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Cerebrospinal Fluid Alzheimer's Disease Biomarkers in Cerebral Amyloid Angiopathy-Related Inflammation

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Abstract.
Background: Decreased cerebrospinal fluid (CSF) amyloid-β 1-40 ($A\beta_{40}$) and amyloid-β 1-42 ($A\beta_{42}$) and increased total and phosphorylated tau (t-tau, p-tau) concentrations have been described in cerebral amyloid angiopathy (CAA).
Objective: Our aim was to analyze these biomarkers in patients with CAA-related inflammation (CAA-I).
Methods: We prospectively recruited nine patients with acute phase CAA-I fulfilling Chung criteria. CSF was analyzed for t-tau, p-tau, $A\beta_{42}$, and $A\beta_{40}$. Data were compared to controls ($n = 14$), patients with Alzheimer's disease (AD, $n = 42$), CAA ($n = 10$), and primary angiitis of the central nervous system (PACNS, $n = 3$).
Results: For the CAA-I group, statistically significant differences were: lower $A\beta_{42}$ ($p = 0.00053$) compared to the control group; lower t-tau ($p = 0.018$), p-tau ($p < 0.001$), and $A\beta_{40}$ ($p < 0.001$) compared to AD; lower $A\beta_{42}$ ($p = 0.027$) compared to CAA; lower $A\beta_{42}$ ($p = 0.012$) compared to PACNS. Nearly significantly lower $A\beta_{40}$ ($p = 0.051$) and higher t-tau ($p = 0.051$) were seen in CAA-I compared to controls.
Conclusion: CSF biomarkers profile similar to that of CAA was observed in CAA-I (with even lower levels of $A\beta_{42}$ compared to CAA). Based on our findings, high p-tau seems more specific for AD, whereas low $A\beta_{42}$ differentiates CAA-I from CAA, PACNS, and controls, and low $A\beta_{40}$ differentiates CAA-I from AD.

Keywords: Alzheimer's disease, amyloid-β, cerebral amyloid angiopathy, cerebrospinal fluid, inflammation, tau

INTRODUCTION

In amyloid β (Aβ)-related angiitis of the central nervous system (CNS) (also called CAA-related inflammation, CAA-I), cerebral amyloid angiopathy

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(CAA) occurs in association with primary vasculitis of small- and medium-sized leptomeningeal and cortical arteries [1–6]. It has been suggested that CAA-I is triggered by vascular Aβ deposition followed by an Aβ-directed (auto)immune response, based on the presence of auto-antibodies against $A\beta_{40}$ and $A\beta_{42}$ and the clinical improvement most frequently encountered in response to immunosuppressive treatment. The apolipoprotein E (ApoE) $\varepsilon4/\varepsilon4$ genotype is predominant in CAA-I. In order to avoid brain biopsy, diagnostic criteria for probable CAA-I have been proposed by Chung et al. including all of the following: acute-subacute symptom onset, >40 years of age, at least one of the clinical features (headache, mental status, or behavioral change, focal neurological signs, seizures), patchy or confluent T2 or FLAIR hyperintensity, evidence of pre-existing CAA on susceptibility-weighted MRI sequences, and absence or neoplastic, infectious, or other cause [2].

Decreased cerebrospinal fluid (CSF) $A\beta_{42}$ and $A\beta_{40}$ and increased total and phosphorylated tau (t-tau and p-tau) concentrations have been described in CAA [7, 8]. In particular, $A\beta_{40}$ levels seemed to be of clinical interest to differentiate CAA from AD. To the best of our knowledge, CSF concentrations of these biomarkers have been analyzed only once in a series of CAA-I patients [9]. The authors found significantly higher t-tau and p-tau, equivalent $A\beta_{40}$, and non-significantly lower $A\beta_{42}$ levels in the acute phase of CAA-I compared with controls. During the remission phase of CAA-I, all biomarker levels decreased to levels significantly lower than in the acute phase. In their study, the authors essentially compared anti-Aβ autoantibodies in patients with CAA-I, CAA, multiple sclerosis, and control subjects, but they did not compare biomarker (CSF $A\beta_{42}$, $A\beta_{40}$, t-tau, and p-tau) levels to CAA or AD patients. Our aim was to analyze CSF biomarkers in clinically and radiologic well characterized patients with acute phase CAA-I and to compare them with patients with CAA, AD, primary angiitis of the CNS (PACNS) patients, and control subjects.

METHODS

Between November 2011 and November 2014, we prospectively included nine patients with CAA-I according to the Chung criteria in the three participating centers (CHU Nîmes, France; CHU Montpellier, France; CH Narbonne, France). None of our CAA-I patients had prior cognitive impairment according to standard criteria [10].

Informed consent to participate in CSF assessments and analysis was obtained from all patients. Lumbar puncture and CSF analyses were performed in conditions and with techniques previously described [8]. Analyses in CAA-I patients were compared with CSF data of prospectively recruited AD patients ($n = 42$, all meeting the criteria of probable AD defined by the NINCDS-ADRDA), CAA patients ($n = 10$), and controls subjects ($n = 14$) previously enrolled for biomarker analyses in CAA and with patients with PACNS ($n = 3$) [8]. In this study, however, we only took into account CAA patients with lobar hemorrhage (and thus excluding patients with isolated superficial siderosis only considered by the modified Boston criteria) according to the classical Boston criteria. All control patients underwent MRI including GRE sequences showing absence of abnormalities seen in CAA or CAA-I. In AD patients at time of CSF analysis, mean, standard deviation, median, and range values were respectively 19/30, 5.5, 21/30, and 5-27/30 for the Mini-Mental State examination and 34.5, 22.5, 34.5, and 2–84 for disease duration (months).

Graphic results were presented as medians and interquartile ranges. Statistical pairwise comparisons were performed with the non-parametric Kruskal–Wallis test using the Conover *post-hoc* method [11]. The H-score corrected for ties is indicated in the text after the *p*-values.

RESULTS

Clinical, radiological, and CSF characteristics of the nine CAA-I patients are summarized in Table 1. Age between the CAA-I ($n = 9$), CAA ($n = 10$), AD ($n = 42$), and control group ($n = 14$) did not differ significantly (mean age 70, 77, 73, and 69, respectively). However, PACNS patients ($n = 3$, mean age 58, range 53–64) were significantly younger than all other groups. CSF analysis was performed in the acute phase in all CAA-I patients, after a mean of 7.5 weeks (range 0.5–44 weeks) after symptoms onset. In one patient (patient nr 3), CAA-I-related symptoms were preceded by a large acute symptomatic lobar hemorrhage a few months earlier. FLAIR and GRE imaging of CAA-I patients are shown in the Supplementary Figure 1. Only two patients (patients 3 and 4) had histological analysis (lobar hemorrhage surgery in patient 3 and biopsy performed for symptoms related to CAA-I in patient 4), confirming CAA-I. ApoE genotype was determined for all CAA-I patients ($\varepsilon4/\varepsilon4$ in patients 1, 2, 4, 7, and 8; $\varepsilon2/\varepsilon3$ in patients 3 and 5; $\varepsilon3/\varepsilon4$ in patient

Table 1
Clinical, radiological, CSF data, and treatment and effect of treatment in CAA-I patients

Nr	Gender	Age	Symptoms	MB	LH	SS	Gado	Time LP	WBC	Protein	OCB	Treatment Effect
1	F	65	seizure, cognitive deficit	+++	0	0	NP	44 weeks	<3	0.45	No	No /
2	F	80	confusion, aphasia, cognitive deficit	+++	0	0	Mild lepto	6 weeks	<3	0.28	No	CS +
3	F	70	seizure, aphasia, cognitive deficit	3	2	FSS	Mild lepto	2 weeks	6	1.09	Yes	CS +
4	M	55	headache, cognitive and visual deficit	+++	0	0	Mild lepto	2 weeks	<3	0.54	No	CS/AZT/CPM +
5	F	78	apathy, gait disturb, confusion, cognitive deficit	+++	0	DSS	No	2 weeks	<3	1.12	NP	CS +
6	F	76	confusion, apathy, left neglect	+++	0	0	No	10 weeks	<3	0.31	No	CS +
7	M	71	apathy, confusion	+++	0	FSS	Mild lepto	4 weeks	<3	0.77	NP	CS +
8	M	66	confusion	+++	0	FSS	No	3 weeks	<3	0.43	NP	CS +
9	M	68	transient aphasia and faciobrachial paresthesias	+++	0	FSS	Mild lepto	0.5 week	<3	0.62	No	No /

Nr, patient number; MB, number of microbleeds; LH, lobar hemorrhage; SS, superficial siderosis; Gado, gadolinium enhancement on MRI; Time LP, delay between symptom onset and performance of lumbar puncture; WBC, number of white blood cells/mm^3 in CSF; Protein, protein level (g/L) in CSF; OCB, oligoclonal bands; +++, innumerable (>25 microbleeds); FSS, focal superficial siderosis (superficial siderosis restricted to 3 or fewer sulci); DSS, disseminated superficial siderosis (superficial siderosis affecting at least 4 sulci); NP, not performed; Mild lepto, mild leptomeningeal enhancement; CS, corticosteroids; AZT, azathioprine; CPM, cyclophosphamide.

6; $\varepsilon 3/\varepsilon 3$ in patient 9), but not systematically for the other groups. Treatment, given to seven patients (all including corticosteroid therapy), led to clinical and radiological improvement in all. The two remaining patients showed spontaneous improvement.

Results of CSF biomarkers in the different patient's groups are shown in Fig. 1. Median values (in pg/mL) for the nine CAA-I patients were: t-tau 333 (range 179–1200), p-tau 37 (range 26–71), $A\beta_{42}$ 400 (range 326–563), and $A\beta_{40}$ 9457 (range 4486–21205).

Compared to controls, CAA-I patients showed significantly lower $A\beta_{42}$ ($p < 0.001$) levels, nearly significantly lower $A\beta_{40}$ ($p = 0.051$) and higher t-tau ($p = 0.051$) levels, and comparable p-tau levels. When compared to AD, CAA-I showed significantly lower t-tau ($p = 0.018$), p-tau ($p < 0.001$), and $A\beta_{40}$ ($p < 0.001$) levels, but not significantly lower $A\beta_{42}$ ($p = 0.35$) levels. Compared to CAA, CAA-I had significantly lower $A\beta_{42}$ ($p = 0.027$) but not significantly lower t-tau ($p = 0.9$), p-tau ($p = 0.51$), and $A\beta_{40}$ ($p = 0.10$) levels. With respect to PACNS, CAA-I patients had significantly lower $A\beta_{42}$ ($p = 0.012$) but not significantly lower Aß$_{40}$ ($p = 0.35$) and higher t-tau ($p = 0.78$) and p-tau ($p = 0.17$). For AD, p-tau was the only biomarker significantly different (i.e., higher) compared to each other groups.

In the group of CAA-I group, biomarker values did not differ significantly between the $\varepsilon 4/\varepsilon 4$ and the non-$\varepsilon 4/\varepsilon 4$ patients.

One patient had a second CSF biomarker analysis (performed because of clinical and radiological relapse) three months after the first analysis/neurological episode (successfully treated by one month-lasting corticoid treatment). This second analysis showed 326 pg/ml (versus initial 286 pg/ml) for t-tau, 31 pg/ml for p-tau (versus initial 37 pg/ml), 416 pg/ml (versus initial 412 pg/ml) for $A\beta_{42}$, and 9,799 pg/ml (versus initial 12633 pg/ml) for $A\beta_{40}$. In this study, only CSF during the first episode was taken into account.

DISCUSSION

To the best of our knowledge, this is the first study to assess CSF $A\beta_{42}$, $A\beta_{40}$, t-tau, and p-tau levels in a series of CAA-I patients and to compare them with CAA, AD, PACNS patients, and controls. Based on our data, especially $A\beta_{42}$ and $A\beta_{40}$ levels seem to be of particular interest since the lowest levels of both biomarkers were found in the CAA-I group. $A\beta_{42}$ seemed to be the most specific CAA-I biomarker when

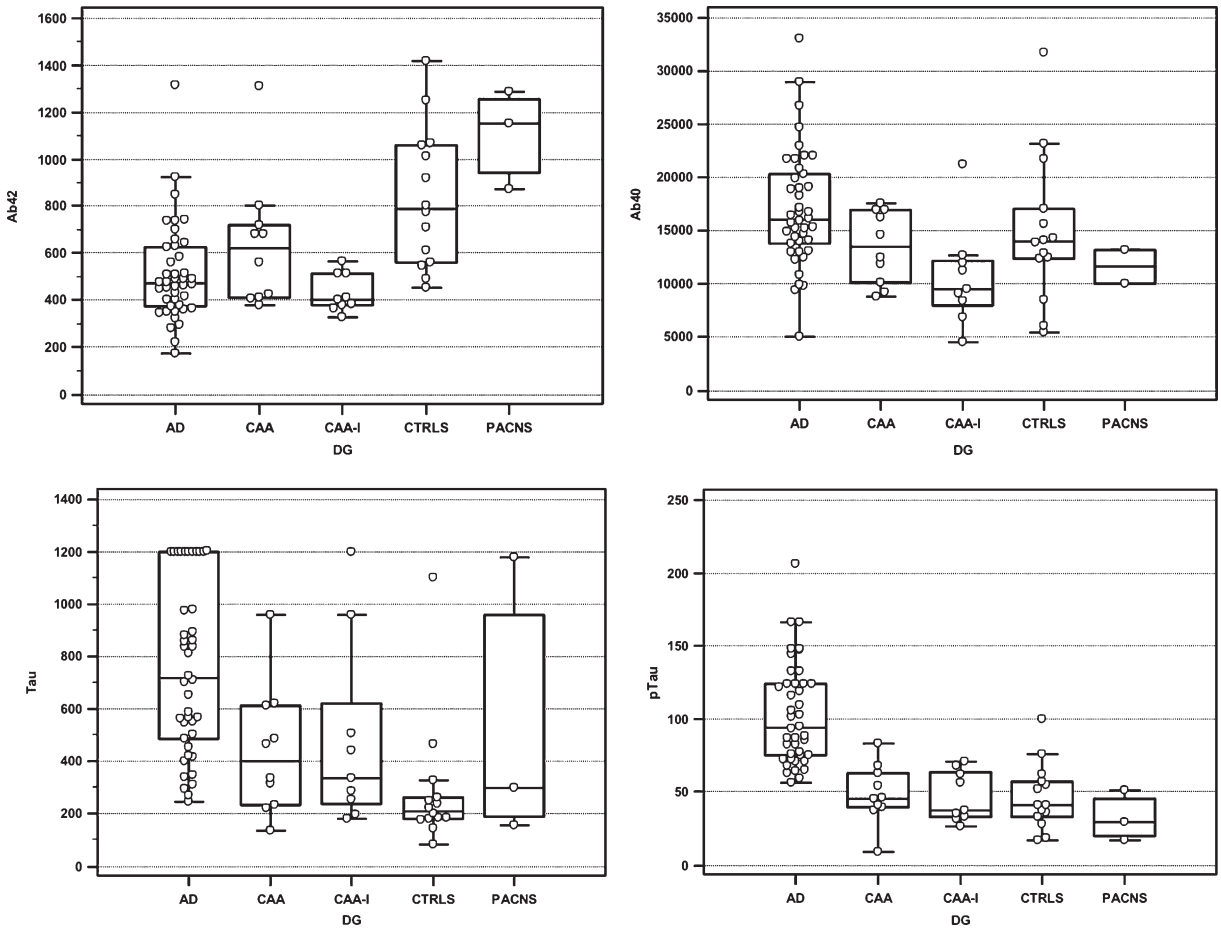

Fig. 1. Box-and-whisker diagrams, presented as medians and interquartile ranges of CSF $A\beta_{42}$, $A\beta_{40}$, t-tau, and p-tau levels in the AD patients ($n = 42$), CAA patients ($n = 10$), CAA-I patients ($n = 9$), controls ($n = 14$), and PACNS patients ($n = 3$).

compared to the other disorders: indeed, we observed levels significantly lower than CAA, PACNS, and control subjects, and non-significantly lower than AD. However, $A\beta_{42}$ and $A\beta_{40}$ levels showed important overlap between the different amyloid-related conditions making interpretation in individual patients difficult. CSF biomarkers, particularly $A\beta_{42}$, may vary with age. In our study, PACNS patients were significantly younger than the other groups, whereas the age between the CAA-I, CAA, AD, and control groups did not differ significantly. We performed statistical analyses comparing CAA-I patients with a subgroup of four controls with comparable age, and still found very significantly lower $A\beta_{42}$ levels, making less likely that the lower $A\beta_{42}$ levels in CAA-I compared to PACNS are only an age-dependent phenomenon. However, interpretation of biomarker levels in the PACNS group should be done with caution because of both a younger age and the limited number of these patients included in the study.

When dosing $A\beta_{42}$ and $A\beta_{40}$ levels, investigators and clinicians should bear in mind that amyloid may play a role (and thus potentially affect $A\beta_{42}$ and $A\beta_{40}$ levels) in other inflammatory and infectious CNS disorders usually not linked to primary amyloid-related pathophysiological processes (e.g., multiple sclerosis, human immunodeficiency virus) [12–15].

Based on our findings, when comparing CSF biomarkers in AD, CAA, CAA-I, PACNS, and control subjects, high p-tau seems to be the most specific biomarker for AD, whereas as mentioned above low $A\beta_{42}$ can differentiate CAA-I well from CAA, PACNS, and controls while low $A\beta_{40}$ can differentiate CAA-I from AD. Aβ concentrations in the CSF are the result of the dynamic equilibrium of brain production, clearance, and accumulation of Aβ. In CAA and

CAA-I, both $A\beta_{40}$ and $A\beta_{42}$ are probably trapped in the cerebral vasculature, whereas in AD predominant deposition of $A\beta_{42}$ in diffuse senile plaques probably leads to selective reduction of CSF $A\beta_{42}$ levels. Our data suggest that entrapment of $A\beta_{42}$ (and to a lesser degree $A\beta_{40}$) seems to be even more severe in CAA-I than in CAA, although other mechanisms influencing $A\beta_{42}$ and $A\beta_{40}$ levels cannot be excluded.

Compared with a previous study performed in CAA-I patients and showing non-significantly lower $A\beta_{42}$ levels in CAA-I acute phase as compared with controls, our study showed that $A\beta_{42}$ levels were significantly lower in CAA-I patients (also analyzed also in the acute phase) [9]. In Piazza and colleagues' study, $A\beta_{42}$ (together with $A\beta_{40}$, tau, and p-tau) levels decreased over time with lowest levels reached in the CAA-I remission phase. The time between symptom onset and CSF analysis seems to not explain the difference between the two studies since in our study CSF analysis was performed also in the acute phase and even earlier (mean of 7.5 weeks versus 3.8 months). Our CAA-I patients were slightly older than in the previous study (mean age of 70 versus 68), but this small age difference is probably insufficient to explain the difference found.

Comparing the biomarkers profile of CAA-I, CAA, and AD seems logical because of shared pathophysiological mechanisms. In daily practice, however, clinical and radiological abnormalities in CAA-I often suggest other differential diagnoses including vasculitis, tumor, and reversible posterior leukoencephalopathy syndrome. In our study, histology was available in a minority of CAA-I cases. Ideally, CSF biomarkers should be analyzed in a larger number of histologically proven CAA-I patients and compared to patients with other disorders mimicking CAA-I. If our observations can be confirmed, it might be interesting to integrate CSF data in the CAA-I diagnostic criteria in the future. Our study was mainly focused on AD biomarkers in CSF. In addition, dosing inflammatory markers (e.g., interleukin-6) and microglial-derived proteins (e.g., monocyte chemotactic protein-1 and YKL-40) might be interesting in order to see if these markers can help to differentiate inflammatory from non-inflammatory CNS disorders.

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SUPPLEMENTARY MATERIAL

The supplementary material is available in the electronic version of this article: http://dx.doi.org/10.3233/JAD-150621.

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