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Clinical significance of the number of oligoclonal bands in patients with clinically isolated syndromes

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ABSTRACT

CSF oligoclonal bands (OCBs) in patients with clinically isolated syndromes (CIS) are a risk factor for clinically definite multiple sclerosis (CDMS). We aimed to address the relevance of the number of OCBs in the prognosis of CIS patients. 219 CIS patients were included in the study, and 42% of them developed the disease during follow-up (median: 5.04 years). Patients with a high number of CSF OCBs (third quartile, 8–12 OCBs) had 2.5-fold increase in CDMS risk, while no further increase in the HR of disease was observed for patients with more than 12 OCBs. The results did not change after adjustment for additional correlates of CDMS development. This association may be due to the epitope-spreading phenomenon and may reflect the stage of the disease at the time of the examination.

1. Introduction

Since its introduction, MRI has led to a significant improvement in the speed and accuracy of the diagnosis of multiple sclerosis (MS) (Young et al., 1981). Cerebrospinal fluid (CSF) analysis remains important, however, in order to support early diagnosis and, above all, to rule out different diagnoses (Stangel et al., 2013). CSF findings that may indicate a diagnosis of MS include a slightly increased cell count, activated B cells in routine cytological analysis, a high IgG index and the presence of IgG oligoclonal bands (OCBs), all of which constitute the pathophysiological evidences of ongoing inflammation within the CNS (Stangel et al., 2013; Mayringer et al., 2005). In particular, it has been shown that the presence OCBs in the CSF of patients with clinically isolated syndromes (CIS) is an independent prognostic factor for the subsequent development of the disease (Kuhle et al., 2015; Tintore et al., 2008). Furthermore, a more favorable long-term prognosis and slower progression of the disease in OCB-negative patients have been demonstrated (Joseph et al., 2009; Zeman et al., 1996).

OCBs are IgG immunoglobulins generated by plasma blasts and plasma cells in the CSF or CNS compartment (Awad et al., 2010). Therefore, the number of CSF OCBs may have a prognostic role in MS as an indicator of the number of intrathecal active B-cell clones and of the possible spreading of autoreactivity to different epitopes. Although some evidence to support this concept is available (Bourahoui et al., 2004; Avasarala et al., 2001), the prognostic value of these CSF parameters has not been evaluated in patients with CIS.

The current study aims to address the relevance of the number of OCBs in the prognosis of CIS patients.

2. Material and methods

2.1. Patient characteristics

In this retrospective study patients admitted to our department between January 1, 2005 and December 31, 2012 for a first time neurological event suggestive of MS have been included. Further inclusion criteria were: 1) age between 15 and 55 years at the time of the CIS; 2) availability of CSF isoelectrofocusing performed at the time of the hospitalization.

We collected demographic, baseline clinical data, as well as all diagnostic results. The data acquired included age at onset, gender, type of onset (monofocal or multifocal), baseline neurological disability according to the Expanded Disability Status Scale (EDSS), and any steroid therapies received in the four weeks prior to the hospitalization. Brain MRIs and analyses of CSF OCBs were performed according to the routine clinical practice at our MS clinic. CSF cell and protein counts, as well as the blood–brain barrier damage index (defined as: [(CSF Albumin) / (Seric Albumin)] × 100), were retrieved from hospital charts. Agarose isoelectric focusing was used to examine OCBs, combined with immunoblotting and avidin–biotin-amplified double antibody peroxidase staining. The number of CSF-restricted OCBs was assessed by two trained readers (GDC and GP) blinded to patient identity and clinical

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Table 1
Baseline characteristics of the study cohort.

Characteristic	Patients who did not develop CDMS	Patients who developed CDMS
No. of subjects	124	95
Age at onset (years)	32.67 ± 9.40	21.73 ± 8.64
Females — %	75 (60.5)	66 (69.5)
Multifocal type of onset — %	20 (16.1)	15 (15.8)
Type of CIS — %		
Optic neuritis syndrome	37 (29.8)	25 (26.3)
Brainstem syndrome	28 (22.6)	30 (31.6)
Spinal cord syndrome	39 (31.5)	25 (26.3)
Other	20 (16.1)	15 (15.8)
CSF findings		
CSF cells	4.18 ± 6.60	5.43 ± 6.71
CSF proteins	35.02 ± 13.93	33.35 ± 11.63
Number of CSF oligoclonal bands	6.73 ± 7.28	8.84 ± 7.01
Blood–brain barrier damage index	0.50 ± 0.25	0.54 ± 0.21
MRI findings		
T2 lesions	5.66 ± 6.02	8.97 ± 5.58
Gd-enhancing lesions	0.51 ± 0.95	0.74 ± 1.17

Plus–minus values are means ± SD.
Abbreviations: CDMS: clinically definite multiple sclerosis, CIS: clinically isolated syndrome, CSF: cerebrospinal fluid.

follow-up. The MRI scans were retrieved from our hospital archive and the number of areas of increased signal intensity present on the T2-weighted images was marked by a trained reader (MJM) who was also blinded to patient identity and clinical follow-up. Four different categories for the number of lesions were considered: 0 lesions, 1 to 3 lesions, 4 to 9 lesions, and 10 or more lesions (brainstem lesions were not considered in the analysis of patients who presented with a brainstem syndrome) (Fisniku et al., 2008; Tintoré et al., 2006). The presence of gadolinium-enhancing lesions was also assessed.

Follow-up information was retrieved from the local MS patient database (iMed) or it was received via the local investigator at the MS center where the patient subsequently moved. The follow-up duration was considered as the interval between the onset of the first neurological event and the patient's most recent neurological visit.

A diagnosis of clinically definite multiple sclerosis (CDMS) was made when new symptoms or signs occurred according to Poser's MS criteria and only when other diagnoses had been accurately excluded (Poser et al., 1983). All patients who developed CDMS received immunomodulatory treatments after the diagnosis, while seventy-nine patients had started therapy after new subclinical brain lesions were detected at follow-up brain MRI.

The protocol was approved from the ethical committee of our hospital.

2.2. Statistical analyses

Normality of the data was tested using the Shapiro–Wilk test. Normally distributed variables were shown as mean (SD), and differences between groups were analyzed using unpaired t tests. Nonnormally distributed variables were shown as medians with 25 and 75% percentiles, and Mann–Whitney U tests were used to test for differences. Categorical variables were shown as proportions, and the differences were analyzed using χ tests. P values less than 0.05 were considered statistically.

We used univariate and multivariate Cox proportional hazards models to estimate the hazard ratios (HRs) and 95% CIs. To allow for nonlinear association of the number of CSF OCBs with CDMS development, restricted cubic-spline Cox proportional hazard models were used with knots chosen at the 25th, 50th and 75th percentiles, and 0 OCBs was used as reference (Harrell, 2001). Person-years of follow-up for CDMS incidence were calculated from the baseline until CDMS diagnosis, death, or the end of the follow-up period. The proportional hazard assumptions for the Cox models were assessed using Schoenfeld's residuals. Variables used for building the multivariate regression models were chosen based on clinically known prognostic factors. Normograms were used to assess the prognostic contribution of the CSF OCBs to MRI prognostic markers. Bootstrap validation with 200 resamples was used to assess the performance of the models and a concordance index was calculated (Iasonos et al., 2008; Shen et al., 2014).

All statistical analyses were performed using the computing environment R (www.r-project.org).

3. Results

We identified 244 patients who received a complete clinical, CSF, and MRI assessment at our hospital after an initial episode of neurological symptoms suggestive of MS. Twenty-five patients were lost during the follow-up. The remaining 219 patients had a median follow-up of 5.04 years (interquartile range: 2.88–6.89). During the follow-up period, 95 patients (43%) developed CDMS: 48 patients (22%) within the first year and 64 (29%) within the second year. Of the 111 patients who had at least a five year follow-up period, 55 patients (50%) had developed CDMS.

The main demographic and clinical characteristics of the patients at baseline are summarized in Table 1. Patients who subsequently

Table 2
Baseline characteristics of patients by CSF oligoclonal IgG band status.

	CSF IgG oligoclonal bands			
	0	1–7	8–12	≥13
No. of subjects	55	55	55	54
Age at onset (years)	32.93 ± 0.88	30.86 ± 0.98	31.77 ± 0.87	31.84 ± 0.89
Females — %	33 (60.0)	37 (67.3)	35 (63.6)	36 (66.7)
Multifocal type of onset — %	11 (20.0)	8 (14.5)	7 (12.7)	9 (16.7)
Type of CIS — %				
Optic neuritis syndrome	16 (29.1)	15 (27.3)	18 (32.7)	13 (24.1)
Brainstem syndrome	10 (18.2)	17 (30.9)	10 (18.2)	21 (38.9)
Spinal cord syndrome	13 (23.6)	16 (29.1)	22 (40.0)	13 (24.1)
Other	11 (20.0)	8 (14.5)	7 (12.7)	9 (16.7)
CSF findings				
CSF cells	2.77 ± 3.93	4.88 ± 5.54	7.32 ± 9.47	3.91 ± 5.71
CSF proteins	33.0 ± 12.13	34.84 ± 13.71	36.13 ± 13.58	32.44 ± 12.49
Blood–brain barrier damage index	0.53 ± 0.24	0.52 ± 0.24	0.55 ± 0.22	0.49 ± 0.24
MRI findings				
T2 lesions	4.58 ± 3.93	4.78 ± 4.76	7.02 ± 5.04	12.11 ± 6.93
Gd-enhancing lesions	0.55 ± 1.14	0.75 ± 1.10	0.54 ± 0.93	0.60 ± 1.05

Plus–minus values are means ± SD.
Abbreviations: CDMS: clinically definite multiple sclerosis, CIS: clinically isolated syndrome, CSF: cerebrospinal fluid.