

# UCSF

## UC San Francisco Previously Published Works

### Title

Multiple comorbid neuropathologies in the setting of Alzheimer's disease neuropathology and implications for drug development

### Permalink

<https://escholarship.org/uc/item/5pv5p99k>

### Journal

Alzheimer's & Dementia: Translational Research & Clinical Interventions, 3(1)

### ISSN

2352-8737

### Authors

Rabinovici, Gil D

Carrillo, Maria C

Forman, Mark

et al.

### Publication Date

2017

### DOI

10.1016/j.trci.2016.09.002

### Copyright Information

This work is made available under the terms of a Creative Commons Attribution-NonCommercial-NoDerivatives License, available at

<https://creativecommons.org/licenses/by-nc-nd/4.0/>

Peer reviewed

Perspective

# Multiple comorbid neuropathologies in the setting of Alzheimer's disease neuropathology and implications for drug development

Gil D. Rabinovici<sup>a</sup>, Maria C. Carrillo<sup>b</sup>, Mark Forman<sup>c</sup>, Susan DeSanti<sup>d</sup>, David S. Miller<sup>e</sup>,  
Nicholas Kozauer<sup>f</sup>, Ronald C. Petersen<sup>g</sup>, Christopher Randolph<sup>h,i</sup>, David S. Knopman<sup>g</sup>,  
Eric E. Smith<sup>j</sup>, Maria Isaac<sup>k</sup>, Niklas Mattsson<sup>l,m</sup>, Lisa J. Bain<sup>n</sup>, James A. Hendrix<sup>b,\*</sup>,  
John R. Sims<sup>o</sup>

<sup>a</sup>Memory & Aging Center, Department of Neurology, University of California, San Francisco, San Francisco, CA, USA

<sup>b</sup>Division of Medical & Scientific Relations, Alzheimer's Association, Chicago IL, USA

<sup>c</sup>Merck, Kenilworth, NJ, USA

<sup>d</sup>Piramal Pharma, Inc., Boston, MA, USA

<sup>e</sup>Bracket Global, Wayne, PA, USA

<sup>f</sup>Quintiles (formerly), Rockville, MD, USA

<sup>g</sup>Department of Neurology, Mayo Clinic and Foundation, Rochester, MN, USA

<sup>h</sup>MedAvante, Hamilton, NJ, USA

<sup>i</sup>Department of Neurology, Loyola University Medical Center, Maywood, IL, USA

<sup>j</sup>Department of Clinical Neurosciences, Hotchkiss Brain Institute, University of Calgary, Calgary, Alberta, Canada

<sup>k</sup>European Medicines Agency (EMA), London, UK

<sup>l</sup>Clinical Memory Research Unit, Department of Clinical Sciences Malmö, Lund University, Malmö, Sweden

<sup>m</sup>Department of Neurology, Skåne University Hospital, Lund, Sweden

<sup>n</sup>Independent Science Writer, Elverson, PA, USA

<sup>o</sup>Eli Lilly & Co., Indianapolis, IN, USA

## Abstract

Dementia is often characterized as being caused by one of several major diseases, such as Alzheimer's disease (AD), cerebrovascular disease, Lewy body disease, or a frontotemporal degeneration. Failure to acknowledge that more than one entity may be present precludes attempts to understand interactive relationships. The clinicopathological studies of dementia demonstrate that multiple pathologic processes often coexist.

How overlapping pathologic findings affect the diagnosis and treatment of clinical AD and other dementia phenotypes was the topic taken up by the Alzheimer's Association's Research Roundtable in October 2014. This review will cover the neuropathologic basis of dementia, provide clinical perspectives on multiple pathologies, and discuss therapeutics and biomarkers targeting overlapping pathologies and how these issues impact clinical trials. High prevalence of multiple pathologic findings among individuals with clinical diagnosis of AD suggests that new treatment strategies may be needed to effectively treat AD and other dementing illnesses.

© 2016 The Authors. Published by Elsevier Inc. on behalf of the Alzheimer's Association. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

**Keywords:** Alzheimer's disease (AD); Cerebrovascular disease; Lewy body disease; Frontotemporal degeneration;  $\beta$ -amyloid; Tau;  $\alpha$ -Synuclein; TDP-43

The views expressed in this article are the personal views of the authors and may not be understood or quoted as being made on behalf of or reflecting the official position of the European Medicines Agency or any of its Committees or Working Parties.

\*Corresponding author. Tel.: 312-604-1650; Fax: 877-444-6282.

E-mail address: [jhendrix@alz.org](mailto:jhendrix@alz.org)

## 1. Introduction

Converging research has shown the complexity of multiple pathologic substrates underlying clinical Alzheimer's disease (AD), with closely related and perhaps synergistic pathologic

processes often involved. A better understanding of these co-pathologies (i.e., cerebrovascular disease, Lewy bodies [LBs], hippocampal sclerosis [HS] with or without TAR DNA-binding protein 43 [TDP-43] inclusions, non-AD tauopathies, neuroinflammation, etc.) could influence clinical trial strategies and outcomes and lead to novel therapeutic approaches.

## 2. Methods

How overlapping pathologic findings affect the diagnosis and treatment of clinical AD and other dementia phenotypes was the topic taken up by the Alzheimer's Association's Research Roundtable in October 2014. The meeting provided a forum for experts from academia, industry, funding and regulatory agencies, and payer groups to consider the implications of overlapping pathologic findings on the discovery of new drug targets, therapeutic approaches, trial designs, and the regulatory approval of new drugs. The objective of this article is to provide a summary of the topics discussed at the Research Roundtable meeting and provide a review of the latest understanding of these issues and their implications for drug development in AD.

## 3. Discussion

### 3.1. The neuropathologic basis of dementia

Although neuropathologic diagnosis is considered the gold standard for determining the etiologic cause of a dementia syndrome, this approach has limitations. Neuropathologic examination is the only technique that enables visualization of abnormal structures at a microscopic level—plaques, tangles, LBs, neuronal loss, infarcts, etc.—yet it provides only a snapshot of the brain at one point in time. Neuropathology provides limited information regarding the age of lesions, their relationship to one another over time, or their relationship to the real-time clinical characteristics of the disease. Longitudinal cohort studies with annual testing have improved clinical correlation with pathology. Indeed, observational studies such as the longitudinal Nun Study, which began in 1986 with annual examinations and by 1997 included autopsies on 146 participants, found pathologic indicators of AD even in the brains of cognitively normal individuals evaluated clinically proximate to death [1]. These studies, along with amyloid positron emission tomography (PET) imaging and cerebrospinal fluid (CSF) biomarkers, support the concept of pre-clinical disease which is now codified into the revised National Institute on Aging and Alzheimer's Association (NIA-AA) diagnostic guidelines [2–4].

After the publication of the revised diagnostic criteria, it became clear that the criteria for postmortem pathologic diagnosis of AD also required updating to reflect knowledge that has accumulated since the last consensus criteria were published in 1997 [5]. These new criteria, also proposed by an NIA-AA working group [6], aim to disentangle the

clinicopathologic term “Alzheimer's disease” from the neuropathologic changes seen in the AD brain, including brain lesions that reflect comorbid conditions that are common among the elderly.

These new criteria recommended classifying “AD neuropathologic change (ADNC)” according to three different staging schemes: the distribution of amyloid  $\beta$  ( $A\beta$ ) deposits with Thal stages [7], neurofibrillary pathology with Braak stages [8,9], and the presence and severity of neuritic plaques according to the Consortium to Establish a Registry for Alzheimer's Disease [10]. Combining results from these three variables yields an estimate of no, low, medium, or high ADNCs. Those with intermediate or high ADNC are considered to have sufficient pathology to confirm a clinical diagnosis of AD dementia during life. The working group also made recommendations regarding how to report findings for common comorbidities, including Lewy body disease (LBD), vascular brain injury (VBI), and HS, and other neuropathologic findings such as TDP-43 inclusions and argyrophilic grain disease (AGD). LBD, VBI, HS, and TDP-43 inclusions have all been shown to independently contribute to cognitive impairment [11–14], whereas the clinical significance of AGD is less well established [15].

The new NIA-AA neuropathologic criteria for AD reflect recent studies suggesting that a minority of persons with clinical diagnosis of AD have “pure AD,” that is, only plaque and tangle pathology. For example, in the State of Florida Department of Elder Affairs Alzheimer's Disease Initiative (ADI) Brain Bank at Mayo Clinic in Jacksonville, which in 2014 included 1242 brains collected from memory disorders clinics across the state, fewer than half of all patients with a primary pathologic diagnosis of AD had “pure AD” neuropathology (Fig. 1) (unpublished data).

Another analysis by the Religious Orders Study (ROS) and the Rush Memory and Aging Project found that 45.8%

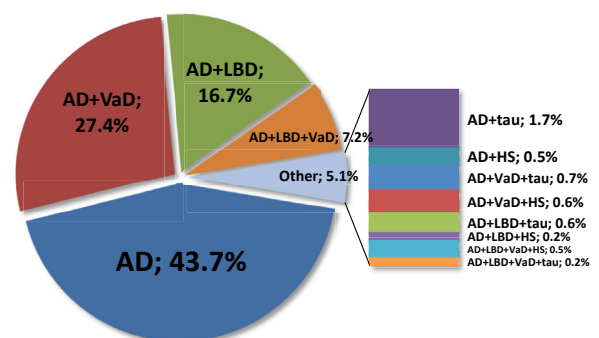


Fig. 1. Multiple pathologic findings when AD is present. A total of 1242 brains were included: all Braak stage IV or greater and Thal phase 3 or greater. Eighty-six percent of patients had a primary clinical diagnosis of dementia and 14% were diagnosed with parkinsonism. “Tau” indicates argyrophilic grain disease, an age-associated medial temporal tauopathy. Abbreviations: AD, Alzheimer's disease; VaD, vascular dementia; LBD, Lewy body disease; HS, hippocampal sclerosis.

of individuals with probable AD dementia and 19.4% of those with mild cognitive impairment (MCI) had multiple pathologies [16]. Other studies have shown that between 30% and 57% of brains with AD pathologic diagnosis have TDP-43 pathology in the medial temporal lobe [17,18] and that cases with TDP-43 pathology have greater regional brain atrophy and more severe cognitive impairment [18–20]. The most significant predictor of TDP-43 pathology in AD is HS [21]. In the Florida ADI Brain Bank, HS was detected in 20% of patients with dementia [22]. In a separate study, almost half of AD cases were shown to have LBs. LB copathology in AD is further classified based on the anatomic distribution of LBs: brainstem, limbic, or diffuse cortical. In some cases, LB copathology in AD is confined to the amygdala. Such cases are sometimes classified as the LB variant of AD [23].

The Rush study also examined the influence of overlapping pathologic processes on cognitive impairment. The majority of patients with an antemortem clinical diagnosis of AD dementia were found to have multiple pathologies at autopsy, with about half having, in addition to AD, one or more of the following: LBs, TDP-43, HS, and vascular diseases. Each pathology appears to have separate but additive effects on cognition. How these different pathologic processes interact remains unclear, although some experimental data suggest that A $\beta$  deposition may affect tau [24,25] and  $\alpha$ -synuclein accumulation [26].

Moreover, the distribution of neurofibrillary tangles in patients with AD does not always conform to typical Braak staging. A study that queried the brain bank database at the Mayo Clinic in Jacksonville, FL, found that of 889 AD cases studied, about a quarter fell into two “atypical” patterns: 11% had hippocampal sparing (HpSp) and 14% had limbic-predominant (LP) tangle pathology [27]. Interestingly, the HpSp subgroup experienced more rapid decline, whereas the LP subgroup showed a slower rate of decline. Those in the HpSp subgroup also tended to be younger, more often present with nonamnestic clinical syndromes, and showed shorter disease duration and less concurrent pathology.

Vascular lesions are also frequently comorbid with AD and contribute to cognitive decline. In the ADI analysis, 44% of AD cases also had vascular pathology, including 28% with small-vessel disease (lacunar infarcts, microinfarcts, or cribriform change), 12% with severe leukoencephalopathy, 10% with severe amyloid angiopathy, 2% with large-vessel infarcts, and <1% with large hematomas (personal communication, presented at Research Roundtable by Dennis Dickson). The ROS/Rush study measured different frequencies: among patients with probable AD, approximately one-third had gross infarcts, one-fourth had microinfarcts, and about one-third had moderate-to-severe cerebral amyloid angiopathy (CAA) [28]. In addition, one-third of probable AD patients had moderate-to-severe atherosclerosis and moderate-to-severe arteriolosclerosis.

### 3.2. Clinical perspectives on multiple pathologies

The prevalence of most neuropathologic findings increases with age. Therefore, the high rate of multiple pathologies found in neuropathologic studies, such as those described previously, may be related to the high mean age of death (mean  $\sim$ 90 years) of participants in most autopsy studies [14]. At the other end of the spectrum, clinical research typically involves younger patients with fewer comorbid pathologic findings, meaning that the problem is less of an issue in younger dementia patient cohorts. Researchers should consider copathologies in clinical trials, although the prevalence and impact may be lower in a younger population than the autopsy studies, composed of older populations, may suggest.

According to data collected by the National Alzheimer's Coordinating Center using the 1997 NIA-Reagan neuropathologic criteria as the gold standard [5], the sensitivity of clinical diagnosis of AD ranged from 70.9% to 87.3% and the specificity ranged from 44.3% to 70.8% [29]. Among those subjects who were misdiagnosed, the primary neuropathologic findings were tauopathies such as AGD, as well as frontotemporal lobar degeneration (FTLD), cerebrovascular disease, LBD, and HS.

FTLD comprises a family of disorders, differentiated clinically by behavioral, language (semantic or agrammatic), or parkinsonian variants (progressive supranuclear palsy and corticobasal degeneration). The neuropathologic diagnosis of FTLD is further broken down by the presence of tau, TDP-43, or the RNA-binding protein fused in sarcoma (FUS) pathology [30]. The links between clinical phenotype and pathology vary for each syndrome, and both the clinical and pathologic features overlap with AD. When evidence of both frontotemporal dementia (FTD) and AD pathologic processes coexists in a patient, both may contribute to the clinical phenotype [31].

LBD symptoms can be differentiated by fluctuating cognition, visual hallucinations, Parkinsonism, and rapid eye movement sleep behavior disorder. While most non-AD pathologic processes increase with age, LB frequency either plateaus or declines in older cohorts, further complicating diagnosis when multiple pathologies are present. Neuropathology studies have found divergent results regarding the clinical effects of concomitant LB pathology in AD. Some studies report that the combination of AD and LB pathology affects the cognitive presentation of dementia [32,33] but not the rate of cognitive decline [33]. However, one recent study failed to find clinical or neuropsychological predictors that could differentiate between AD with or without concomitant LB [34].

In AD, TDP-43 pathology is found in about 20% to 50% of AD brains, particularly in the amygdala and medial temporal lobe [18,35], and correlates with more rapid cognitive decline, even after controlling for other pathologies, particularly in the domains of episodic and working memory (Fig. 2) [20]. Moreover, TDP-43 deposition appears to progress in a stereotypic manner similar to tau, which

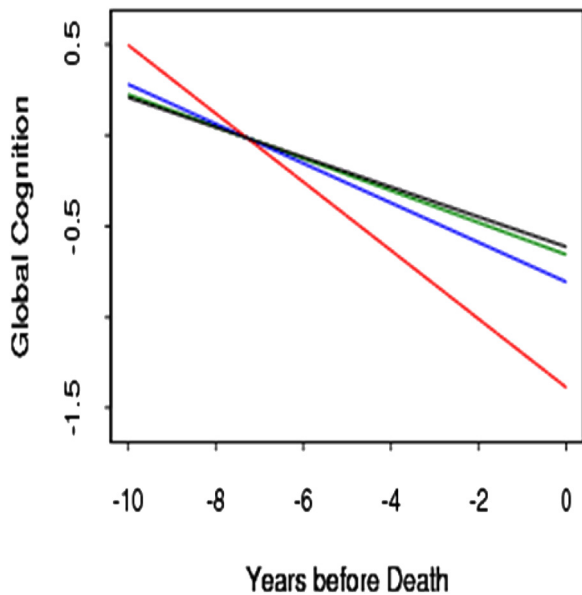


Fig. 2. Ten-year paths of global cognitive decline in 130 participants from the Religious Order Study with varying levels of TDP-43 pathology, adjusted for age at death, amyloid, tangles, and hippocampal sclerosis. Black line, no TDP-43; green line, 10th percentile; blue line, 50th percentile; and red line, 90th percentile TDP-43 [20].

gives rise to a staging scheme [18] that correlates with cognition, memory loss, and medial temporal atrophy [36].

The presence of FTLN or LBD pathology can be predicted albeit with moderate sensitivity and specificity, on clinical grounds. For cerebrovascular disease, the Hachinski Ischemic Score has been widely used to identify vascular dementia on clinical grounds but also with only moderate sensitivity and specificity for separating pure AD from AD with cerebrovascular copathology [37]. The addition of magnetic resonance imaging (MRI) allows identification of clinically unrecognized brain infarcts, microbleeds, and white matter hyperintensities of presumed vascular origin. However, MRI signs of cerebrovascular disease may not always be clinically relevant for cognition, and our understanding of the relationship between cerebral small-vessel disease burden and risk for dementia is incomplete. Furthermore, commonly available MRI (1.5 or 3.0 T) cannot detect microinfarcts, which are very common and are associated with dementia risk [38]. Therefore, MRI is a useful adjunct to increase the sensitivity for detection of cerebrovascular disease but still does not capture the full spectrum of cerebrovascular pathology. In the ROS, macroscopic infarcts, microinfarcts, and CAA were independently and additively associated with antemortem cognitive decline and/or risk for dementia, including when AD pathology was additionally present [39–41]. In clinical trials, comorbid cerebrovascular pathology could affect trial results or even obscure beneficial effects on AD if there are off-target drug effects. For example, patients with CAA appear to be at higher risk for amyloid-related imaging abnormalities in AD immunotherapy trials [42].

### 3.3. Therapeutics/targeting overlapping pathology

Emerging evidence suggests that AD and other neurodegenerative diseases may be driven by cell-to-cell transmission of pathologic proteins such as A $\beta$ , tau,  $\alpha$ -synuclein, and TDP-43 [43]. These proteins appear to become pathologic through conformational changes such as misfolding, oligomerization, fibrillization, and aggregation. These abnormal conformers then appear to serve as seeds or templates for assembly of additional protein aggregates that can be transmitted neuron to neuron [44–46]. This process could explain the spatiotemporal progression of pathology in the brain. While the mechanisms of this process are only beginning to be understood, it has nevertheless given rise to novel therapeutic approaches. For example, antibodies or small molecules may be generated that target different protein structures, either stabilizing them so they do not form fibrils or aggregates or by preventing their transmission [47,48]. Clinical trials are already in progress testing this strategy. For example, there is a phase I trial of an antibody against  $\alpha$ -synuclein in patients with Parkinson's disease (PD) (NCT02095171 and NCT02157714), antitau antibodies have shown promising results in animal models of tauopathies [49], and early human studies are just getting underway.

Elucidating the mechanisms whereby abnormal conformers of key pathogenic proteins induce neurodegeneration could also reveal new treatment approaches. Toxicity from the RNA-binding protein TDP-43, for example, appears to result from both gain- and loss-of-function mechanisms, with posttranslational modifications conferring toxic properties that disrupt normal transport of mRNA to the synapse [50]. The mechanisms of TDP-43 misregulation and propagation are unknown. New animal models and a TDP-43-specific imaging agent could speed the identification of therapeutic targets and the development of new drugs.

Targeting amyloid in the vasculature presents additional complexity. In the ROS/Rush studies mentioned earlier, CAA was present in about 80% of individuals at autopsy, and the severity of CAA correlated with cognitive impairment [51]. Candidate treatment approaches to prevent deposition of A $\beta$  in the arteries include approaches that reduce A $\beta$  production, increase its clearance, or block its toxicity. Immunotherapy approaches to clear amyloid from the vessels could have negative consequences, such as increasing the risk of amyloid-related microbleeds as noted by imaging abnormalities [42,52].

Vascular pathology caused by atherosclerosis, arteriosclerosis, or embolism is the only common comorbid pathology of AD that is potentially treatable with current pharmacotherapies and lifestyle modification. However, although there is some evidence that blood pressure control may prevent dementia, there is not yet strong evidence for treatment of other vascular risk factors [53]. Investigators planning future trials in AD may wish to take steps to ensure that vascular risk factors are balanced between randomized

groups and to implement prospective standardized protocols to ensure similar treatment of blood pressure and other vascular risk factors in all trial patients.

Treating the comorbid pathologic processes so common in AD and related neurodegenerative diseases may ultimately require combination therapy; however, evaluating the effectiveness of combination approaches presents challenges in both drug discovery and clinical trials. For example, there currently are no accepted animal models that reflect therapeutic pathways for testing treatments or understanding multiple pathologies. As a result, combinatorial approaches may have to be advanced to human studies by demonstrating target engagement in humans.

### 3.4. Biomarkers and imaging

Progress in understanding nonamyloid pathologic processes and developing treatments for them requires improved biomarkers and imaging tools. Recent advances in developing radioligands for tau PET imaging, for example, may enable the differential diagnosis of the various tauopathies [54]. Although tau is molecularly heterogeneous, low in abundance, and found intracellularly, several tau radioligands are currently in development [55–57].

Novel methods for imaging vascular cognitive impairment are also emerging, including higher resolution imaging to resolve small infarcts and white matter lesions; physiologic imaging approaches such as cerebral blood flow and permeability; and diffusion tensor imaging and resting-state functional MRI to assess structural and functional connectivity [58]. However, spatial resolution limits will continue to prevent direct measurement of microinfarcts, whose mean diameter may be as small as 200 microns [59]. Functional imaging of the dopaminergic system using <sup>123</sup>I-FP-CIT SPECT (DaTscan™, GE Healthcare, Chicago, IL, USA) may be used to identify dopaminergic deficiency in parkinsonian diseases, including LBD [60], but it remains to be determined how this technology can be used to identify concomitant LB pathology in AD patients.

Several studies have shown that the CSF biomarkers A $\beta$ <sub>1–42</sub>, tau, and phosphorylated tau may detect AD neuropathology and be useful in clinical practice for diagnosis and prognosis even in early disease stages [61–65]. These CSF biomarkers may also aid in differential diagnosis of AD versus other dementias and other neurologic diseases [66]. However, very few studies have examined CSF biomarkers in AD patients with well-defined copathologies, especially with neuropathologic confirmation. In a study of 142 autopsy-confirmed dementia patients (with 38 patients with AD with LBD or FTLN copathology), CSF A $\beta$ <sub>1–42</sub>, tau, and phosphorylated tau had robust sensitivity and specificity in AD with coincidence of other pathologies but did not differentiate between pure AD and AD with copathologies [67]. Likewise, in autopsy-characterized subjects from Alzheimer's Disease Neuroimaging Initiative, CSF A $\beta$ <sub>1–42</sub>, tau, and phosphorylated tau did not differ between pure

AD and AD with coincident TDP-43 or LB pathology [63]. Novel CSF biomarkers are needed for in vivo identification of copathologies in AD patients. One candidate is CSF neurofilament light protein (NFL), which may indicate damage of large-diameter myelinated axons and white matter lesions [68]. Specifically, high CSF NFL levels are seen in FTD [69], especially in tau-negative FTLN [70], and in patients with small-vessel disease in the brain, including AD dementia with vascular copathology [71]. CSF NFL may also provide prognostic information because it correlates with higher mortality in patients with and without AD [71]. Another candidate biomarker is CSF  $\alpha$ -synuclein, which is reduced in the presence of LB pathology [72,73]. Several studies have found reduced levels of CSF  $\alpha$ -synuclein in synucleinopathies (including LBD, PD, and multiple system atrophy) compared with AD [73–76]. Hypothetically, this could inform on LB copathology in AD because LBD-like symptoms are correlated with lower CSF  $\alpha$ -synuclein levels in AD dementia patients [77]. However, two neuropathologic studies did not find any difference in CSF  $\alpha$ -synuclein levels between AD patients with and without LB copathology [63,75]. The use of CSF  $\alpha$ -synuclein is complicated by several confounding factors, including increased CSF  $\alpha$ -synuclein as response to neuronal injury [78,79] and susceptibility to analytical factors, especially blood contamination of CSF [80]. Other  $\alpha$ -synuclein-related biomarkers, including  $\alpha$ -synuclein oligomers, may be more specific to the presence of LB pathology [81]. Recently, CSF biomarkers of synaptic function have been developed and tested in AD [82,83], but studies on differential diagnosis and copathologies are lacking. Likewise, CSF markers of inflammation [84] and microglial function [85–87] are altered in AD and may be explored further for differential diagnosis and tested in autopsy-confirmed subjects.

### 3.5. Implications of multiple/variable pathologies on clinical trials

Multiple pathologic processes have substantial implications on the design of clinical trials. Because the various pathologic processes have additive effects on cognition, when enrolling a heterogeneous population, greater numbers of subjects will be needed to achieve the power necessary to see an effect from an agent targeting just one pathology (e.g., amyloid) that is related to decline. Study cohorts in which diagnostic heterogeneity is not well defined could also lead to erroneous conclusions because decline may be different among the various subgroups. Although randomization should equally allocate both measured and unmeasured accompanying pathologies between treatment arms and thus prevent confounding, the potential for off-target drug effects on accompanying pathologic processes could lead to erroneous conclusions. The alternative—screening out individuals with non-AD pathologies including cerebrovascular disease—comes with its own set of problems,

including extremely high numbers of screen failures, the potential to eliminate decliners, and the resulting lack of generalizability because so many people with these other pathologic findings also have AD. Another alternative may be clinical trials focused on people with sporadic early-onset AD, that is, onset before 65 years of age, which may reduce the likelihood of copathologies. The current trend is to intervene early in the disease process when amyloid and/or tangles may not have reached some theoretical critical threshold. However, the slope of decline is less steep, requiring even more power to see an effect. In fact, treatment in secondary prevention or primary intervention typically translates into longer treatment periods and larger patient numbers to detect the smaller absolute change on outcome measures.

Postmortem pathology studies have been invaluable in illuminating underlying disease processes, and there was a suggestion that signed intent of willingness of participants to enroll in a brain donation program should be required for participation in a clinical trial; however, this requirement could complicate recruitment and retention efforts, and it would take many years to accrue sufficient postmortem examinations, particularly in trials enrolling MCI patients. It is worth noting that this has not been an impediment to development of neuroimaging markers [88].

In the future, disease-modifying trials will need to be large, and international trials will be needed, increasing the variability across centers. Identifying people who are likely to have progressive disease will thus become even more important for early disease treatments. Although the expertise for identifying such subjects has largely been located in academic medical centers, these centers have not demonstrated the ability to enroll the large numbers of subjects needed. Better education of general neurologists and general practitioners could help rectify this problem by encouraging them to refer potential subjects for clinical trials. There are also systems-based barriers that hinder enrollment in clinical trials because detailed interviews are needed to evaluate and accurately diagnose patients. On the other hand, clinicians in current medical practice are often incentivized to see more patients in less time.

Although investigators debate the pros and cons of testing drugs in homogenous versus heterogeneous populations, they agree on the need for better imaging and fluid biomarkers for vascular disease, tau,  $\alpha$ -synuclein, and TDP-43. In addition, even if the trial targets one pathologic process, measuring markers of other pathologies will undoubtedly lead to improved understanding of the results of interventions and prepare the field for future treatments that target multiple pathologies simultaneously.

#### 4. Conclusions

The criteria used to classify neurodegenerative diseases are based on the assumption of unitary clinicopathologic syndromes—namely that the underlying neuropathology

can be accurately predicted based on the clinical phenotype and that, as Occam's razor would postulate, a dementia syndrome in an individual patient is due to a single neuropathologic process. However, postmortem studies have revealed the real picture to be much more complex. The relationship between clinical syndrome and underlying neuropathology is probabilistic rather than deterministic: the same phenotype can be caused by different pathologies, and the same pathology can produce different phenotypes. Furthermore, a dementia syndrome is uncommonly related to a single process: Multiple pathologies seem to be the rule rather than the exception, especially in older individuals.

Multiple pathologic findings also complicate clinical trials. Although extremely common, they are mostly undetectable with current clinical, imaging, or fluid biomarker tools. New tools are needed to evaluate proteins other than amyloid and tau and to distinguish clinically relevant from "silent" cerebrovascular disease. If these tools were available and used to exclude subjects with multiple pathologic findings, the remaining cohort would likely not be representative of larger population of elderly individuals with dementia.

Roundtable participants debated the question of whether clinical phenotyping has limited promise in therapy development. They concluded that clinical phenotyping remains important and may itself be considered a biomarker, but biochemical and imaging biomarkers that can detect multiple pathologies are also needed to enable the development of better treatments. The challenge to the field is to define different etiologies according to the most definitive available biomarkers and then to assess how those biomarkers are affected by disease-modifying therapies.

In terms of therapy development, the high prevalence of multiple pathologic findings among individuals with clinical diagnosis of AD suggests that treatments for each pathologic entity may need to be developed, as well as treatments that target upstream or downstream events such as inflammation, network dysfunction, neurochemical changes, synaptic loss, and neuronal death. These treatments may then need to be used in combination to effectively treat and eventually cure AD and other dementing illnesses.

#### Acknowledgments

None of the coauthors report any funding conflicts related to this paper. Eric Smith reports grant funding from the Canadian Institutes of Health Research, Canadian Partnership Against Cancer, and Alberta Innovates-Health Solutions. Maria C. Carrillo and James A. Hendrix are full-time employees of the Alzheimer's Association. Mark Forman is a full-time employee of Merck. Susan DeSanti is a full-time employee of Piramal Pharma, Inc. David S. Miller is a full-time employee of Bracket Global. Nicholas Kozauer was a full-time employee of Quintiles in October 2014 at the time of the Research Roundtable meeting. Christopher Randolph is an employee of MedAvante and Loyola University Medical Center. John R. Sims is a full-time employee of

Eli Lilly & Co. Maria Isaac is a full-time employee of the European Medicines Agency. Lisa J. Bain is an independent science writer under contract from the Alzheimer's Association for writing this publication.

The authors thank Meredith McNeil for logistical planning of the roundtable as well as the contributing speakers: Adriano Aguzzi, MD, PhD, Dennis Dickson, MD, Matthew Frosch, MD, PhD, Steve Greenberg, MD, PhD, William Jagust, MD, Keith Josephs, MD, Brad Miller, MD, PhD, Leonard Petrucelli, PhD, Julie Schneider, MD, Andrew Siderowf, MD, Harry Vinters, MD, and Wagner Zago, PhD.

## RESEARCH IN CONTEXT

1. **Systematic review:** This review summarizes the presentations made at the October 2014 Research Roundtable meeting. Each presenter reviewed the literature of recent work of their specific topic area within the overall area of how overlapping pathologies affect the diagnosis and treatment of clinical Alzheimer's disease (AD) and other dementia phenotypes.
2. **Interpretation:** This article posits that dementia syndrome is most commonly related to multiple pathologies especially in older individuals, rather than a single process.
3. **Future directions:** A better understanding of the complexity of clinical phenotypes is needed. Biochemical and imaging biomarkers that can detect multiple pathologies may help enable the development of better treatments.

## References

- [1] Snowdon DA. Aging and Alzheimer's disease: lessons from the Nun Study. *Gerontologist* 1997;37:150-6.
- [2] Sperling RA, Aisen PS, Beckett LA, Bennett DA, Craft S, Fagan AM, et al. Toward defining the preclinical stages of Alzheimer's disease: Recommendations from the National Institute on Aging and the Alzheimer's Association workgroup. *Alzheimers Dement* 2011; 7:280-92.
- [3] Albert MS, Dekosky ST, Dickson D, Dubois B, Feldman HH, Fox NC, et al. The diagnosis of mild cognitive impairment due to Alzheimer's disease: Recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. *Alzheimers Dement* 2011;7:270-9.
- [4] McKhann GM, Knopman DS, Chertkow H, Hyman BT, Jack CR Jr, Kawas CH, et al. The diagnosis of dementia due to Alzheimer's disease: Recommendations from the National Institute on Aging and the Alzheimer's Association workgroup. *Alzheimers Dement* 2011; 7:263-9.
- [5] Hyman BT, Trojanowski JQ. Consensus recommendations for the postmortem diagnosis of Alzheimer disease from the National Institute on Aging and the Reagan Institute Working Group on diagnostic criteria for the neuropathological assessment of Alzheimer disease. *J Neuropathol Exp Neurol* 1997;56:1095-7.
- [6] Hyman BT, Phelps CH, Beach TG, Bigio EH, Cairns NJ, Carrillo MC, et al. National Institute on Aging-Alzheimer's Association guidelines for the neuropathologic assessment of Alzheimer's disease. *Alzheimers Dement* 2012;8:1-13.
- [7] Thal DR, Rub U, Orantes M, Braak H. Phases of A beta-deposition in the human brain and its relevance for the development of AD. *Neurology* 2002;58:1791-800.
- [8] Braak H, Alafuzoff I, Arzberger T, Kretschmar H, Del Tredici K. Staging of Alzheimer disease-associated neurofibrillary pathology using paraffin sections and immunocytochemistry. *Acta Neuropathol* 2006;112:389-404.
- [9] Braak H, Braak E. Neuropathological staging of Alzheimer-related changes. *Acta Neuropathol* 1991;82:239-59.
- [10] Mirra SS, Heyman A, McKeel D, Sumi SM, Crain BJ, Brownlee LM, et al. The Consortium to Establish a Registry for Alzheimer's Disease (CERAD). Part II. Standardization of the neuropathologic assessment of Alzheimer's disease. *Neurology* 1991;41:479-86.
- [11] Nag S, Yu L, Capuano AW, Wilson RS, Leurgans SE, Bennett DA, et al. Hippocampal sclerosis and TDP-43 pathology in aging and Alzheimer disease. *Ann Neurol* 2015;77:942-52.
- [12] Esiri MM, Wilcock GK, Morris JH. Neuropathological assessment of the lesions of significance in vascular dementia. *J Neurol Neurosurg Psychiatry* 1997;63:749-53.
- [13] Haroutunian V, Serby M, Purohit DP, Perl DP, Marin D, Lantz M, et al. Contribution of Lewy body inclusions to dementia in patients with and without Alzheimer disease neuropathological conditions. *Arch Neurol* 2000;57:1145-50.
- [14] White L. Brain lesions at autopsy in older Japanese-American men as related to cognitive impairment and dementia in the final years of life: a summary report from the Honolulu-Asia aging study. *J Alzheimers Dis* 2009;18:713-25.
- [15] Ferrer I, Santpere G, van Leeuwen FW. Argyrophilic grain disease. *Brain* 2008;131:1416-32.
- [16] Schneider JA, Arvanitakis Z, Leurgans SE, Bennett DA. The neuropathology of probable Alzheimer disease and mild cognitive impairment. *Ann Neurol* 2009;66:200-8.
- [17] Amador-Ortiz C, Lin WL, Ahmed Z, Personett D, Davies P, Duara R, et al. TDP-43 immunoreactivity in hippocampal sclerosis and Alzheimer's disease. *Ann Neurol* 2007;61:435-45.
- [18] Josephs KA, Murray ME, Whitwell JL, Parisi JE, Petrucelli L, Jack CR, et al. Staging TDP-43 pathology in Alzheimer's disease. *Acta Neuropathol* 2014;127:441-50.
- [19] Josephs KA, Whitwell JL, Knopman DS, Hu WT, Stroh DA, Baker M, et al. Abnormal TDP-43 immunoreactivity in AD modifies clinicopathologic and radiologic phenotype. *Neurology* 2008;70:1850-7.
- [20] Wilson RS, Yu L, Trojanowski JQ, Chen EY, Boyle PA, Bennett DA, et al. TDP-43 pathology, cognitive decline, and dementia in old age. *JAMA Neurol* 2013;70:1418-24.
- [21] Dickson DW, Baker M, Rademakers R. Common variant in GRN is a genetic risk factor for hippocampal sclerosis in the elderly. *Neurodegener Dis* 2010;7:170-4.
- [22] Dickson DW, Davies P, Bevona C, Van Hoeven KH, Factor SM, Grober E, et al. Hippocampal sclerosis: a common pathological feature of dementia in very old (> or = 80 years of age) humans. *Acta Neuropathol* 1994;88:212-21.
- [23] Uchikado H, Lin WL, DeLucia MW, Dickson DW. Alzheimer disease with amygdala Lewy bodies: a distinct form of alpha-synucleinopathy. *J Neuropathol Exp Neurol* 2006;65:685-97.
- [24] Lewis J, Dickson DW, Lin WL, Chisholm L, Corral A, Jones G, et al. Enhanced neurofibrillary degeneration in transgenic mice expressing mutant tau and APP. *Science* 2001;293:1487-91.
- [25] Choi SH, Kim YH, Hebsich M, Sliwinski C, Lee S, D'Avanzo C, et al. A three-dimensional human neural cell culture model of Alzheimer's disease. *Nature* 2014;515:274-8.



- [26] Jellinger KA. Interaction between pathogenic proteins in neurodegenerative disorders. *J Cell Mol Med* 2012;16:1166–83.
- [27] Murray ME, Graff-Radford NR, Ross OA, Petersen RC, Duara R, Dickson DW. Neuropathologically defined subtypes of Alzheimer's disease with distinct clinical characteristics: a retrospective study. *Lancet Neurol* 2011;10:785–96.
- [28] Bennett DA, Wilson RS, Arvanitakis Z, Boyle PA, de Toledo-Morrell L, Schneider JA. Selected findings from the Religious Orders Study and Rush Memory and Aging Project. *J Alzheimers Dis* 2013;33:S397–403.
- [29] Beach TG, Monsell SE, Phillips LE, Kukull W. Accuracy of the clinical diagnosis of Alzheimer disease at National Institute on Aging Alzheimer Disease Centers, 2005–2010. *J Neuropathol Exp Neurol* 2012;71:266–73.
- [30] Josephs KA, Hodges JR, Snowden JS, Mackenzie IR, Neumann M, Mann DM, et al. Neuropathological background of phenotypical variability in frontotemporal dementia. *Acta Neuropathol* 2011;122:137–53.
- [31] Naasan G, Rabinovici GD, Ghosh P, Eloffson JD, Miller BL, Coppola G, et al. Amyloid in dementia associated with familial FTL D: not an innocent bystander. *Neurocase* 2016;22:76–83.
- [32] Connor DJ, Salmon DP, Sandy TJ, Galasko D, Hansen LA, Thal LJ. Cognitive profiles of autopsy-confirmed Lewy body variant vs pure Alzheimer disease. *Arch Neurol* 1998;55:994–1000.
- [33] Johnson DK, Morris JC, Galvin JE. Verbal and visuospatial deficits in dementia with Lewy bodies. *Neurology* 2005;65:1232–8.
- [34] Yoshizawa H, Vonsattel JP, Honig LS. Early neuropsychological discriminants for Lewy body disease: an autopsy series. *J Neurol Neurosurg Psychiatry* 2013;84:1326–30.
- [35] Hu WT, Josephs KA, Knopman DS, Boeve BF, Dickson DW, Petersen RC, et al. Temporal lobar predominance of TDP-43 neuronal cytoplasmic inclusions in Alzheimer disease. *Acta Neuropathol* 2008;116:215–20.
- [36] Josephs KA, Whitwell JL, Weigand SD, Murray ME, Tosakulwong N, Liesinger AM, et al. TDP-43 is a key player in the clinical features associated with Alzheimer's disease. *Acta Neuropathol* 2014;127:811–24.
- [37] Moroney JT, Bagiella E, Desmond DW, Hachinski VC, Molsa PK, Gustafson L, et al. Meta-analysis of the Hachinski Ischemic Score in pathologically verified dementias. *Neurology* 1997;49:1096–105.
- [38] Raman MR, Preboske GM, Przybelski SA, Gunter JL, Senjem ML, Vemuri P, et al. Antemortem MRI findings associated with microinfarcts at autopsy. *Neurology* 2014;82:1951–8.
- [39] Arvanitakis Z, Leurgans SE, Barnes LL, Bennett DA, Schneider JA. Microinfarct pathology, dementia, and cognitive systems. *Stroke* 2011;42:722–7.
- [40] Boyle PA, Yu L, Nag S, Leurgans S, Wilson RS, Bennett DA, et al. Cerebral amyloid angiopathy and cognitive outcomes in community-based older persons. *Neurology* 2015;85:1930–6.
- [41] Schneider JA, Wilson RS, Bienias JL, Evans DA, Bennett DA. Cerebral infarctions and the likelihood of dementia from Alzheimer disease pathology. *Neurology* 2004;62:1148–55.
- [42] Sperling RA, Jack CR Jr, Black SE, Frosch MP, Greenberg SM, Hyman BT, et al. Amyloid-related imaging abnormalities in amyloid-modifying therapeutic trials: recommendations from the Alzheimer's Association Research Roundtable Workgroup. *Alzheimers Dement* 2011;7:367–85.
- [43] Guo JL, Lee VM. Cell-to-cell transmission of pathogenic proteins in neurodegenerative diseases. *Nat Med* 2014;20:130–8.
- [44] Aguzzi A, Gitler AD. A template for new drugs against Alzheimer's disease. *Cell* 2013;154:1182–4.
- [45] Aguzzi A. Neurodegeneration: Alzheimer's disease under strain. *Nature* 2014;512:32–4.
- [46] Freund EC, Maynard N, Clancy EK, Roy S, Bousset L, Sourigues Y, et al. Neuron-to-neuron transmission of alpha-synuclein fibrils through axonal transport. *Ann Neurol* 2012;72:517–24.
- [47] Games D, Valera E, Spencer B, Rockenstein E, Mante M, Adame A, et al. Reducing C-terminal-truncated alpha-synuclein by immunotherapy attenuates neurodegeneration and propagation in Parkinson's disease-like models. *J Neurosci* 2014;34:9441–54.
- [48] Mandler M, Valera E, Rockenstein E, Weninger H, Patrick C, Adame A, et al. Next-generation active immunization approach for synucleinopathies: implications for Parkinson's disease clinical trials. *Acta Neuropathol* 2014;127:861–79.
- [49] Yanamandra K, Kfoury N, Jiang H, Mahan TE, Ma S, Maloney SE, et al. Anti-tau antibodies that block tau aggregate seeding in vitro markedly decrease pathology and improve cognition in vivo. *Neuron* 2013;80:402–14.
- [50] Gendron TF, Petrucelli L. Rodent models of TDP-43 proteinopathy: investigating the mechanisms of TDP-43-mediated neurodegeneration. *J Mol Neurosci* 2011;45:486–99.
- [51] Arvanitakis Z, Leurgans SE, Wang Z, Wilson RS, Bennett DA, Schneider JA. Cerebral amyloid angiopathy pathology and cognitive domains in older persons. *Ann Neurol* 2011;69:320–7.
- [52] Sperling RA, Salloway S, Arrighi HM, Morris K, Lu Y, Liu E, et al. Revised estimates of incidence and risk factors for amyloid related imaging abnormalities (ARIA) in the phase 2 studies of bapineuzumab for mild-to-moderate Alzheimer's disease. *Alzheimers Dement* 2011;7:e64.
- [53] Gorelick PB, Scuteri A, Black SE, Decarli C, Greenberg SM, Iadecola C, et al. Vascular contributions to cognitive impairment and dementia: a statement for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke* 2011;42:2672–713.
- [54] Fodero-Tavoletti MT, Furumoto S, Taylor L, McLean CA, Mulligan RS, Birchall I, et al. Assessing THK523 selectivity for tau deposits in Alzheimer's disease and non-Alzheimer's disease tauopathies. *Alzheimers Res Ther* 2014;6:11.
- [55] Chien DT, Bahri S, Szardenings AK, Walsh JC, Mu F, Su MY, et al. Early clinical PET imaging results with the novel PHF-tau radioligand [F-18]-T807. *J Alzheimers Dis* 2013;34:457–68.
- [56] Hashimoto H, Kawamura K, Igarashi N, Takei M, Fujishiro T, Aihara Y, et al. Radiosynthesis, photoisomerization, biodistribution, and metabolite analysis of <sup>11</sup>C-PBB3 as a clinically useful PET probe for imaging of tau pathology. *J Nucl Med* 2014;55:1532–8.
- [57] Okamura N, Furumoto S, Fodero-Tavoletti MT, Mulligan RS, Harada R, Yates P, et al. Non-invasive assessment of Alzheimer's disease neurofibrillary pathology using <sup>18</sup>F-THK5105 PET. *Brain* 2014;137:1762–71.
- [58] Wardlaw JM, Smith EE, Biessels GJ, Cordonnier C, Fazekas F, Frayne R, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. *Lancet Neurol* 2013;12:822–38.
- [59] Smith EE, Schneider JA, Wardlaw JM, Greenberg SM. Cerebral microinfarcts: the invisible lesions. *Lancet Neurol* 2012;11:272–82.
- [60] Brigo F, Turri G, Tinazzi M. <sup>123</sup>I-FP-CIT SPECT in the differential diagnosis between dementia with Lewy bodies and other dementias. *J Neurol Sci* 2015;359:161–71.
- [61] Seppala TT, Nerg O, Koivisto AM, Rummukainen J, Puli L, Zetterberg H, et al. CSF biomarkers for Alzheimer disease correlate with cortical brain biopsy findings. *Neurology* 2012;78:1568–75.
- [62] Kester MI, Goos JD, Teunissen CE, Benedictus MR, Bouwman FH, Wattjes MP, et al. Associations between cerebral small-vessel disease and Alzheimer disease pathology as measured by cerebrospinal fluid biomarkers. *JAMA Neurol* 2014;71:855–62.
- [63] Toledo JB, Cairns NJ, Da X, Chen K, Carter D, Fleisher A, et al. Clinical and multimodal biomarker correlates of ADNI neuropathological findings. *Acta Neuropathol Commun* 2013;1:65.
- [64] Toledo JB, Weiner MW, Wolk DA, Da X, Chen K, Arnold SE, et al. Neuronal injury biomarkers and prognosis in ADNI subjects with normal cognition. *Acta Neuropathol Commun* 2014;2:26.

- [65] Mattsson N, Zetterberg H, Hansson O, Andreassen N, Parnetti L, Jonsson M, et al. CSF biomarkers and incipient Alzheimer disease in patients with mild cognitive impairment. *JAMA* 2009;302:385–93.
- [66] Ewers M, Mattsson N, Minthon L, Molinuevo JL, Antonell A, Popp J, et al. CSF biomarkers for the differential diagnosis of Alzheimer's disease: A large-scale international multicenter study. *Alzheimers Dement* 2015;11:1306–15.
- [67] Toledo JB, Brettschneider J, Grossman M, Arnold SE, Hu WT, Xie SX, et al. CSF biomarkers cutoffs: the importance of coincident neuropathological diseases. *Acta Neuropathol* 2012;124:23–35.
- [68] Jonsson M, Zetterberg H, van Straaten E, Lind K, Syversen S, Edman A, et al. Cerebrospinal fluid biomarkers of white matter lesions - cross-sectional results from the LADIS study. *Eur J Neurol* 2010;17:377–82.
- [69] Scherling CS, Hall T, Berisha F, Klepac K, Karydas A, Coppola G, et al. Cerebrospinal fluid neurofilament concentration reflects disease severity in frontotemporal degeneration. *Ann Neurol* 2014;75:116–26.
- [70] Landqvist Waldo M, Frizell Santillo A, Passant U, Zetterberg H, Rosengren L, Nilsson C, et al. Cerebrospinal fluid neurofilament light chain protein levels in subtypes of frontotemporal dementia. *BMC Neurol* 2013;13:54.
- [71] Skillback T, Farahmand B, Bartlett JW, Rosen C, Mattsson N, Nagga K, et al. CSF neurofilament light differs in neurodegenerative diseases and predicts severity and survival. *Neurology* 2014;83:1945–53.
- [72] Mollenhauer B, El-Agnaf OM, Marcus K, Trenkwalder C, Schlossmacher MG. Quantification of alpha-synuclein in cerebrospinal fluid as a biomarker candidate: review of the literature and considerations for future studies. *Biomark Med* 2010;4:683–99.
- [73] Hall S, Ohrfelt A, Constantinescu R, Andreasson U, Surova Y, Bostrom F, et al. Accuracy of a panel of 5 cerebrospinal fluid biomarkers in the differential diagnosis of patients with dementia and/or parkinsonian disorders. *Arch Neurol* 2012;69:1445–52.
- [74] Mollenhauer B, Locascio JJ, Schulz-Schaeffer W, Sixel-Doring F, Trenkwalder C, Schlossmacher MG. alpha-Synuclein and tau concentrations in cerebrospinal fluid of patients presenting with parkinsonism: a cohort study. *Lancet Neurol* 2011;10:230–40.
- [75] Slaets S, Vanmechelen E, Le Bastard N, Decraemer H, Vandijck M, Martin JJ, et al. Increased CSF alpha-synuclein levels in Alzheimer's disease: correlation with tau levels. *Alzheimers Dement* 2014;10:S290–8.
- [76] Wang Y, Shi M, Chung KA, Zabetian CP, Leverenz JB, Berg D, et al. Phosphorylated alpha-synuclein in Parkinson's disease. *Sci Transl Med* 2012;4:121ra20.
- [77] Mackin RS, Insel P, Zhang J, Mohlenhoff B, Galasko D, Weiner M, et al. Cerebrospinal fluid alpha-synuclein and Lewy body-like symptoms in normal controls, mild cognitive impairment, and Alzheimer's disease. *J Alzheimers Dis* 2015;43:1007–16.
- [78] Toledo JB, Korff A, Shaw LM, Trojanowski JQ, Zhang J. CSF alpha-synuclein improves diagnostic and prognostic performance of CSF tau and Abeta in Alzheimer's disease. *Acta Neuropathol* 2013;126:683–97.
- [79] Kasai T, Tokuda T, Ishii R, Ishigami N, Tsuboi Y, Nakagawa M, et al. Increased alpha-synuclein levels in the cerebrospinal fluid of patients with Creutzfeldt-Jakob disease. *J Neurol* 2014;261:1203–9.
- [80] Korff A, Liu C, Gingham C, Shi M, Zhang J. Alzheimer's Disease Neuroimaging I. alpha-Synuclein in cerebrospinal fluid of Alzheimer's disease and mild cognitive impairment. *J Alzheimers Dis* 2013;36:679–88.
- [81] Hansson O, Hall S, Ohrfelt A, Zetterberg H, Blennow K, Minthon L, et al. Levels of cerebrospinal fluid alpha-synuclein oligomers are increased in Parkinson's disease with dementia and dementia with Lewy bodies compared to Alzheimer's disease. *Alzheimers Res Ther* 2014;6:25.
- [82] Kester MI, Teunissen CE, Crimmins DL, Herries EM, Ladenson JH, Scheltens P, et al. Neurogranin as a Cerebrospinal Fluid Biomarker for Synaptic Loss in Symptomatic Alzheimer Disease. *JAMA Neurol* 2015;72:1275–80.
- [83] Portelius E, Zetterberg H, Skillback T, Tornqvist U, Andreasson U, Trojanowski JQ, et al. Cerebrospinal fluid neurogranin: relation to cognition and neurodegeneration in Alzheimer's disease. *Brain* 2015;138:3373–85.
- [84] Wiltfang J, Lewczuk P, Riederer P, Grunblatt E, Hock C, Scheltens P, et al. Consensus paper of the WFSBP Task Force on Biological Markers of Dementia: the role of CSF and blood analysis in the early and differential diagnosis of dementia. *World J Biol Psychiatry* 2005;6:69–84.
- [85] Kleinberger G, Yamanishi Y, Suarez-Calvet M, Czirr E, Lohmann E, Cuyvers E, et al. TREM2 mutations implicated in neurodegeneration impair cell surface transport and phagocytosis. *Sci Transl Med* 2014;6:243ra86.
- [86] Mattsson N, Tabatabaei S, Johansson P, Hansson O, Andreasson U, Mansson JE, et al. Cerebrospinal fluid microglial markers in Alzheimer's disease: elevated chitotriosidase activity but lack of diagnostic utility. *Neuromolecular Med* 2011;13:151–9.
- [87] Rosen C, Andersson CH, Andreasson U, Molinuevo JL, Bjerke M, Rami L, et al. Increased Levels of Chitotriosidase and YKL-40 in Cerebrospinal Fluid from Patients with Alzheimer's Disease. *Demen Geriatr Cogn Dis Extra* 2014;4:297–304.
- [88] Clark CM, Schneider JA, Bedell BJ, Beach TG, Bilker WB, Mintun MA, et al. Use of florbetapir-PET for imaging beta-amyloid pathology. *JAMA* 2011;305:275–83.