, 19, 25–31].

In our previous prospective study on etiology and principal clinical features of acute serous meningitis in children conducted in 1997, the diagnosis of LM was based only on findings of *B. burgdorferi* sensu lato IgM and/or IgG antibodies in patient's blood (16th annual meeting of the European Society for Paediatric Infectious Diseases, abstract P 92). LM was diagnosed in 4.9% out of 102 patients. The majority of patients with established etiology had enteroviral meningitis (46.1%).

In present study we didn't search for enteroviral infection. LM was established in 34% out of 122 patients with acute serous meningitis. We believe that the higher percentage of patients with LM is partially the result of different diagnostic approach, using not only measurement of specific borrelial antibodies in blood but also in CSF, as well as with culturing of blood and CSF for *B. burgdorferi* sensu lato. Our patients had no EM or other typical or highly suggestive clinical sign of LB. For this reason, a direct comparison of demographic, clinical and laboratory characteristics of patients between studies reported in the literature is ambiguous. In our patients the duration of illness prior to the hospitalization was longer in patients with LM compared to those with non-LM, but the difference was not statistically significant (Table 2). At the time of admission to the hospital, fever was more common in non-LM and inappetence in LM, however headache, neck pain and fatigue were comparable between the two groups (Tables 3). On physical examination no significant difference in clinical and neurologic signs was found between the two groups (Table 3).

We agree with Porwancher that the most difficult problem with identifying patients with LM without EM is the need to rely on laboratory criteria for diagnosis of LB [27].

The diagnosis of LB is based on clinical manifestations and history of exposure to ticks in an endemic area [1–3]. Detection of borrelial antibodies represents a fundamental aid to diagnosis. Because neurologic symptoms, by definition, require disseminated infection, serologic tests are markedly positive, often with a prominent IgM component. Very rarely, neurologic symptoms proceed to development of a measurable antibody response so that convalescent serologic tests may be necessary [20].

At the time of admission to the hospital positive borrelial IgM and IgG antibodies were found in 5% and 20% of our patients with LM, respectively. Seropositivity was lower than reported in the literature [1–3, 32]. The reason may be using a less sensitive diagnostic serological method [33]. Seroconversion to borrelial antigens was detected in 39%.

Although not standardized, the most widely accepted laboratory method for diagnosis of acute neuroborreliosis is intrathecal antibody production to *B. burgdorferi* sensu lato [23, 25, 27, 34]. Studies have shown variable sensitivity (33 – 92%), but excellent specificity (93 – 100%) for acute neuroborreliosis [27]. The detection of intrathecally produced IgM antibodies shows a high degree of sensitivity in neuroborreliosis with short duration of symptoms, especially

in children [23, 32]. According to Christen in 10% of neuroborreliosis cases, the CSF is positive for borrelial IgM antibodies but serum is negative when tested first [35].

In our study, we couldn't find any child with *B. burgdorferi* sensu lato-IgM intrathecal antibody production. Borrelial IgG intrathecal antibody production was found in only 2 patients with LM (Table 1). This was expected since the median duration of illness before hospitalization in patients with LM was seven days (Table 2). Intrathecal antibody response to *B. burgdorferi* sensu lato usually begins in the second week after onset of neurologic symptoms [21].

Additional laboratory diagnostic method for LM is CSF culture for *B. burgdorferi* sensu lato. Although time consuming and relatively insensitive, it is still the gold standard for diagnosis of neuroborreliosis [27, 28, 36, 37]. Sensitivity is 10 – 30%: up to 50% in patients with disease duration of fewer than two weeks and 13% in patients with disease duration of greater than two weeks [32].

Our results support these findings since the median duration of illness in our patients with LM was seven days, and the rate of isolation of *B. burgdorferi* sensu lato from CSF was 41%.

To our knowledge this is the first report on isolation of *B. burgdorferi* sensu lato from CSF in prospectively evaluated children with acute serous meningitis without EM or highly suggestive clinical sign of LB. There was no significant difference between isolation rate of *B. garinii* and *B. afzelii*. This is in contrast to the data from the literature regarding neuroborreliosis [1–3, 20]. We couldn't find any patient with isolation of *B. burgdorferi* from CSF. This was expected since *B. burgdorferi* is infrequent pathogen of neuroborreliosis [20].

B. burgdorferi sensu lato typically elicits CSF pleocytosis, altered blood-brain barrier permeability, increased protein and often a local antibody response resulting in a relative increase in CSF Ig concentration [38]. We found the same changes in our patients. However, patients with LM differ significantly from those with non-LM only in lymphocytic predominance, which was more common in patients with LM. There were no other significant differences regarding CSF findings between the two groups (Table 4).

Hypoglycorrhachia in areas endemic for LB is suggestive for acute LM [23]. Decreased CSF glucose concentration in our patients with LM and those with non-LM was found in 25% and 11%, respectively (Table 4). The difference was not statistically significant ($p = 0.0873$), however the median concentration of CSF glucose concentration was significantly lower in patients with LM [2.95 (range 2.1 – 3.9) versus 3.1 (range 2.2 – 4.3), $p = 0.0184$].

The proposed treatment of LM, among others, is ceftriaxone intravenously for two to four weeks [20]. In our present study we treated all patients with LM with intravenous ceftriaxone for 14 days.

5. Conclusion

Antibiotics are indicated in children for LM but not when

causes of acute serous meningitis are viral. In this prospective clinical study LM was found in 34% of children with acute serous meningitis with no signs of LB at presentation. In 27% we diagnosed LM only by isolation of *B. burgdorferi* sensu lato from blood or CSF. Isolation rates of *B. garinii* and *B. afzelii* from CSF were comparable. It is impossible to distinguish LM from non-LM only from medical history, clinical examination and basic blood and CSF investigations. For this reason, other signs of LB and microbiological studies on LB are compulsory. We believe that the direct detection of *B. burgdorferi* sensu lato from CSF should become an integral part of routine diagnostic procedures in children with acute serous meningitis in endemic regions for LB. Adding direct detection to routine diagnostic procedures would rationalize the use of antibiotics in children with acute serous meningitis.

References

- [1] Stanek G, Wormser GP, Gray J, Strle F (2012). Lyme borreliosis. *Lancet* 397, 461–473.
- [2] Stanek G, Strle F (2018). Lyme borreliosis—from tick bite to diagnosis and treatment. *FEMS Microbiol Rev* 42, 233–258.
- [3] Steere AC, Strle F, Wormser GP, Hu LT, Branda JA, Hovius JWR, et al (2017). Lyme borreliosis. *Nat Rev Dis Primers*; 2: 16090. doi: 10.1038/nrdp.2016.90.
- [4] Sood SK (2015). Lyme disease in children. *Infect Dis Clin N Am* 29, 281–294.
- [5] Shapiro E (2018). Lyme borreliosis in 2018. What is new (and what is not). *JAMA* 320, 635–636.
- [6] Avery RA, Frank G, Glutting JJ, Eppes SC (2006). Prediction of Lyme meningitis in children from Lyme disease-endemic region: a logistic-regression model using history, physical, and laboratory findings. *Pediatrics* 117, e1–e7.
- [7] Esposito S, Bosis S, Sabatini C, Tagliaferri L, Principi N (2013). *Borrelia burgdorferi* infection and Lyme disease in children. *Intern J Infect Dis* 17, e153–e158.
- [8] Sanchez E, Vannier E, Wormser GP, Hu LT (2016). Diagnosis, treatment, and prevention of Lyme disease, Human granulocytic anaplasmosis, and Babesiosis. *JAMA* 315, 1767–1777.
- [9] Barstad B, Quarsten H, Tveitnes D, Noraas S, Ask IS, Saeed M, et al (2018). Direct molecular detection and genotyping of *Borrelia burgdorferi* sensu lato in cerebrospinal fluid of children with Lyme neuroborreliosis. *J Clin Microbiol* 56: e01868-17. <https://doi.org/10.1128/JCM.01868-17>.
- [10] Rothbart HA (1995). Enteroviral infections of the central nervous system. *Clin Infect Dis* 20, 971–981.
- [11] Garro AC, Rutman M, Simonsen K, Jaeger JL, Chapin K, Lockhart G (2009). Prospective validation of a clinical prediction model of Lyme meningitis in children. *Pediatrics* 123, e829–e834.
- [12] Arnez M, Pleterski-Rigler D, Ahčan J, Ružič-Sabljic E, Strle F (2001). Demographic features, clinical characteristics and laboratory findings in children with multiple erythema migrans in Slovenia. *Wien Klin Wochenschr* 113, 98–101.
- [13] Reiber H, Peter JB (2001). Cerebrospinal fluid analysis: disease-related data patterns and evaluation programs. *J Neurol Sci* 184, 101–122.
- [14] Wilske B, Schierz G, Preac-Mursic V, Weber K, Pfister HW, Einhaupl K (1984). Serological diagnosis of erythema migrans disease and related diseases. *Infection* 5, 331–335.
- [15] Preac-Mursic V, Wilske B, Schierz G (1986). European *Borrelia burgdorferi* isolation from humans and ticks culture conditions and antibiotic susceptibility. *Zentralbl Bakteriol Mikrobiol Hyg A* 263, 112–118.
- [16] Ružič-Sabljic E, Arnez M, Logar M, Maraspin V, Lotrič-Furlan S, Cimperman J, et al (2005). Comparison of *Borrelia burgdorferi* sensu lato strains isolation from specimens obtained simultaneously from two different sites of infection in individual patients. *J clin Microbiol* 43, 2194–2200.
- [17] Postic D, Assous MV, Grimont PA, Baranton G (1994). Diversity of *Borrelia burgdorferi* sensu lato evidenced by restriction fragment length polymorphism of rrf (5S)-rrl (23S) intergenic spacer amplicon. *Int J Syst Bacteriol* 44, 743–752.
- [18] Arnez M, Lužnik-Bufon T, Avšič-Županc T, Ružič-Sabljic E, Petrovec M, Lotrič-Furlan S, et al (2003). Causes of febrile illnesses after a tick-bite in Slovenian children. *Pediatr Infect Dis J* 22, 1078–1083.
- [19] Shah SS, Zaoutis TE, Turnquist J, Hodinka RL, Coffin SE (2005). Early differentiation of Lyme from enteroviral meningitis. *Pediatr Infect Dis J* 24, 542–545.
- [20] Halperin JJ (2015). Nervous system Lyme disease. *Infect Dis Clin N Am* 29, 241–253.
- [21] Garro AC, Rutman MS, Simonsen K, Jaeger JL, Chapin K, Lockhart G (2011). Prevalence of Lyme meningitis in children with aseptic meningitis in a Lyme disease-endemic region. *Pediatr Infect Dis J* 30, 990–992.
- [22] Øymar R, Tveitnes D (2009). Clinical characteristics of childhood Lyme neuroborreliosis in an endemic area of northern Europe. *Scand J Infect Dis* 41, 88–94.
- [23] Dayan NE, Rubin LG, Di John DD, Sood SK (2004). Hypoglycorrhachia in Lyme meningitis. *Pediatr Infect Dis J* 23, 370–371.
- [24] Broekhuijsen-van Henten D, Braun KP, Wolfs TFW (2010). Clinical presentation of childhood neuroborreliosis; neurological examination may be normal. *Arch Dis Child* 95, 910–914.
- [25] Eppes SC, Nelson DK, Lewis L, Klein JD (1999). Characterization of Lyme meningitis and comparison with viral meningitis in children. *Pediatrics* 103, 957–960.
- [26] Tuerlinckx D, Bodart E, Garrino MG, de Bilderling G (2003). Clinical data and cerebrospinal fluid findings in Lyme meningitis versus aseptic meningitis. *Eur J Pediatr* 162, 150–153.
- [27] Porwancher R (2006). Predictive model of Lyme meningitis. *Pediatrics* 118, 438–439.
- [28] Tveitnes D, Natås OB, Skadberg Ø, Øymar K (2012). Lyme meningitis, the major cause of childhood meningitis in an endemic area: a population based study. *Arch Dis Child* 97, 215–220.

- [29] Cohn KA, Thompson AD, Shah SS, Hines EM, Lyons TW, Welsh EJ, et al (2012). Validation of clinical prediction rule to distinguish Lyme meningitis from aseptic meningitis. *Pediatrics* 129, e46–e53.
- [30] Tuerlinckx D, Bodart E, Jamart J, Glupczynski Y (2009). Prediction of Lyme meningitis based on a logistic regression model using clinical and cerebrospinal fluid analysis. *Pediatr Infect Dis J* 28, 394–397.
- [31] Waespe N, Steffen I, Heininger U (2010). Etiology of aseptic meningitis, peripheral facial nerve palsy, and a combination of both in children. *Pediatr Infect Dis J* 29, 453–456.
- [32] Wilske B, Fingerle V, Schulte-Spechtel V (2007). Microbiological and serological diagnosis of Lyme borreliosis. *FEMS Immunol Med Microbiol* 49, 13–21.
- [33] Cerar T, Ruzic-Sabljic E, Cimperman J, Stle F (2006). Comparison of immunofluorescence assay (IFA) and LIAISON® in patients with different clinical manifestations of Lyme borreliosis. *Wien Klin Wochenschr* 118, 686–690.
- [34] Theel ES, Agüero-Rosenfeld ME, Pritt B, Adem PV, Wormser GP (2019). Limitations and confusing aspects of diagnostic testing for neurologic Lyme disease in the United States. *J Clin Microbiol* 57: e01406-18. <https://doi.org/10.1128/JCM.01406-18>.
- [35] Christen HJ (1996). Lyme neuroborreliosis in children. *Ann Med* 28, 235–40.
- [36] Agüero-Rosenfeld ME, Wang G, Schwartz I, Wormser GP (2005). Diagnosis of Lyme borreliosis. *Clin Microbiol Rev* 18, 484–509.
- [37] Schutzer SE, Body BA, Boyle J, Branson BM, Dattwyler RJ, Fikrig E, et al (2019). Direct diagnostic tests for Lyme disease. *Clin Infect Dis* 68, 1052–1057.
- [38] Halperin JJ (2002). Nervous system Lyme disease. *Vector Borne Zoonotic Dis* 2, 241–247.