Evidence of Intrathecal Immunoglobulin Synthesis in Stroke

A Cohort Study

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Background: Immune mechanisms are included in stroke pathophysiologic factors, but the frequency and role of intrathecal antibodies is unclear and diagnostic tests are not routinely performed on cerebrospinal fluid (CSF).

Objective: To determine the frequency of intrathecal immunoglobulin synthesis in a well-characterized cohort of patients who experienced “noninflammatory” acute stroke.

Design: Retrospective cohort study.

Setting: University hospital neurology department.

Patients: Patients (n=318) with stroke who were undergoing lumbar puncture during diagnostic workup and 79 control patients.

Results: Cerebrospinal fluid–specific immunoglobulin (IgG, IgM, and IgA) synthesis was significantly (P<.001) more frequent after stroke (24.8%) compared with the incidence in age- and sex-matched controls (2.5%). Furthermore, 31.3% of stroke patients demonstrated blood-brain barrier dysfunction and 18.1% displayed pleocytosis.

Conclusion: The strong association between CSF-specific immunoglobulin synthesis and stroke suggests a role in the development of cerebral ischemia and might constitute an immunologically defined stroke subgroup.


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uncertainty. All patients underwent cerebral computed tomography or magnetic resonance imaging and lumbar puncture within 96 hours after symptom onset. After exclusion of inflammatory disease, consecutive age- and sex-matched patients who had received lumbar puncture during a diagnostic workup for headache (n = 24), diabetic oculomotor or abducens nerve palsy (n = 16), idiopathic facial nerve palsy (n = 29), and dizziness (n = 10) were used as controls (n = 79).

INTRATHECAL IMMUNOGLOBULIN SYNTHESIS

Albumin, immunoglobulin (Ig) G, IgA, and IgM from CSF and serum samples were quantified by routine nephelometry. Blood-brain barrier dysfunction was determined on the basis of age-related albumin quotients of CSF/serum. For detection of intrathecal antibody synthesis, the antibody index was calculated as the ratio between the CSF/serum quotient for IgG, IgM, and IgA antibodies and the CSF/serum albumin quotient using the Reiber gram calculation.¹ ² Oligoclonal bands were detected by isoelectric focusing with silver stain.³ Participation in the quarterly German quality-control survey for the detection of OBs revealed a 100% match during the study.

RESULTS

All 318 patients with stroke had radiologically confirmed cerebral ischemia and exclusion of CNS infection (eg, neurotropic viruses, Borrelia serologic findings, and antineuronal antibodies), demyelinating disease, head trauma, and intracerebral neoplasm. Age and sex data were not significantly different from those in the control group (Figure; A; Mann-Whitney and Fisher exact tests used for analysis of age and sex, respectively). The CSF cell counts ranged from 0 to 120/µL (mean [SD], 4.7 [13.3]/µL); 18.1% had pleocytosis (CSF cell count, ≥5/µL). Protein concentration ranged from 9.7 to 363.4 mg/dL (mean [SD], 64.6 [51.0] mg/dL); 32.9% had increased CSF protein (>45 mg/dL), and 33.1% had blood-brain barrier dysfunction. Immunoglobulin synthesis in the CSF compartment was present in 24.8% of patients with stroke: 17.9% revealing CSF-specific oligoclonal IgG bands using isoelectric focusing (several also with increased IgG antibody indices) and 6.9% showing increased CSF/serum antibody indices for IgM and IgA. Representative images show multiple strong OBs in selected patients with stroke (Figure, B). The frequency of OBs was significantly different from that of the 79 control patients without CNS disease, of whom only 2.5% had OBs (P < .001, Fisher exact test), and none of the control patients had pleocytosis.

In contrast to the population in a small study (N = 16),⁸ stroke patients with OBs were not significantly different in age from those without OBs (P = .87, Mann-Whitney 2-tailed test). Also, there was no association between the presence of OBs and sex, type of ischemia, frequency of pleocytosis, or blood-brain barrier dysfunction (Table). Pleocytosis was lymphocyte-dominant in both groups, with significantly fewer macrophages in patients with OB-positive stroke. Relevant previous illnesses (eg, tumor, rheumatoid disease) or current systemic infections were not significantly more frequent in patients with OB-positive stroke (Table).

To analyze whether increased frequency of OBs might be related to previous infarcts, we compared patients with first-ever stroke (n = 233) with those who definitely had old infarcts in addition to the current stroke (n = 50). The frequency of OBs, pleocytosis, and increased CSF protein, as well as age and sex, was not significantly different between the groups (data not shown).

Of the stroke patients without OBs, 12 underwent a second lumbar puncture beyond the 96-hour window of the present study. After 6 to 199 days (median, 19 days), 50% of the patients developed intrathecal immunoglobulin synthesis (4 with IgM after 6-19 days and 2 with IgG after 17-23 days). On the basis of this observation, the percentage of patients with OB-positive stroke might increase further with longer follow-up.

COMMENT

Intrathecal immunoglobulin synthesis was determined by the presence of oligoclonal IgG bands and of IgM and IgA antibody serum to CSF indices and was present in 24.8% of stroke patients in whom no infectious or autoimmune cause was identified during clinical workup. This unexpectedly high prevalence of OBs in this population may point to a direct association between CSF-specific immunoglobulin synthesis and focal cerebral ischemia. Because of the invasive nature of lumbar puncture, CSF samples from healthy individuals are not available for comparison. Our control group without any evident CNS disease had a low OB frequency of 2.5%, which is likely in the range of healthy individuals. Indeed, published studies on noninflammatory cohorts of between 134 and 207 patients (eg, having disk prolapse, headache, or dizziness) revealed OBs in none to 3.9% of the population.
Table. Characteristics of Patients With Ischemic Stroke

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Intrathecal Immunoglobulin Synthesis</th>
<th>No Intrathecal Immunoglobulin Synthesis</th>
<th>P Valuea</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, mean (SD), y</td>
<td>58.4 (14.4)</td>
<td>57.6 (16.0)</td>
<td>.22</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>55.3</td>
<td>47.4</td>
<td>.24</td>
</tr>
<tr>
<td>Female</td>
<td>44.3</td>
<td>52.3</td>
<td>.24</td>
</tr>
<tr>
<td>Pleocytosis, ≥5 WBC/µL</td>
<td>21.5</td>
<td>17.1</td>
<td>.40</td>
</tr>
<tr>
<td>BBB dysfunction</td>
<td>39.2</td>
<td>28.6</td>
<td>.28</td>
</tr>
<tr>
<td>Territorial infarction</td>
<td>87.3</td>
<td>80.9</td>
<td>.64</td>
</tr>
<tr>
<td>Lacunar infarction</td>
<td>7.7</td>
<td>5.4</td>
<td>.59</td>
</tr>
<tr>
<td>Macrophages, mean (SD), %</td>
<td>13.4 (8.8)</td>
<td>21.3 (14.9)</td>
<td>.01</td>
</tr>
<tr>
<td>Neutrophils, mean (SD), %</td>
<td>15.9 (29.4)</td>
<td>11.7 (22.0)</td>
<td>.42</td>
</tr>
<tr>
<td>History of tumor</td>
<td>7.3</td>
<td>13.2</td>
<td>.23</td>
</tr>
<tr>
<td>History of rheumatoid diseases (eg, SLE, APS, Sjögren syndrome)</td>
<td>9.8</td>
<td>9.6</td>
<td>&gt;.99</td>
</tr>
<tr>
<td>Systemic infection (eg, sinusitis, urinary tract infection, gastritis)</td>
<td>9.8</td>
<td>10.1</td>
<td>&gt;.99</td>
</tr>
</tbody>
</table>

Abbreviations: APS, antiphospholipid syndrome; BBB, blood-brain barrier; SLE, systemic lupus erythematosus; WBC, white blood cell.

aPoststroke cerebrospinal fluid cells were primarily lymphocytes.

bFisher exact test or Mann-Whitney test, 2-tailed, used in analysis.

In previous reports on cerebrovascular disease, limited by small sample size, the percentage of OBs varied widely between 7 of 14 patients (50%) with stroke identified using isoelectric focusing and 4 of 85 patients (5%) with acute cerebrovascular disease identified using agar gel electrophoresis. Oligoclonal bands were detected in 10 patients with stroke of another study, and 23 patients with infarct had higher CSF IgG levels compared with healthy control participants.

Diagnostic tests using CSF are not routinely performed in patients after cerebral ischemia events. In our cohort, additional symptoms (although not uncommon in stroke, eg, agitation, disorientation, and seizure) led to lumbar puncture to rule out inflammatory CNS disease. We cannot exclude the possibility that this selectivity enriched the retrospective cohort with more severely ill stroke patients.

Stroke-associated intrathecal immunoglobulin synthesis may result from (1) an underlying unidentified inflammatory disease, (2) undetected previous ischemic degeneration of neuronal tissue with repeated presentation of CNS antigen to the immune system, or (3) polyclonal nonspecific B-cell activation secondary to brain damage. The second explanation might be relevant to the high proportion of patients with OBs already present at the time of their first clinically detected stroke. The finding of OBs in patients with transient ischemic attacks supports this notion and implies relevance for predisease stages.

Because of the retrospective study design, it is unclear whether stroke-related intrathecal immunoglobulins represent specific antibody-mediated autoreactivity and whether this is relevant for pathologic factors that lead to stroke and clinical outcome (eg, determined with the use of the National Institutes of Health Stroke Scale). In a rodent study, induction of anti-neurofilament antibodies was associated with cognitive deficits. Similarly, our findings stimulate the question whether the high frequency of poststroke dementia (30%, often with atrophy) is associated with intrathecal immunoglobulin synthesis. Along these lines, it is tempting to speculate whether the recently defined CNS injury–induced immune depression syndrome might suppress overt humoral and cellular autoreactivity after CNS injury such as stroke.

We conclude that the unexpectedly high prevalence of intrathecal immunoglobulin synthesis in patients with stroke demands a systematic prospective analysis of CSF and serum samples to determine the time kinetics and pathogenicity of antibodies. Future experiments should evaluate antigen specificity and the relation to cellular immunity after stroke.

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REFERENCES