Cerebrospinal Fluid Markers in Dementia With Lewy Bodies Compared With Alzheimer Disease

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Background: Most patients with dementia with Lewy bodies (DLB) exhibit diffuse plaque–only pathology with rare neocortical neurofibrillary tangles (NFTs), as opposed to the widespread cortical neurofibrillary-tau involvement in Alzheimer disease (AD). Another pathological difference is the astrocytic and microglial inflammatory responses, including release of interleukins (ILs), around the neuritic plaques and NFTs in AD brains that are absent or much lower in DLB. We analyzed cerebrospinal fluid (CSF) markers that reflect the pathological differences between AD and DLB.

Objective: To determine CSF concentrations of tau, β-amyloid, IL-1β, and IL-6 as potential diagnostic clues to distinguish between AD and DLB.

Methods: We measured total tau, β-amyloid1-42, IL-1β, and IL-6 levels in CSF samples of 33 patients with probable AD without parkinsonism, 25 patients with all the core features of DLB, and 46 age-matched controls.

Results: Patients with AD had significantly higher levels of tau protein than patients with DLB and controls (P<.001). The most efficient cutoff value provided 76% specificity to distinguish AD and DLB cases. Patients with AD and DLB had lower, but not significantly so, β-amyloid levels than controls. The combination of tau and β-amyloid levels provided the best sensitivity (84%) and specificity (79%) to differentiate AD vs controls but was worse than tau values alone in discriminating between AD and DLB. β-Amyloid levels had the best correlation with disease progression in both AD and DLB (P=.01). There were no significant differences in IL-1β levels among patients with AD, patients with DLB, and controls. Patients with AD and DLB showed slightly, but not significantly, higher IL-6 levels than controls.

Conclusions: The tau levels in CSF may contribute to the clinical distinction between AD and DLB. β-Amyloid CSF levels are similar in both dementia disorders and reflect disease progression better than tau levels. Interleukin CSF concentrations do not distinguish between AD and DLB.

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A L Z H E I M E R D I S E A S E (AD) and dementia with Lewy bodies (DLB) are the 2 most common neurodegenerative dementias. Consensus clinical criteria for their diagnosis are commonly used with high sensitivity and specificity in specialized settings, but biological markers to differentiate both disorders are still lacking. Most cases with pure DLB exhibit diffuse-plaque–only pathology with rare or absent neocortical neurofibrillary tangles (NFTs), as opposed to the widespread cortical neurofibrillary-tau involvement in AD. In addition, astrocytic and microglial inflammatory responses, including release of interleukins, that occur around the neuritic plaques and NFTs of AD brains are absent or much lower around the diffuse plaques or Lewy body inclusions present in DLB brains.

The objective of this study was to determine the cerebrospinal fluid (CSF) concentrations of tau, β-amyloid, interleukin (IL) 1β (IL-1β), IL-6 in patients with clinically diagnosed AD and DLB as markers of the underlying pathological processes and potential diagnostic clues to distinguish between the 2 diseases.

METHODS

Cerebrospinal fluid samples were collected prospectively at the dementia clinics of the Fundación Jiménez Díaz and the Hospital Severo Ochoa, Madrid, Spain, from 1998 to 2002. To minimize the overlap between AD and DLB, cases were selected in accordance with strict clinical criteria. Patients with AD (n=33) were selected according to NINCDS-ADRDA (National Institute of Neurological and Communicative Disorders and Stroke–Alzheimer’s Disease and Related Disorders Association) criteria...
for “probable AD,”3 excluding patients with any parkinsonian features or visual hallucinations. Patients with DLB (n=25) were selected only if they fulfilled all the core criteria from the Consortium on DLB International Workshop.7 All selected patients had progressive dementia and spontaneous parkinsonism concomitant in the first 2 years of the disease, spontaneous visual hallucinations, and fluctuating cognition. Hallucinations and parkinsonism could not be attributed to medication. In 1 case, the diagnosis was pathologically confirmed.

Demographic and clinical characteristics are summarized in Table 1. Both groups included patients with a wide range of Mini-Mental State Examination (MMSE) scores (range, 5-26). There was no difference between the 2 groups in the mean MMSE scores or the mean duration of the disease when CSF samples were collected.

Samples of CSF from age-matched controls (n=46) were obtained from the neurology and emergency departments. Control cases had no cognitive impairment or symptoms of any neurodegenerative disease and were not in the acute phases of stroke or any inflammatory brain disease. Samples of CSF from control cases did not have pleocytosis or abnormal glucose or protein levels. Subjects with inflammatory diseases of the peripheral nervous system or with a possible inflammatory systemic disease (ie, blood erythrocyte sedimentation rate >25 mm/h) were also excluded as controls for the IL analysis.

After informed consent was obtained from subjects, CSF samples were collected by lumbar puncture, aliquoted, and stored at −80°C. Levels of β-amyloid1-42 and total tau protein were measured by sandwich enzyme-linked immunosorbent assay (Innotest Aβ1-42 and hTAU-Ag, respectively; Innogenetics NV, Gent, Belgium). Interleukin 1β and IL-6 were measured by sandwich enzyme-linked immunosorbent assay using commercial kits (Diaclone Research, Besançon, France). The sensitivity of these kits is less than 4 pg/mL for IL-1β and less than 2 pg/mL for IL-6. A positive control with a well-known IL concentration was included. In all analyses, duplicate samples were used according to the manufacturer’s protocol, and cases with more than 20% variability between samples were reanalyzed or excluded.

Groups were compared using analysis of variance or Kruskal-Wallis tests as appropriate according to the distribution of the data, with post hoc analyses. When differences were significant, diagnostic sensitivity and specificity were determined for several cutoff values. The sex distribution of the data, with post hoc analyses. When differences were significant, diagnostic sensitivity and specificity were determined for several cutoff values. The sex distribution of the data, with post hoc analyses. When differences were significant, diagnostic sensitivity and specificity were determined for several cutoff values. The sex distribution of the data, with post hoc analyses.

The distributions of the levels of the studied CSF markers were shown in the Figure. In general, the standard deviations for the markers were quite high in the 3 groups.

Patients with AD and DLB showed slightly higher IL-6 levels than controls, but the differences among the 3 groups were not significant. The combination of τ and β-amyloid levels by calculating the τ/β-amyloid quotient was significantly different in patients with AD vs controls (P<.001), but there were no significant differences when comparing patients with AD and patients with DLB or patients with DLB and controls. The τ/β-amyloid quotient best differentiated between the AD and control groups (cutoff point, 0.50; sensitivity, 84%; specificity, 79%; Table 3) but was less effective than τ values alone at discriminating between AD and DLB.

In all 3 groups, IL-1β levels were within the lowest values of the standard curve. In 10 cases (2 controls, 4 patients with DLB, and 6 patients with AD), IL-1β levels were undetectable (considered to be 0). There were no significant differences in IL-1β levels among patients with AD, patients with DLB, and controls.

Patients with AD and DLB showed slightly higher IL-6 levels than controls, but the differences among the 3 groups were not significant.

The distributions of the levels of the studied CSF markers are shown in Table 1. Correlations were observed among all 3 groups. When both dementia groups were analyzed together, the strongest correlation was between MMSE and β-amyloid levels (r=0.70, P=.007) and tau (r=0.52, P=.03). When both dementia groups were analyzed together, the strongest correlation was between MMSE and β-amyloid levels.

### Table 1. Demographic and Clinical Characteristics*<br>

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Controls (n = 46)</th>
<th>AD (n = 33)</th>
<th>DLB (n = 25)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>72.7 ± 7.8</td>
<td>73.8 ± 6.3</td>
<td>75.2 ± 4.8</td>
</tr>
<tr>
<td>Sex, FrM</td>
<td>22/24</td>
<td>23/10</td>
<td>12/13</td>
</tr>
<tr>
<td>Disease duration, mo</td>
<td>NA</td>
<td>26.7 ± 18.6</td>
<td>27.0 ± 24.6</td>
</tr>
<tr>
<td>MMSE score</td>
<td>NA</td>
<td>16.8 ± 6.0</td>
<td>18.0 ± 4.8</td>
</tr>
</tbody>
</table>

Abbreviations: AD, Alzheimer disease; DLB, dementia with Lewy bodies; MMSE, Mini-Mental State Examination; NA, not applicable.

*Data are presented as mean ± SD unless otherwise indicated. No comparisons were statistically significant.

Correlations between CSF MARKERS AND CLINICAL VARIABLES

None of the CSF markers correlated significantly with age in any of the 3 groups. There was no significant effect of sex in levels of CSF markers in any of the groups. In the DLB group, disease duration was significantly correlated with β-amyloid (r=−0.71, P=.007) and tau (r=−0.40, P=.06) levels and with MMSE score (r=−0.48, P=.04). In this group, there were no significant correlations between MMSE scores and the levels of any CSF marker. In the AD group, MMSE score was correlated with β-amyloid levels (r=0.49, P=.01), and disease duration was correlated with IL-1β levels (r=0.52, P=.03). When both dementia groups were analyzed together, the strongest correlation was between MMSE and β-amyloid levels (r=0.46, P=.006). Therefore, β-amyloid level was the CSF marker that best reflected disease progression, decreasing with a longer disease duration in DLB.

## RESULTS

### CSF MARKERS

Patients with AD had significantly higher tau levels than both patients with DLB and controls (P<.001 for all 3 groups; post hoc comparisons: control vs AD, P<.001; AD vs DLB, P<.01) (Table 2). There were no differences in tau levels between the DLB and control groups. The most efficient cutoff point (250 pg/mL) provided a 73% sensitivity for the diagnosis of AD with an 80% specificity vs controls and a 76% specificity vs DLB (Table 3).
Table 2. Comparison of Cerebrospinal Fluid (CSF) Markers Among the 3 Study Groups

<table>
<thead>
<tr>
<th>CSF Marker</th>
<th>Controls Mean ± SD, pg/mL (No. of Cases)</th>
<th>AD Mean ± SD, pg/mL (No. of Cases)</th>
<th>DLB Mean ± SD, pg/mL (No. of Cases)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>tau</td>
<td>200 ± 243 (46)</td>
<td>500 ± 399 (33)</td>
<td>213 ± 243 (25)</td>
<td>&lt;.001*</td>
</tr>
<tr>
<td>β-amyloid</td>
<td>462 ± 352 (34)</td>
<td>346 ± 309 (31)</td>
<td>320 ± 233 (14)</td>
<td>.25</td>
</tr>
<tr>
<td>tau/β-amyloid quotient</td>
<td>0.96 ± 3.37 (29)</td>
<td>5.26 ± 10.18 (25)</td>
<td>1.28 ± 1.87 (14)</td>
<td>&lt;.001†</td>
</tr>
<tr>
<td>Interleukin 1β</td>
<td>2.02 ± 3.28 (17)</td>
<td>2.17 ± 3.00 (20)</td>
<td>2.60 ± 2.97 (20)</td>
<td>.84</td>
</tr>
<tr>
<td>Only samples &gt;0</td>
<td>2.29 ± 3.41 (15)</td>
<td>3.10 ± 3.17 (14)</td>
<td>3.24 ± 2.98 (16)</td>
<td>.67</td>
</tr>
<tr>
<td>Interleukin 6</td>
<td>4.96 ± 4.72 (24)</td>
<td>7.14 ± 5.74 (22)</td>
<td>6.14 ± 5.00 (22)</td>
<td>.36</td>
</tr>
</tbody>
</table>

Abbreviations: AD, Alzheimer disease; DLB, dementia with Lewy bodies; NS, not significant.

*Post hoc comparison significant for tau: control vs AD P<.001; AD vs DLB P<.01.
†Post hoc comparison significant for tau/β-amyloid quotient: control vs AD P<.001.

Table 3. Sensitivity and Specificity for the Diagnosis of AD for Different Cutoff Points of tau Cerebrospinal Fluid Levels and tau/β-Amyloid Quotient

<table>
<thead>
<tr>
<th>Cutoff Point</th>
<th>Sensitivity for AD, %</th>
<th>Specificity, % AD vs Controls</th>
<th>Specificity, % AD vs DLB</th>
</tr>
</thead>
<tbody>
<tr>
<td>tau Level, pg/mL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>79</td>
<td>70</td>
<td>64</td>
</tr>
<tr>
<td>225</td>
<td>76</td>
<td>74</td>
<td>72</td>
</tr>
<tr>
<td>250</td>
<td>73</td>
<td>80</td>
<td>76</td>
</tr>
<tr>
<td>tau/β-amyloid quotient</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.50</td>
<td>84</td>
<td>79</td>
<td>36</td>
</tr>
<tr>
<td>0.75</td>
<td>68</td>
<td>90</td>
<td>57</td>
</tr>
<tr>
<td>1.00</td>
<td>56</td>
<td>93</td>
<td>71</td>
</tr>
</tbody>
</table>

Abbreviations: AD, Alzheimer disease; DLB, dementia with Lewy bodies.

This study shows that CSF concentrations of tau protein are significantly lower in DLB than in AD and that they may contribute to the clinical distinction of both neurodegenerative disorders. The most efficient cutoff point (250 pg/mL) provided a 76% specificity in distinguishing between the 2 types of dementia. Tau levels in CSF have been found to be significantly increased and well correlated with the amount of cortical NFTs in AD. In contrast, the normal tau levels found in patients with DLB are consistent with the pathological pattern described in DLB and likely reflect the low cortical neuritic deposition in these cases. Although clinical diagnostic criteria may be highly specific, a certain amount of error cannot be excluded, especially because dementia disorders share many clinical features. However, the difference in tau concentrations strongly supports the assertion that patients diagnosed with DLB and those with AD belong to different populations with different burdens of tau and NFT pathology in the central nervous system.

β-Amyloid levels were similar in patients with AD and DLB and tended to be lower than in controls, and they were not helpful in distinguishing between the 2 dementia groups. However, β-amyloid level best reflected disease progression in both dementia disorders, decreasing with a longer disease duration in DLB and with lower MMSE scores in AD. Previous studies have reported this correlation between β-amyloid CSF concentrations and several measures of disease severity in patients with AD.

The calculation of a tau/β-amyloid quotient provided the best distinction between patients with AD and controls, but it was worse than tau values alone at distinguishing between AD and DLB. Our data are similar to findings from previous studies in which the normality of tau levels distinguished DLB from AD cases but β-amyloid concentrations were similar in both groups.

A recent study suggests that the concentration of phosphorylated tau in CSF, which is well correlated with total tau levels, may provide a higher specificity to differentiate AD and DLB. Conversely, Buerger et al did not find that phosphorylated tau differentiated between AD and DLB better than did total tau levels, although phosphorylated tau better discriminates between AD and other degenerative dementias, particularly frontotemporal dementia.

Interleukin CSF levels did not distinguish between the 2 dementia disorders. To our knowledge, this is the first study specifically comparing these CSF inflammatory markers in AD and DLB. Interleukins are released by activated astrocytes and microglia and are detected around neuritic plaques and NFTs in AD brains. Some studies have reported increased CSF concentrations of IL-1β and IL-6 in patients with Alzheimer-type dementia compared with controls, although the increases were not always statistically significant. Variable results have been reported for groups of mixed non-Alzheimer dementia cases. This raises the issue of whether these markers are increased in any disorder with neuronal damage or whether there is a different pattern according to the amount of inflammatory response in the brain. Because glial responses are not much stimulated by diffuse plaques or Lewy body inclusions, we hypothesized that this pathological pattern could be reflected in CSF IL concentrations, and, therefore, we expected IL
levels to be lower in DLB than in AD. However, we did not find significant differences among the 3 groups, perhaps because most IL levels in controls but also in patients with dementia were clustered in the lowest concentrations. Interleukin 1β levels significantly increased with longer disease duration in AD cases.

In conclusion, our data show that CSF tau concentrations can serve as a clinical marker to differentiate DLB from AD and that β-amyloid levels probably reflect disease progression better than tau levels in DLB cases.

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Author contributions: Study concept and design (Drs Go´mez-Tortosa, García Ye´benes, and del Ser); acquisition of data (Drs Go´mez-Tortosa, Gonzalo, Fanjul, Sainz, Cantarero, and Cemillan); analysis and interpretation of data (Dr Go´mez-Tortosa); drafting of the manuscript (Drs Go´mez-Tortosa and Gonzalo); critical revision of the manuscript for important intellectual content (Drs Go´mez-Tortosa, Gonzalo, Fanjul, Sainz, Cantarero, Cemillan, García Ye´benes, and del Ser); obtained funding (Drs Go´mez-Tortosa, García Ye´benes, and del Ser); administrative, technical, and material support (Drs Go´mez-Tortosa, Gonzalo, Fanjul, Sainz, Cantarero, and García Ye´benes); study supervision (Dr Go´mez-Tortosa).

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Distribution of cerebrospinal fluid levels of tau protein (A), β-amyloid (B), the tau/β-amyloid quotient (C), and interleukins (IL) 6 (IL-6) (D) and 1β (IL-1β) (E) in controls, patients with Alzheimer disease (AD), and patients with dementia with Lewy bodies (DLB). Horizontal lines indicate the mean value for each group.

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