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Blood-CSF-barrier dysfunction is a marker for encephalitic involvement in patients with aseptic meningitis/meningoencephalitis

C. Hoyer[a,1], P. Eisele[a,1], A.D. Ebert[a], S. Schneider[b], A. Gass[a], M. Fatar[a], K. Szabo[a], A. Alonso[a,*]

[a] Department of Neurology, Medizinische Fakultät Mannheim, University of Heidelberg, Theodor-Kutzer-Ufer 1-3, 68135 Mannheim, Germany
[b] Institute for Clinical Chemistry, Medizinische Fakultät Mannheim, University of Heidelberg, Theodor-Kutzer-Ufer 1-3, 68135 Mannheim, Germany

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ABSTRACT

Background: The term "aseptic meningitis" encompasses cases of meningitis with negative bacterial CSF culture, which predominantly are of viral etiology. While the clinical course is usually benign, complications such as encephalitic involvement resulting in a more severe clinical course may occur. Dysfunction of the blood-brain-barrier (BBB), which is a prerequisite for viral entry into the brain parenchyma, can be approximated using the CSF/serum albumin ratio, readily obtainable in routine CSF analysis.
Objecitves: Analysis of CSF patterns in patients with aseptic meningitis/meningoencephalitis with a focus on BBB dysfunction as a marker for encephalitic involvement.
Study design: Retrospective chart review of patients admitted to our hospital between 2004 and 2016 with a diagnosis of aseptic meningitis/meningoencephalitis.
Results: Patients with aseptic meningitis displaying clinical, MR-tomographic or electroencephalographic signs of encephalitic involvement were significantly older than patients without these features (47.4 vs. 35.5 yrs., $p = 0.002$). In patients with meningoencephalitis, CSF analysis revealed a more severe disruption of BBB, approximated by the CSF/serum albumin ratio ($p = 0.002$). Compromised BBB function correlated positively with length of hospitalization ($p = 0.007$), indicative of a more severe clinical course. The number of CSF lymphocytes was found to predict the severity of the BBB disruption, which additionally was more frequently observed when herpesviridae were identified as infectious agents.
Conclusions: We suggest that the CSF/serum albumin ratio as an estimate for BBB function should be attended to in the evaluation of patients with aseptic meningitis. Severe BBB dysfunction, older age and infection with herpesviridae appear to raise the risk for encephalitic involvement.

1. Background and objectives

The term "aseptic meningitis" subsumes all types of meningitis with negative bacterial culture from cerebrospinal fluid (CSF) [1,2]. Characteristically, CSF analyses of patients with aseptic meningitis reveal a lymphocytic pleocytosis accompanied by mild elevation of CSF protein concentration and normal glucose levels [3]. Viral infections are the most common etiology of aseptic meningitis, accounting for about 55–65% of all cases [4,5]. Among these, enteroviruses are the major causative agents in children as well as in adults, followed by herpesviridae [4–6]. Clinical outcome is mostly benign, with complete symptom resolution within the first weeks in most of the patients or milder sequel such as persisting headache and fatigue in some patients over several weeks [5]. However, complications in terms of encephalitic involvement, intracranial hypertension or cerebral vasculitis with subsequent ischemia may occur [7,8]. Among others, the probability of encephalitic involvement is related to the infectious agent: the herpesviridae varicella zoster virus (VZV) and herpes simplex virus (HSV) 1 have been identified as most commonly involved pathogens in aseptic encephalitis whereas encephalitis due to enterovirus is a rarity [7,9].

As a precondition for encephalitic involvement, viral agents need to enter the central nervous system (CNS). This critically implies compromised blood-brain barrier (BBB) function, either directly through viral infection of brain microvascular endothelial cells or the transgression of tight junctions because of high levels of viremia or inflammation, or indirectly when viral particles are transported into the CNS by infected leukocytes [10]. Even when

* Corresponding author.
E-mail address: alonso@neuro.ma.uni-heidelberg.de (A. Alonso).
[1] These authors contributed equally to the study.

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viruses circumvent the BBB by retrograde axonal transport strategies, secondary impairment of BBB function due to the action of proinflammatory cytokines may ensue.

The CSF/serum albumin ratio, indicating the amount of leakage of blood-borne proteins into the CSF, is a well-known marker for estimating the integrity of the BBB [11]. In a previous work, we have shown that leptomeningeal contrast enhancement and increased CSF/serum albumin ratios as corollaries of BBB dysfunction in patients with aseptic meningitis are associated with a prolonged hospitalization [12], suggesting a more severe clinical course. The purpose of this study was to investigate CSF patterns of patients with aseptic meningitis and meningoencephalitis, with a focus on blood-CSF-barrier dysfunction as a potential risk factor for encephalitic involvement and a more severe clinical course.

2. Study design

The study was approved by the local Institutional Review Board (Medizinische Ethikkommission II der Medizinischen Fakultät Mannheim, University of Heidelberg).

We retrospectively analyzed the charts of 204 consecutive immunocompetent patients with CSF-confirmed diagnosis of aseptic meningitis or meningoencephalitis from 10/2004 to 01/2016. The diagnosis of aseptic meningitis was established by the combination of typical clinical features (headache, fever, neck stiffness), a white blood cell (WBC) count of >5/mm^3 in CSF analysis and negative bacterial CSF culture. Meningoencephalitis was diagnosed if patients presented with a combination of clinical features typical for meningitis and either 1) at least one of the following clinical features: newly developed focal neurologic deficit; acute disturbance of consciousness, orientation or psycho-motorics; new-onset epileptic seizures, or 2) parenchymal pathologies compatible with encephalitis on cerebral MR imaging, or 3) EEG findings compatible with encephalitis, together with a white blood cell (WBC) count of >5/mm^3 in CSF analysis and negative bacterial culture of CSF.

In all patients, we collected clinical data including age, gender, presenting symptoms, duration of hospitalization and medication. CSF was analyzed for WBC count, CSF protein concentration, CSF/serum albumin ratio (Q_{alb}), CSF/serum IgG ratio (Q_{IgG}). The upper limit of the reference range for an intact brain barrier function is age-dependent and was calculated for each patient according to the formula $Q_{alb} = (4+ \text{ age}/15) \times 10^3$ [13]. Blood-brain barrier dysfunction was graduated as mild (Q_{alb} <10 × 103), moderate (Q_{alb} 10–20 × 103) or severe (Q_{alb} >20 × 103) [14]. Routine testing for infectious agents included CSF culture, CSF serology (Herpes simplex IgG/IgM, varicella/zoster IgG/IgM, measles IgG/IgM, mumps IgG/IgM, Borrelia burgdorferi IgG/IgM) and PCR analysis for neurotropic viruses (Herpes simplex 1/2, varicella/zoster, CMV, Epstein-Barr virus (EBV), enterovirus, HHV-6). Further CSF serology (syphilis, HIV) was performed when appropriate in the context of the clinical presentation. As CSF albumin concentration is known to decrease with increasing volume of spinal fluid extracted [13], the total CSF sample volume taken from each patient was standardized to 6 ml. Demographics, clinical and CSF characteristics of our cohort are given in Table 1.

3. Statistics

Statistical analysis was performed using the Statistical Package for the Social Sciences (SPSS), version 22.0.0.0 (IBM, USA), implementing χ^2-test or Fisher's exact test as appropriate for group comparisons of categorical, Mann-Whitney-U-test for ordinal and *t*-test for metric variables. To detect possible predictors of the extent of BBB-dysfunction we performed multiple regression analysis (stepwise regression with backward elimination). For correlation analyses we calculated Pearson correlations for metric variables and Spearman correlations for ordinal variables. A p value < 0.05 was considered to indicate statistical significance.

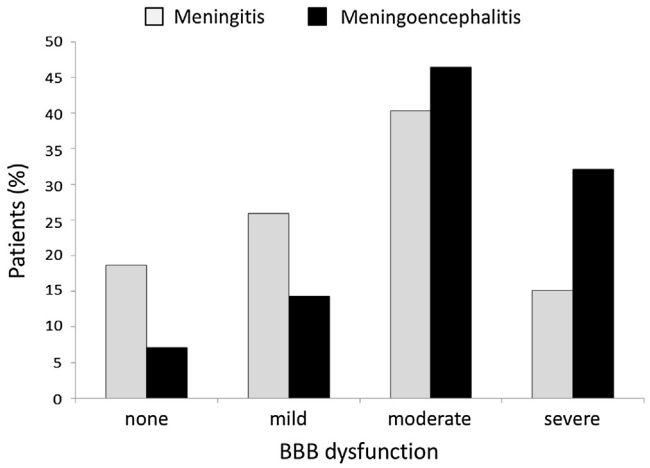

Fig. 1. Graduation of BBB dysfunction (mild, moderate, severe) in patients with meningitis and meningoencephalitis.

4. Results

Of 204 patients with clinically suspected aseptic meningitis or meningoencephalitis and confirming CSF analyses, 173 patients (77 male, 96 female, mean age 35.5 +/− 12.8 years) presented with isolated meningitis. Major clinical feature at presentation was headache (161/173, 93.1%), followed by fever (81/173, 46.8%) and neck stiffness (76/173, 43.9%). Thirty-one patients (19 male, 12 female, mean age 47.4 +/− 18.9 years) had clinical (26/31, 83.9%), electroencephalographic (23/31, 74.2%) or MR-tomographic (11/31, 35.5%) signs of encephalitic involvement. Electroencephalographic findings encompassed generalized slowing (n = 5), focal slowing (n = 15, one of them with focal epileptiform discharges) and intermitted generalized rhythmic activity (n = 3). The most frequent clinical features were aphasia (11/31, 35.5%), acute confusional state (15/31, 48.4%) and epileptic seizures (7/31, 22.6%). Patients with meningoencephalitis were significantly older than patients with isolated meningitis (p = 0.002) and had a significantly longer hospital stay (p < 0.001). There was a trend towards longer timespans between symptom onset and spinal tap between patients presenting with meningitis or meningoencephalitis (p = 0.066) (Table 1). Patients with meningoencephalitis received a follow-up spinal tap more frequently in comparison with patients diagnosed with meningitis (p = 0.001).

CSF findings in patients with aseptic meningitis differed in several aspects from those in patients with encephalitic involvement. The absolute cell count/μl as well as the differential lymphocyte count in patients with meningitis did not differ from that in patients with meningoencephalitis (p = 0.479 and p = 0.111) while the differential neutrophil count was significantly higher in patients with meningitis (p = 0.004). The CSF/serum albumin ratio as a marker for the integrity of the blood-CSF barrier was significantly higher in patients with encephalitic involvement (p = 0.002), indicating a more severe barrier disturbance BBB in patients with meningoencephalitis. In addition, CSF protein content as a raw estimate of BBB dysfunction was significantly higher in patients with encephalitic involvement (p < 0.001, Table 1).

Following graduation of BBB dysfunction as mild, moderate or severe, patients with meningoencephalitis showed significantly higher degrees of BBB disturbance (p = 0.009; Fig. 1). Further, CSF