Sharpen That Needle

The article by De Meyer et al[1] in this month's issue of the *Archives* presents a novel method of analyzing cerebrospinal fluid (CSF) biomarker data and determining how these data map onto the clinical diagnoses of Alzheimer disease (AD), mild cognitive impairment (MCI), and healthy control subjects. Using a 2-component mixture model, they identified a signature of low β-amyloid 1-42 (Aβ1-42) level, high total tau protein (T-tau) level, and elevated phosphorylated tau protein 181 (P-tau 181) level that was detected in more than 90% of the AD group and only 39% of the control group; the MCI group was intermediate with 73%. Overall, the diagnostic sensitivity was 90% for AD with a specificity of 64% in 3 independent data sets. Further, the AD CSF signature of low Aβ1-42 level and high P-tau 181 level correctly identified 100% of MCI cases that progressed to AD within 5 years. This report solidifies the diagnostic importance of measuring Aβ, T-tau, and P-tau in CSF[2] and confirms the value of CSF measurements in predicting the conversion of MCI to AD.[3]

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There are 2 broad categories of biomarkers: biochemical and imaging. The quest for biochemical markers of AD in body fluids began in earnest with the "Consensus Report of the Working Group on Molecular and Biochemical Markers of Alzheimer's Disease" published in 1998.[4] Sponsored by the Ronald and Nancy Reagan Research Institute of the Alzheimer's Association and the National Institute on Aging, the working group provided guidelines for an ideal AD biomarker: it should detect a fundamental feature of the neuropathology and be validated in neuropathologically confirmed cases with a sensitivity of more than 80% for detecting AD and a specificity of more than 80% for distinguishing other dementias. Other desirable features included being technically reliable and reproducible, noninvasive, simple to perform, and inexpensive. The report concluded by citing low Aβ and high tau protein levels in CSF as coming closest to fulfilling these criteria. In the past 11 years, scores of studies have been published from investigators worldwide reporting that the formula of low Aβ1-42 level and high T-tau and P-tau 181 levels denote AD with sensitivities between 75% and 100% and specificities between 65% and 90%.[2]

The CSF Aβ and tau measurements meet the first and most important biomarker criterion of reflecting an important neuropathologic feature of AD. The chief histopathologic hallmarks of AD are senile plaques in the brain parenchyma and intraneuronal fibrillary tangles with neuropil threads. What is measured in the CSF reflects the biochemical composition of the anatomic lesions: the plaques are composed of amyloid fragments derived from a large amyloid precursor protein, whereas the tangles and threads are composed of hyperphosphorylated tau protein. Levels of Aβ1-42 and tau protein in the CSF accurately reflect the presence of substantial amyloid load and neurofibrillary abnormalities in the brain. Tapiola et al[5] analyzed antemortem CSF samples from 123 patients who subsequently had postmortem confirmation of diagnoses; 79 had a clinical diagnosis of AD and 34 had diagnoses of other neurological disease and non-AD dementias. They found that levels of Aβ1-42 inversely correlated with the total Aβ load in the brain and that tau protein levels correlated directly with the results of immunohistochemistry for hyperphosphorylated tau protein and with the presence of neocortical neurofibrillary tangles. In this study, the CSF formula gave an overall accuracy of 90% for the presence of pathologic neuritic plaques in the brain. In another autopsy-confirmed series of cases, Shaw et al[6] reported that low levels of CSF Aβ1-42 collected before death accurately distinguished subjects with a pathologic diagnosis of AD from healthy control subjects with a sensitivity of 96%. With the advent of molecular neuroimaging with amyloid radioligands such as Pittsburgh Compound B (PiB), it is now possible to visualize fibrillar Aβ deposits in the brain during life. Uptake of PiB corresponds well with the amount of Aβ in the brain examined at death[7] and also correlates well with CSF Aβ measurements. In a study by Fagan et al,[8] individuals with positive PiB binding had the lowest CSF Aβ1-42 levels, whereas those with negative PiB binding had the highest levels. These reports indicate that CSF Aβ and tau protein measurements accurately reflect major aspects of pathologic processes in the brain and that detecting low levels of Aβ1-42 in the CSF predicts high amyloid load in the brain.

The results of CSF Aβ and tau protein measurements do not stand alone as a single diagnostic test for AD as the positive percentage of agreement with diagnosis is never 100%. Low levels of Aβ1-42 may indeed always predict a high brain amyloid load, but brain amyloid deposits may occur in non-AD cases such as Parkinson disease and dementia with Lewy bodies as well as in cognitively normal individuals. Similarly, high tau protein levels are fairly nonspecific and may be seen after stroke, after traumatic head injury, and in Creutzfeldt-Jakob dis-

ease. Instead, the CSF findings should be viewed as an important adjunct to the diagnostic algorithm and complement the recommended practice of obtaining a careful history, conducting a comprehensive examination, completing neuropsychological testing, and performing brain imaging. This recommendation is consistent with the revised research criteria for the diagnosis of AD proposed by Dubois et al.[9] According to these guidelines, the core diagnostic criterion is a gradual, slowly progressive episodic memory impairment that is documented on formal testing. In addition to this core criterion, there must be at least 1 supporting feature from among 4 possible measurements: (1) presence of medial temporal lobe atrophy on magnetic resonance brain scan; (2) abnormal CSF profile of low Aβ1-42 level, increased T-tau level, and increased P-tau level; (3) reduced glucose metabolism in bilateral temporal parietal regions or increased Aβ radioligand uptake on positron emission tomographic scans; or (4) a proven autosomal dominant genetic mutation. Diagnostic certainty increases with confirmation from each additional supporting domain.

The AD signature of altered Aβ and tau protein levels in CSF may have other applications in addition to aiding in diagnosis. De Meyer and colleagues called attention to its possible predictive value in detecting AD in advance of clinical signs and symptoms. In their series, 39% of the nondemented healthy control subjects had levels of Aβ1-42 that fell in the AD range. As they pointed out, this percentage is similar to that in healthy control subjects with positive PiB scan results and with amyloid plaques in brains of nondemented individuals at autopsy. There are some data suggesting that these individuals may be on the path to develop AD.[10,11] If this were true, it would account for specificities being lower than sensitivities as these cases would be considered errors—that is, having a pattern of Aβ and tau protein levels in the CSF that is suggestive of AD but not having AD clinically. Nonetheless, these individuals as a group may be exactly the ones who should be targeted for antiamyloid therapies when they become available. It is already clear that CSF biomarkers can distinguish individuals with MCI and accurately identify those who will progress to AD.[1,3,6] The CSF biomarkers may have additional value in predicting the course of AD. Snider et al[12] found that the rate of dementia progression was significantly more rapid in those with the lowest baseline levels of Aβ1-42 and higher levels of T-tau and P-tau. Repeated analyses of Aβ over the course of illness are not useful in tracking the course of degeneration as the levels tend to remain constant, whereas levels of T-tau and P-tau tend to increase somewhat with dementia severity. Whether these analytes can be used as change measurements during therapeutic intervention must await discovery of a safe and effective drug that alters the trajectory of decline in AD.

Other stipulations of an ideal biomarker were that it be noninvasive, simple to perform, and inexpensive. Collecting CSF is simple for an experienced physician, although it requires more time and preparation than obtaining a blood sample from a vein. Whether it is noninvasive or not is in the eye of the beholder: performing a lumbar puncture (LP) is no more invasive than other outpatient procedures such as endoscopies that millions of Americans tolerate each year. The adverse effect profile is favorable: there is virtually no mortality and the risk of morbidities such as infection and post-LP headache is well under 5%. Expense is a relative measurement and should be viewed in a cost-benefit context; in most centers, the consulting physician's bill, the charge for neuropsychological testing, and the cost of a magnetic resonance brain scan (let alone that of a fludeoxyglucose F 18–positron emission tomographic or PiB scan) are all greater than obtaining CSF Aβ and tau protein values.

Measurements of CSF Aβ and tau protein levels are not perfect biomarkers for AD, but they fulfill almost all of the desirable features outlined by the working group in 1998. We believe that the scientific evidence accumulated during the past decade indicates that CSF analyses do provide important information that should be part of clinical diagnosis and care. In the future, additional analytes in CSF may be discovered to supplement Aβ and tau protein measurements. Of putative markers examined to date, however, only isoprostanes show promise; however, this lead is not yet as fully documented as Aβ and tau levels.

The next step in biomarker development should be education—educating patients that the test is safe and gives useful information, and educating physicians that the Aβ and tau protein measurements aid in their diagnosis and care of patients. There are some biases to overcome. For patients, the procedure is often perceived as painful, difficult to perform, and dangerous. Apocryphal stories circulate about chronic post-LP pain and paralysis. Even the terms *spinal tap* and *lumbar puncture* are threatening; perhaps a more neutral description such as *spinal fluid collection*, comparable to *blood draw*, followed by a full explanation of the procedure would be helpful. Physicians, nurses, and lay organizations such as the Alzheimer's Association need to undertake this education process in much the same way as autopsy and brain donation became a mantra among patients and families of patients with AD. The initial concern that discussions of autopsy would upset people was quickly dispelled, and autopsy rates in academic AD centers are well over 50%.

Physician education is also necessary, and articles such as the one by De Meyer and colleagues go a long way toward building a compelling case for CSF analyses, defining the conditions in which these are likely to be helpful, and showing how to interpret the results. There are practical matters as well in that most patients with MCI and AD are diagnosed and treated by their family physicians; neurologists see only a tiny minority of the estimated 3 to 4 million patients with AD in the United States. Most family physicians and internists are not skilled or experienced in performing an LP and therefore would not embrace CSF analyses unless they could refer patients to a central facility. Neurologists are trained to perform LPs, but those in busy practice and on the senior staff at teaching hospitals may have done very few since their residency. They too might welcome the idea of establishing or expanding an LP clinical unit in their outpatient practice or hospital setting and devoting a morning every week (or month, according to the need) to collecting CSF.

To date, CSF analyses have not been a routine component of assessment and care for patients with cognitive impairments and suspected AD in the United States. There is now ample evidence that these measurements have value; physicians need to formulate when and how to incorporate CSF measurements into their practice. We strongly recommend CSF analyses of Aβ1-42, T-tau, and P-tau in circumstances where having a definitive diagnosis of AD is important for counseling patients about such concerns as work, driving, and making other lifestyle changes. The CSF biomarkers will also improve accuracy for determining treatment in clinical situations where other conditions, such as normal-pressure hydrocephalus, depression, or vascular ischemic changes, figure in the differential diagnosis. There is already ample evidence that the AD CSF signature has a place in predicting which individuals with MCI are most at risk to progress to dementia, and it may even have value in predicting the rate of cognitive decline. The CSF analyses of Aβ, T-tau, and P-tau should have a central place in experimental clinical trials to increase the likelihood that participants have AD and eliminate other diagnoses that might dilute treatment effects. Gazing into the future when there are neuroprotective medications for AD, we can envision a recommendation that CSF analyses be implemented as a screening test to identify clinically healthy individuals at risk for MCI and AD. The information gained would enable early application of treatments to delay onset of symptoms or slow progression of cognitive impairments.

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Correction

Errors in Table. In the article "Evidence of Syntaxin 1A Involvement in Migraine Susceptibility: A Portuguese Study," published in the April issue of the *Archives* (2010;67[4]:422-427), there were a few errors in Table 3. In the rs941298 section of the table, the CC and TT genotype cells in the "All Patients With Migraine" column should be switched. The number and frequency for the TT genotype should be 28 (14.9%) and for the CC genotype, 86 (45.7%). The CC and TT genotype cells should also be switched in the "Controls" column: the number and frequency for the TT genotype should be 28 (9.8%) and for the CC genotype, 142 (49.5%).