Plasma tau and neurofilament light in frontotemporal lobar degeneration and Alzheimer's disease

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Abstract

Objective: To test the hypothesis that plasma total tau (t-tau) and neurofilament light chain (NfL) concentrations may have a differential role in the study of frontotemporal lobar degeneration syndromes (FTLD-S) and clinically-diagnosed Alzheimer's disease (AD-S), we determined their diagnostic and prognostic value in FTLD-S and AD-S and their sensitivity to pathologic diagnoses.

Methods: We measured plasma t-tau and NfL with the Simoa platform in 265 participants: 167 FTLD-S, 43 AD-S, and 55 healthy controls (HC), including 82 pathology-proven cases (50 FTLD-Tau, 18 FTLD-TDP, 2 FTLD-FUS, and 12 AD) and 98 participants with amyloid PET. We compared cross-sectional and longitudinal biomarker concentrations between groups, their correlation with clinical measures of disease severity, progression and survival and cortical thickness.

Results: Plasma NfL, but not plasma t-tau discriminated FTLD-S from HC and AD-S from HC. Both plasma NfL and t-tau were poor discriminators between FLTD-S and AD-S. In pathology confirmed cases, plasma NfL was higher in FTLD than AD and in FTLD-TDP compared to FTLD-Tau, after accounting for age and disease severity. Plasma NfL, but not plasma t-tau, predicted clinical decline and survival and correlated with regional cortical thickness in both FTLD-S and AD-S. The combination of plasma NfL with plasma t-tau did not outperform plasma NfL alone.

Conclusions: Plasma NfL is superior to plasma t-tau for the diagnosis and prediction of clinical progression of FTLD-S and AD-S.

Classification of evidence: This study provides Class III evidence that plasma NfL has superior diagnostic and prognostic performance than plasma t-tau in FTLD and AD.

Introduction

Frontotemporal lobar degeneration (FTLD) and Alzheimer's disease (AD) are two heterogeneous neuropathological constructs characterized by the neurodegeneration of distinct but partially overlapping cerebral regions.[1–5] Both entities present with a wide range of phenotypes, and have widely variable rates of clinical progression at the single subject level.[4] This creates important diagnostic and prognostic barriers that impact participant selection for clinical trials testing disease-modifying drugs and other therapy development efforts.

Fluid biomarkers represent potentially powerful diagnostic and prognostic clinical tools in neurodegenerative dementias.[6] To date, several fluid biomarkers have been proposed to reflect neurodegeneration-driven cerebral changes. The microtubule associated protein tau modulates the dynamic stability of axonal microtubules and has been implicated in the pathophysiology of multiple neurodegenerative diseases, including AD and FTLD.[7] CSF concentrations of both total tau (herein, tau) and phosphorylated tau (p-tau) are specifically increased in pathology-confirmed cases with AD (either alone or as a comorbid pathology in FTLD cases) and are considered biomarkers of neurodegeneration and tau pathology in the recently proposed AD research framework.[1,6] In turn, neurofilament light (NfL) is an abundant intermediate filament cytoskeletal protein that is elevated in CSF and blood upon neuronal injury, irrespective of cause, and is associated with clinical progression and survival in FTLD-S and AD.[8–12] In multimodal biomarker studies, NfL in biofluids has been used for the stratification of patients with neurodegenerative dementias according to the "intensity" (*i.e.*, rate of clinical progression) of neurodegeneration.[13]

Previous studies have found that CSF levels of both NfL and tau may be useful for the diagnosis and prognosis in FTLD.[6,14–16] Tau and NfL, however, can now be detected in plasma with ultrasensitive technology,[17] but neither their diagnostic and prognostic value nor their relationship with AD pathophysiology has been previously compared. We hypothesized that plasma concentrations of tau and NfL will differentially reflect neurodegeneration in FTLD-S and AD and that their use, alone or in combination may be helpful to identify underlying pathology and inform about disease severity.

In this multimodal biomarker study, we compared the diagnostic and prognostic value of both plasma tau and NfL concentrations (alone or in combination) in a large sample of FTLD-S, AD and HC. We also compared the utility of these plasma biomarkers for the detection FTLD major subtypes and underlying AD pathophysiology, as well as their ability to track neurodegeneration, assessed by clinical measures of disease severity, progression, survival and cortical thickness.

Methods

Study participants and classification.

Participants were recruited at the University of California, San Francisco (UCSF) Memory and Aging Center from November 2011 to January 2015. A total of 267 research participants provided written informed consent and underwent neurological, neuropsychological, functional assessment with informant interview, and blood sampling. A subgroup of participants also underwent structural brain MRI (n=240) and CSF sample collection (n=181). Participants were diagnosed at a

multidisciplinary consensus conference and met criteria for behavioral variant frontotemporal dementia (bvFTD)[18], primary progressive aphasia (PPA)[19], progressive supranuclear palsy (PSP)[20], corticobasal syndrome (CBS)[21], amyotrophic lateral sclerosis with frontotemporal dementia (ALS-FTD)[22] or AD.[23] Participants in the control group (HC) were cognitively healthy participants enrolled through a healthy aging cohort.

Neurocognitive and disease-staging measures.

Participants underwent a comprehensive neuropsychological battery at the time of plasma sampling. Four major cognitive domains were covered as previously described:[24] memory (delayed recall of the Californian Verbal Learning Test and Benson Figure Test), executive functioning (Digit Span backwards, Trail Making Test part B, Stroop Color-Word card subtask and Letter Fluency), language (Category Fluency at both sites, Boston Naming Test) and visuo-spatial functioning [Number Location from the Visual Object Space and Perception battery and (modified) Rey Complex Figure copy test]. To obtain composite scores for each cognitive domain, we used the means and standard deviations of the healthy control group to convert raw cognitive scores into Z-scores. Subsequently, patient's Z-scores were averaged within each cognitive domain. Additionally, the Mini-Mental State Examination (MMSE)[25] was used as a general measure of global cognition and, the Clinical Dementia Rating sum-of-the-boxes (CDR-sb) score was used as a measure of disease severity.[26]

Plasma and CSF biomarkers.

Plasma and CSF collections were performed according to the Alzheimer's Disease Neuroimaging Initiative protocol.[9] Plasma NfL and plasma tau concentrations were

determined with commercially-available ultrasensitive Single molecule array technology using an HD-1 analyzer (Quanterix, Billerica, MA), by researchers blinded to clinical data, as previously described.[9] CSF concentrations of both total-tau (tau) and phosphorylated tau (p-tau$_{181}$) were measured with the INNO-BIA AlzBio3 platform (Fujirebio, Gent, Belgium). CSF NfL concentration was measured using the UmanDiagnostics (Umeå, Sweden) ELISA kit (NF-Light kit), as previously described.[27]

Amyloid PET.

98 participants had available brain amyloid PET data (71 PiB tracer and 27 florbetapir) at plasma sampling. We used visual impression for the dichotomization of amyloid PET results. Participants with a positive amyloid PET were included in the subgroup of participants with increased certainty of underlying AD pathophysiology for secondary analyses.

MRI acquisition and preprocessing.

A total of 240 participants (160 FTLD-S, 29 AD and 51 HC) underwent an MRI at the time of plasma sampling (mean time from plasma sampling to scan 1 month, with a maximum time between plasma sampling to MRI of 6 months). MRIs were acquired on a 3T Siemens Tim Trio system equipped with a 12-channel head coil. Fifteen MRIs were excluded from final neuroimaging analyses: eight because of low image quality (*i.e.*, significant movement artifact) or pre-processing errors and seven because they were performed in a different MRI scan. The remaining 225 MRIs (160 FTLD-S, 29 AD and 51 HC) were processed with CAT12 toolbox (http://www.neuro.uni-jena.de/cat/, version 1450) within SPM12 (http://www.fil.ion.ucl.ac.uk/spm/software/spm12/, version 7487, running in

MATLAB r2019b) to gather cortical thickness estimates, as previously described.[28] Briefly, the CAT12 toolbox uses tissue segmentation to estimate the white matter distance, and it then projects the local maxima (which is equal to cortical thickness) to other gray matter voxels by using a neighbor relationship described by the white matter distance. Previous studies have shown that projection-based thickness allows the handling of partial volume information, sulcal blurring, and sulcal asymmetries without explicit sulcus reconstruction.[28] Topological correction, spherical mapping, and spherical registration were performed to obtain vertex wise cortical thickness. Finally, surface maps were smoothed using a 15 mm-FWMH for group comparisons and correlations with plasma biomarkers.

Neuroimage analyses.

Cortical thickness comparisons between FTLD-S and AD groups and controls were performed in CAT12 with a 2-sample t test using age and sex as covariates. For group comparisons, a significant statistical threshold of $p < 0.05$, false discovery rate (FDR)-corrected, was considered using an extent threshold of the expected vertices per cluster. Correlation of regional cortical thickness maps with plasma tau and NfL concentrations was performed in both FTLD-S and AD groups using multiple regressions with individual plasma biomarker levels as the variable of interest, and age and sex as covariates. For correlation analyses, in order to detect moderate correlation coefficients, we show uncorrected $p < 0.001$ results.

Genetic analysis.

Genetic screening was conducted for mutations known to cause autosomal dominant FTLD or AD (*MAPT*, *C9orf72*, *GRN*, *TARDBP*, *FUS*, *PSEN1*, *PSEN2*, and *APP*) at the Coppola Lab at the University of California, Los Angeles.

Neuropathological assessment.

Neuropathological assessments performed at UCSF followed previously described procedures.[29] Participants were classified into FTLD major molecular classes (Tau, TDP-43, or FUS) and subtypes[30,31] or AD.[32,33] AD pathology was classified according to the National Institute on Aging – Reagan criteria for likelihood of AD pathology as low, intermediate or high.[34] For secondary analyses, we considered that participants with either a positive amyloid PET, or at least comorbid AD (as defined by at least intermediate likelihood of AD pathology) has an increased certainty of underlying AD (either as a primary or contributing neuropathological diagnosis).

Statistical analysis.

Data was explored for normality using the Shapiro-Wilk test. Between-group differences were determined with ANOVA or t-test for continuous variables, and the Chi-square for dichotomous or categorical data. in addition to main clinical group comparisons (namely, FTLD-S, AD and HC) we also performed secondary analyses comparing plasma biomarkers between FTLD subtypes in cases with pathological confirmation. Moreover, to explore the impact of AD pathophysiology on the studied plasma biomarker levels, we also compared participants with an increased certainty of underlying AD (as defined in the neuropathological assessment section). Fluid biomarker concentrations were log-transformed using the natural log to fulfill the normal distribution assumptions needed for ANOVA analyses. We studied the correlation between cognitive composites and plasma biomarkers with partial correlations adjusting by the effect of age and education, which were demographic variables that showed between-group differences. Statistical significance for all

tests was set at 5% (α= .05), and all statistical tests were two-sided. Linear mixed effects analyses controlling for age, sex, disease severity (as measured with CDR-sb) and baseline clinical syndrome (only in FTLD-S) were used to determine the ability of baseline plasma biomarkers to predict a change in disease severity as measured by the CDR-sb score over time. A compound symmetry covariance matrix was used in all models. To account for the effect of baseline values, a term for biomarker by time interaction was introduced in addition to the random intercepts. Survival was calculated from the date of blood draw until death. Patients alive at analysis were censored at that date. For survival analyses, we first evaluated the association of age at diagnosis, sex and disease severity at symptom onset with survival in FTLD-S and AD. In FTLD-S, we also controlled for the clinical phenotype at plasma sampling. We applied Cox regression analyses to estimate survival while taking into account age at diagnosis, sex, disease severity at symptom onset and main clinical phenotype (only in the FTLD-S group). We next introduced plasma biomarkers in the Cox regression models to test if plasma biomarkers were independent predictors of survival. All analyses were performed using SPSS 24 (Armonk, NY: IBM Corp.).

Standard protocol approvals, registrations, and patient consent.

The study was approved by the local ethics committee and was conducted following the Declaration of Helsinki. All participants gave their written informed consent to participate in the study.

Data availability.

The datasets analyzed during the current study are available from the corresponding authors on reasonable request.

Results

Sample composition and demographics.

From an initial sample of 304 participants with available plasma tau measurements, we excluded 29 participants with preclinical FTD (asymptomatic mutation carriers), 9 participants with a clinical or pathological diagnosis of Lewy body disease and 1 participant with a final diagnosis of primary psychiatric disease. The final sample included 265 participants: 167 FTLD-S (FTLD-S) [(43 bvFTD, 28 non-fluent variant PPA (nfvPPA), 18 semantic variant PPA (svPPA), 36 PSP, 32 CBS and 10 ALS-FTD], 43 AD and 55 healthy controls (HC). Age at plasma sampling, education, MMSE and CDR-sb were similar between participants in the FTLD-S and AD groups. The HC group, however, was younger than both disease groups (**Table 1**).

Relationship between plasma biomarkers, age and clinical measures.

There were no correlations between plasma tau and NfL concentrations within any clinical group. Plasma tau concentrations did not correlate with age at plasma sampling in any clinical group. In contrast, plasma NfL and age showed a weak correlation in the AD and HC groups (r=.51, 95% CI .23 to .70 and r=.63, 95% CI .43 to .80, respectively, all p<.001) but not in the FTLD-S group (r=.02, 95% CI -.15 to .20, p=796). In the whole sample, plasma NfL correlated with MMSE (r=-.26, 95% CI -.36 to -.16, p<.001), the executive cognitive composite (r=-.15, 95% CI -.29 to -.03, p=.037) and CDR-sb (r=.27, 95% CI .12 to .41, p<.001) scores. However, when we restricted the analyses to each clinical group, we only observed a significant

correlation between plasma NfL and CDR-sb in the AD group (r=.50, 95% CI .09 to .77, *p*=.003).

Relationship between plasma and CSF biomarkers.

We did not find any significant correlation between plasma tau and CSF tau. However, as shown in **Table e1**, plasma and CSF measures of NfL were strongly correlated in the whole sample (r=.82, p<.001) and in all clinical groups (r=.63, r=.64, and r=.66, in FTLD-S, AD and HC, respectively).

Differences in plasma tau and NfL concentrations by clinical syndrome.

We observed different concentrations of plasma NfL, but not tau between FTLD-S, AD and controls (**Figures 1A and 1B**). Plasma NfL concentrations in the FTLD-S group (50.2 ± 31.7 pg/mL) were higher than AD and HC (28.5 ± 11.5 pg/mL and 12.1 ± 4.7 pg/mL, *p*<.001 and *p*<.001, respectively). Plasma NfL concentrations were also increased in AD compared to HC (*p*<.001). As shown in **Figure 1C and 1D**, within the FTLD-S group, the FTD-ALS subgroup showed decreased levels of plasma tau (1.6 ± 0.9 pg/mL) and increased levels of plasma NfL (99.1 ± 46 pg/mL). All clinical subgroups included in the FTLD-S groups had higher plasma NfL concentrations than the AD group. Of note, participants in the FTLD-S group without a mutation had similar plasma NfL levels than *GRN* and MAPT carriers, but lower plasma NfL levels than *C9orf72* (*p*=.006). However, plasma levels of tau did not differ between mutation carriers and sporadic FTLD-S (**Supplementary Table e2)**.

Differences in plasma tau and NfL concentrations between pathological subtypes of FTLD.

Plasma tau concentrations did not differ between FTLD subtypes (**Figure 1E**). However, we observed higher plasma NfL concentrations, in the FTLD-TDP subgroup (85.6 ± 46.6 pg/mL) when compared to the FTLD-Tau subgroup (50.4 ±

26.9 pg/mL; *p*=.001). The effect size of this difference was small but remained significant after accounting for age, sex and disease severity at plasma sampling (*p*<.001; partial η^2=.20) and also after including participants with FTLD-related mutations without neuropathological confirmation (*p*=.001; partial η^2=.14). As shown in **Figure 1F**, both FTLD-Tau and FTLD-TDP groups had higher plasma NfL concentrations than participants in the AD group with pathological confirmation or positive amyloid PET (n=30; 29.0 ± 12.2 pg/mL). Of note, plasma tau or NfL concentrations did not differ between pathology-confirmed FTLD cases with and without comorbid AD (data not shown).

Differences in plasma tau concentrations between participants with and without increased certainty of underlying AD.

Since a significant proportion of FTLD-S were found to have some degree of comorbid AD (**Table 1**), we also investigated if plasma tau and NfL concentrations varied between participants with increased certainty of underlying AD (either positive amyloid PET and/or at least intermediate AD likelihood on autopsy, n=43) and participants without AD (n=103, negative amyloid PET and/or absent/low comorbid AD at autopsy). Importantly. there were no differences in plasma tau or NfL concentrations regarding on presence or absence of AD pathophysiology.

Diagnostic value of plasma tau and NfL.

Plasma tau had no diagnostic utility for the differentiation of FTLD-S, AD and HC. Nevertheless, plasma NfL showed an excellent performance in the differentiation of FTLD-S from HC (AUC=.97, 95% CI .95 to .99, *p*<.001) and of AD from controls (AUC=.94, 95% CI .89 to .98, *p*<.001), but a poor performance for the discrimination between FTLD-S and AD (AUC=.75, 95% CI .68 to .82) (**Figure 2**). Importantly, the

combination of plasma NfL and plasma tau in a ratio did not improve the diagnostic performance of plasma NfL alone.

Longitudinal changes in plasma tau and NfL.

We explored the longitudinal changes in tau and NfL plasma concentrations in the subgroup of participants with a second longitudinal sample available (n=103, mean time between samples = 1.2 ± 0.4 years). After controlling for age, sex, baseline CDR-sb and time between samples, we observed an increase of mean plasma NfL concentrations in FTLD-S, compared to baseline (10.3 pg/mL, 95% CI: 5.7 to 14.9, *p*<.001). There were no longitudinal increases of NfL in AD or of tau in either diagnostic group.

Relationship between baseline plasma biomarkers and clinical progression.

Figure 3 shows the association between baseline plasma tau and NfL concentrations and longitudinal CDR-sb score. Baseline plasma tau concentrations did not predict the rate of change in CDR-sb in any clinical group (**Figure 3, A-B**). In contrast, baseline NfL concentration by time interactions were observed with plasma NfL concentrations related to faster annual worsening in CDR-sb, for both FTLD-S (1.6 points per log NfL ng/mL increase per year, 95% CI 0.78 to 2.4, *p*<.001) and AD (3.4 points per log NfL ng/mL increase per year, 95% CI 1.2 to 5.5, *p*=.002)(**Figure 3, C-D**). Importantly, the combination of plasma NfL and tau levels did not improve the ability of plasma NfL alone to predict longitudinal CDR-sb score changes (data not shown).

Relationship between plasma biomarkers and cortical thickness.

When compared to the HC group, the FTLD-S group showed expected decreases in cortical thickness in dorsolateral prefrontal, superior frontal, inferior frontal,

temporal poles and medial and lateral temporal regions. The AD group also showed the expected pattern of atrophy in temporal and parietal regions (**Figure 4A and 4B**). Plasma tau concentrations showed no correlations with cortical thickness in neither the FTLD-S or AD groups (**Figure 4C and 4D**). In contrast, plasma NfL correlated with cortical thickness in frontal regions in FTLD-S and in the right lateral temporal lobe, right inferior parietal and left superior frontal in the AD group (**Figure 4E and 4F**).

Survival analyses.

As shown in **Table 2**, in FTLD-S group, only the clinical phenotype was independently associated with a shorter survival while in the AD group, only the CDR-sb score was independently associated with a shorter survival. When we introduced plasma biomarkers in the Cox regression models, only in FTLD-S, plasma NfL, but not plasma tau, predicted survival after accounting for age and CDR-sb at plasma sampling, sex, and clinical phenotype. For illustrative purposes, **Figure 5** shows survival curves in the FTLD-S group after a median split based on plasma NfL levels. FTLD-S participants with high NfL levels (> 42 ng/mL) showed an increased mortality compared to those with lower concentrations (Log-Rank=14.423, $p<.001$. Of note, neither plasma tau nor plasma NfL predicted survival in the AD group (**Table 2**).

Discussion

In this multimodal biomarker study, we compared for the first time the diagnostic and prognostic value of plasma tau and NfL in FTLD-S and AD participants with

deep clinical, neuropsychological, and neuroimaging phenotyping. The main findings of this study were that (i) plasma NfL was the only biomarker providing between-group clinical discrimination, predicting disease progression and survival, and correlating with neuroimage measures of neurodegeneration; and (ii) the combination of plasma NfL and plasma tau did not improve the performance of plasma NfL alone.

Plasma NfL was higher in both FTLD-S and AD than controls and it was higher in FTLD-S compared to AD. Within FTLD-S, the highest plasma NfL levels were observed in the FTD-ALS subgroup. Importantly, in pathology-confirmed cases, plasma NfL was higher in FTLD-TDP, compared to FTLD-tau independently of the inclusion of FTLD-related mutations. In both FTLD-S and AD, plasma NfL correlated with more severe clinical deterioration overtime, and in FTLD-S, it was associated with shorter survival. In addition, plasma NfL correlated with reduced frontal cortical thickness in FTLD-S and with reduced cortical thickness in parietotemporal regions in AD. In marked contrast, plasma tau showed none of these associations and showed no clinical value, except for being low in FTD-ALS, compared to other FTLD phenotypes, and an association more aggressive disease course over time in the bvFTD subgroup. In pathology-confirmed cases, most FTLD cases had at least some degree of AD co-pathology, but it did not influence the performance of plasma tau or NfL.

The striking contrast between the performances of plasma tau and NfL suggest that they may reflect different aspects of neurodegeneration and also highlights the importance of the accurate definition of pathophysiological categories in biomarker-based classification systems.[35] Indeed, accumulating evidence indicates that tau and NfL measurements may provide different information compared to other

neurodegeneration biomarkers, such as FDG PET, or structural neuroimaging biomarkers,[35] and their longitudinal trajectories may be differently affected by demographic variables or disease stage.[36]

Tau is a microtubule-stabilizing protein encoded by *MAPT* and has been implicated in the pathophysiology of AD and FTLD. Tau hyperphosphorylation leads to formation of paired helical filaments that aggregate in neurofibrillary tangles, a defining pathological hallmark of AD.[1] Elevated CSF levels of tau and p-tau are considered markers of neurodegeneration and tau pathology in AD and are used in the clinical setting to increase the diagnostic certainty of AD.[6,37] Moreover, high CSF tau levels have been related to clinical progression in AD[38] and in FTLD, high CSF tau is associated with more severe clinical progression and brain atrophy.[39] Only a single previous study investigated plasma tau levels in FTLD-S.[17] In this study, plasma tau was elevated in bvFTD, PPA and symptomatic *MAPT* mutation carriers, but the effect sizes where small, pathological data were not available and analyzes for prediction of disease progression with clinical scales, neuropsychological testing and survival were not conducted. In agreement with our results, no associations where found between plasma tau and baseline measures of disease severity or brain volume, and AD pathophysiology (as measured by the CSF tau/Aβ_{1-42} ratio), did not influence plasma tau clinical performance.[17] Also in agreement with the present results are those from two large AD cohorts, in which plasma tau was associated with faster clinical decline.[40] Plasma tau was also previously found to show weak elevations compared to controls and correlate with more severe longitudinal hypometabolism in AD.[40] Studies of plasma tau in AD and FTLD, including the present one, have found no relationship between plasma and CSF tau. This contrasts with relatively strong associations between plasma and CSF tau and

strong prediction of survival by plasma tau in Creutzfeldt Jakob disease, in which the range of tau concentrations in plasma and CSF is higher than in AD or FTLD[41]. Together, the data support a potential prognostic role of plasma tau in AD, but not in FTLD. Diagnostically, plasma tau has a poor performance and will likely be of little value at a single subject level. These results may be related to limitations of the methodology to measure plasma tau and other approaches to tau quantification may have better clinical performance.[42] In acute conditions, like traumatic brain injury and hypoxic brain injury, plasma tau concentration, measured using the same technology as the one employed here, increases rapidly and shows an apparent half-life of around 10 hours, which contrasts the half-life of tau in CSF, which is around 20 days. This may also explain the weak correlation of plasma with CSF tau and the poor diagnostic performance of plasma tau in chronic neurodegeneration.

NfL has emerged as a non-specific CSF and plasma biomarker of neuronal injury in degenerative and non-degenerative disorders.[10] Our results add to a large body of evidence showing that plasma or serum NfL concentrations are elevated in both FTLD and AD, compared to healthy individuals,[8,43,44] but it is non-specific and has poor discriminatory power between FTLD and AD or between FTLD clinical subtypes. Our results, however, support that plasma NfL has high prognostic value in FTLD, which has been previously been demonstrated in previous specific clinical subtypes, including bvFTD,[43] PPA,[42] ALS[47] and PSP.[9] This study found predictive value of plasma NfL in bvFTD and FTD-ALS, but not in PPA (nfvPPA or svPPA). This may be related to small PPA sample size or the inability of CDR-sb to capture the relatively slow progression of these phenotypes, as compared to FTD-ALS or bvFTD. This study also replicated the findings of previous investigations supporting

that, in AD, high plasma NfL correlates with faster worsening in global cognition and faster atrophy rates.[8,48] Our results are also consistent with previous reports showing a high correlation between plasma and CSF NfL.[9] This supports that plasma NfL may provide the similar valuable prognostic information available through CSF NfL, with the added value of being more convenient for clinical use. The most relevant novel contributions of this study are the study of plasma NfL in relation to specific pathology-confirmed FTLD subtypes and the analysis of its prognostic value for survival in FLTD-S. Seventy cases, (42% of the FTLD-S sample) had available neuropathology data. We studied a large FTLD-S cohort with available pathological data (*i.e.*, 70 cases or 42% of the FTLD-S sample). Plasma NfL was higher in FLTD-TDP than FTLD-tau or AD, a result driven by high plasma NfL concentrations in patients with FTD-ALS. Nevertheless, variability of plasma NfL is high, especially in FTLD-TDP, which makes it a poor discriminator of FLTD pathology subtypes.

The main strengths of this study are the inclusion of a wide range of phenotypes with follow-up information and the relatively high number of participants with available pathological information. But this study also has some limitations. We observed an association between baseline plasma NfL and the longitudinal CDR-sb change, but not with baseline CDR-sb. This may seem counterintuitive, but it is likely due to the poor ability of CDR-sb to accurately reflect FTLD-S disease severity. This problem could be solved with the use of novel clinical scales, specific for FTLD-S, such as the CDR plus NACC FTLD[49] or the multidimensional Impairment Rating (MIR).[50] The AD group was used mainly for contrast purposes, but conclusions about diagnostic and prognostic performance in this group should be interpreted cautiously and considering its relative small size. Finally, our study

included a small number of participants with genetic FTLD, but our results are in line with a recent European multicenter study of plasma NfL in genetic FTLD.[51]

In summary, this study supports the superiority of plasma NfL compared with plasma tau as a diagnostic and prognostic biomarker for both FTLD-S and AD.

References

1. Jack CR, Bennett DA, Blennow K, et al. NIA-AA Research Framework: Toward a biological definition of Alzheimer's disease. Alzheimers Dement J Alzheimers Assoc. 2018;14:535–562.

2. Dubois B, Feldman HH, Jacova C, et al. Advancing research diagnostic criteria for Alzheimer's disease: the IWG-2 criteria. Lancet Neurol. 2014;13:614–629.

3. Scheltens P, Blennow K, Breteler MMB, et al. Alzheimer's disease. The Lancet. 2016;388:505–517.

4. Elahi FM, Miller BL. A clinicopathological approach to the diagnosis of dementia. Nat Rev Neurol. 2017;13:457–476.

5. Bang J, Spina S, Miller BL. Frontotemporal dementia. The Lancet. 2015;386:1672–1682.

6. Lleo A, Irwin DJ, Illan-Gala I, et al. A 2-Step Cerebrospinal Algorithm for the Selection of Frontotemporal Lobar Degeneration Subtypes. JAMA Neurol. 2018;75:738–745.

7. Iqbal K, Liu F, Gong C-X. Tau and neurodegenerative disease: the story so far. Nat Rev Neurol. 2016;12:15–27.

8. Mattsson N, Andreasson U, Zetterberg H, Blennow K, for the Alzheimer's Disease Neuroimaging Initiative. Association of Plasma Neurofilament Light With Neurodegeneration in Patients With Alzheimer Disease. JAMA Neurol. 2017;74:557.

9. Rojas JC, Karydas A, Bang J, et al. Plasma neurofilament light chain predicts progression in progressive supranuclear palsy. Ann Clin Transl Neurol. 2016;3:216–225.

10. Bridel C, van Wieringen WN, Zetterberg H, et al. Diagnostic Value of Cerebrospinal Fluid Neurofilament Light Protein in Neurology: A Systematic Review and Meta-analysis. JAMA Neurol. 2019;76:1035.

11. Meeter LH, Dopper EG, Jiskoot LC, et al. Neurofilament light chain: a biomarker for genetic frontotemporal dementia. Ann Clin Transl Neurol. 2016;3:623–636.

12. Meeter LHH, Steketee RME, Salkovic D, et al. Clinical value of cerebrospinal fluid neurofilament light chain in semantic dementia. J Neurol Neurosurg Psychiatry. Epub 2019 May 23.

13. Alirezaei Z, Pourhanifeh MH, Borran S, Nejati M, Mirzaei H, Hamblin MR. Neurofilament Light Chain as a Biomarker, and Correlation with Magnetic Resonance Imaging in Diagnosis of CNS-Related Disorders. Mol Neurobiol [online serial]. Epub 2019 Aug 5. Accessed at: http://link.springer.com/10.1007/s12035-019-01698-3. Accessed September 29, 2019.

14. Irwin DJ, Lleo A, Xie SX, et al. Ante mortem cerebrospinal fluid tau levels correlate with postmortem tau pathology in frontotemporal lobar degeneration. Ann Neurol. 2017;82:247–258.

15. Grossman M, Elman L, McCluskey L, et al. Phosphorylated Tau as a Candidate Biomarker for Amyotrophic Lateral Sclerosis. JAMA Neurol. 2014;71:442–7.

16. Pijnenburg YAL, Verwey NA, van der Flier WM, Scheltens P, Teunissen CE. Discriminative and prognostic potential of cerebrospinal fluid phosphoTau/tau ratio and neurofilaments for frontotemporal dementia subtypes. Alzheimers Dement Diagn Assess Dis Monit. 2015;1:505–512.

17. Foiani MS, Woollacott IO, Heller C, et al. Plasma tau is increased in frontotemporal dementia. J Neurol Neurosurg Psychiatry. 2018;89:804–807.

18. Rascovsky K, Hodges JR, Knopman D, et al. Sensitivity of revised diagnostic criteria for the behavioural variant of frontotemporal dementia. Brain. 2011;134:2456–2477.

19. Gorno-Tempini ML, Hillis AE, Weintraub S, et al. Classification of primary progressive aphasia and its variants. Neurology. 2011;76:1006–1014.

20. Litvan I, Agid Y, Calne D, et al. Clinical research criteria for the diagnosis of progressive supranuclear palsy (Steele-Richardson-Olszewski syndrome): Report of the NINDS-SPSP International Workshop. Neurology. 1996;47:1–9.

21. Armstrong MJ, Armstrong MJ, Litvan I, et al. Criteria for the diagnosis of corticobasal degeneration. Neurology. 2013;80:496–503.

22. Brooks BR, Miller RG, Swash M, Munsat TL. El Escorial revisited: Revised criteria for the diagnosis of amyotrophic lateral sclerosis. Amyotroph Lateral Scler Other Motor Neuron Disord. 2000;1:293–299.

23. McKhann GM, Knopman DS, Knopman DS, et al. The diagnosis of dementia 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 J Alzheimers Assoc. Department of Neurology, Johns Hopkins University School of Medicine, Baltimore, MD, USA. guy.mckhann@jhu.edu; 2011. p. 263–269.

24. Knopman DS, Kramer JH, Boeve BF, et al. Development of methodology for conducting clinical trials in frontotemporal lobar degeneration. Brain. 2008;131:2957–2968.

25. Folstein MF, Folstein SE, McHugh PR. "Mini-mental state." J Psychiatr Res. 1975;12:189–198.

26. O'Bryant SE, Lacritz LH, Hall J, et al. Validation of the New Interpretive Guidelines for the Clinical Dementia Rating Scale Sum of Boxes Score in the National Alzheimer's Coordinating Center Database. Arch Neurol [online serial]. 2010;67. Accessed at: http://archneur.jamanetwork.com/article.aspx?doi=10.1001/archneurol.2010.115. Accessed December 19, 2019.

27. Rojas JC, Bang J, Lobach IV, et al. CSF neurofilament light chain and phosphorylated tau 181 predict disease progression in PSP. Neurology. 2018;90:e273–e281.

28. Dahnke R, Yotter RA, Gaser C. Cortical thickness and central surface estimation. NeuroImage. 2013;65:336–348.

29. Perry DC, Brown JA, Possin KL, et al. Clinicopathological correlations in behavioural variant frontotemporal dementia. Brain J Neurol. 2017;140:3329–3345.

30. Mackenzie IRA, Neumann M, Bigio EH, et al. Nomenclature for neuropathologic subtypes of frontotemporal lobar degeneration: consensus recommendations. Acta Neuropathol (Berl). 2009;117:15–18.

31. Mackenzie IRA, Neumann M, Bigio EH, et al. Nomenclature and nosology for neuropathologic subtypes of frontotemporal lobar degeneration: an update. Acta Neuropathol (Berl). 2010;119:1–4.

32. 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;56:1095–1097.

33. Hyman BT, Phelps CH, Beach TG, et al. National Institute on Aging-Alzheimer's Association guidelines for the neuropathologic assessment of Alzheimer's disease. Alzheimers Dement J Alzheimers Assoc. 2012;8:1–13.

34. Montine TJ, Phelps CH, Beach TG, et al. National Institute on Aging-Alzheimer's Association guidelines for the neuropathologic assessment of Alzheimer's disease: a practical approach. 2012.

35. Illan-Gala I, Pegueroles J, Montal V, et al. Challenges associated with biomarker-based classification systems for Alzheimer's disease. Alzheimers Dement Amst Neth. 2018;10:346–357.

36. Lleó A, Alcolea D, Martínez-Lage P, et al. Longitudinal cerebrospinal fluid biomarker trajectories along the Alzheimer's disease continuum in the BIOMARKAPD study. Alzheimers Dement. 2019;15:742–753.

37. Illan-Gala I, Pegueroles J, Montal V, et al. APP-derived peptides reflect neurodegeneration in frontotemporal dementia. Ann Clin Transl Neurol. 2019;6:2518–2530.

38. Wattmo C, Blennow K, Hansson O. Cerebro-spinal fluid biomarker levels: phosphorylated tau (T) and total tau (N) as markers for rate of progression in Alzheimer's disease. BMC Neurol. 2020;20:10.

39. Ljubenkov PA, Staffaroni AM, Rojas JC, et al. Cerebrospinal fluid biomarkers predict frontotemporal dementia trajectory. Ann Clin Transl Neurol. 2018;5:1250–1263.

40. Mattsson N, Zetterberg H, Janelidze S, et al. Plasma tau in Alzheimer disease. Neurology. 2016;87:1827–1835.

41. Staffaroni AM, Kramer AO, Casey M, et al. Association of Blood and Cerebrospinal Fluid Tau Level and Other Biomarkers With Survival Time in Sporadic Creutzfeldt-Jakob Disease. JAMA Neurol. Epub 2019 May 6.

42. Blennow K, Chen C, Cicognola C, et al. Cerebrospinal fluid tau fragment correlates with tau PET: a candidate biomarker for tangle pathology. Brain. Epub 2019 Dec 13.:awz346.

43. Lewczuk P, Ermann N, Andreasson U, et al. Plasma neurofilament light as a potential biomarker of neurodegeneration in Alzheimer's disease. Alzheimers Res Ther. 2018;10:71.

44. Rohrer JD, Woollacott IOC, Dick KM, et al. Serum neurofilament light chain protein is a measure of disease intensity in frontotemporal dementia. Neurology. 2016;87:1329–1336.

45. Steinacker P, Semler E, Anderl-Straub S, et al. Neurofilament as a blood marker for diagnosis and monitoring of primary progressive aphasias. Neurology. 2017;88:961–969.

46. Steinacker P, Anderl-Straub S, Diehl-Schmid J, et al. Serum neurofilament light chain in behavioral variant frontotemporal dementia. Neurology. 2018;91:e1390–e1401.

47. Lu C-H, Macdonald-Wallis C, Gray E, et al. Neurofilament light chain: A prognostic biomarker in amyotrophic lateral sclerosis. Neurology. 2015;84:2247–2257.

48. Mattsson N, Cullen NC, Andreasson U, Zetterberg H, Blennow K. Association Between Longitudinal Plasma Neurofilament Light and Neurodegeneration in Patients With Alzheimer Disease. JAMA Neurol. 2019;76:791.

49. Miyagawa T, Brushaber D, Syrjanen J, et al. Utility of the global CDR® plus NACC FTLD rating and development of scoring rules: Data from the ARTFL/LEFFTDS Consortium. Alzheimers Dement J Alzheimers Assoc. 2020;16:106–117.

50. Boxer AL, Gold M, Feldman H, et al. New directions in clinical trials for frontotemporal lobar degeneration: Methods and outcome measures. Alzheimers Dement. Epub 2019 Oct.:S1552526019353609.

51. van der Ende EL, Meeter LH, Poos JM, et al. Serum neurofilament light chain in genetic frontotemporal dementia: a longitudinal, multicentre cohort study. Lancet Neurol. 2019;18:1103–1111.

Tables

Table 1. Sample characteristics

Characteristics	FTLD-S	AD	HC	*p*-value
Number (%)	167 (63)	43 (16)	55 (21)	-
Age, y	65.8 ± 8.7[c]	65.2 ± 10.3[c]	52.2 ± 13.8[a,b]	***p*<.001**
Sex, males/females	83/84	16/27	25/30	*p*=.334
Education, y	15.8 ± 3.8[c]	16.4 ± 2.8	17.1± 2.5[a]	***p*=.005**
MMSE	23.1 ± 7[c]	21.5 ± 6.8[c]	28.9 ± 1.2[a,b]	***p*<.001**
CDR-sb[Ψ]	4.5 ± 3.4[c]	5.2 ± 2.7[c]	0 ± 0[a,b]	***p*<.001**
Memory composite	-2.4 ± 1.9[bc]	-4.1 ± 1.8[ac]	0.0 ± 0.7[a,b]	***p*<.001**
Language composite	-2.2 ± 1.8[c]	-1.7± 1.1[c]	0.0 ± 0.8[a,b]	***p*<.001**
Executive composite	-2.3 ± 1.6[c]	-2.8 ± 1.7[c]	0.0 ± 0.4[a,b]	***p*<.001**
Visuospatial composite	-1.6 ± 2.1[bc]	-3.1 ± 3.7[ac]	0.0 ± 0.7[a,b]	***p*<.001**
Longitudinal plasma sample, n (%)	72 (43)	27 (63)	24 (44)	*p*=.063
Clinical diagnosis at plasma sampling	43 bvFTD 28 nfvPPA 18 svPPA 36 PSP 32 CBS 10 FTD-ALS	36 amnestic 7 non-amnestic (3 language, 1 visuospatial and 3 frontal)	-	-
Clinical follow-up time, y	2.7 ± 1.8	3.1 ± 2	2.5 ± 2.5	*p=.111*
Deceased, n (%)	97 (58)[c]	18 (42)[c]	0 (0)[a,b]	***p*<.001**
Main pathological diagnosis	50 FTLD-Tau 18 FTLD-TDP 2 FTLD-FUS	12 AD	-	-
Genetic cases, n	11 *C9orf72* 7 *GRN* 4 *MAPT*	1 *PSEN*	-	-

Positive amyloid PET, n (%)	3/61 (5)[b]	22/25 (88)[ac]	1/12 (8)[b]	***p*<.001**
NIA-AA AD score,				
Absent, n (%)	20/70 (29)	0/12 (0)	-	-
Low, n (%)	39/70 (56)	0/12 (0)	-	-
Intermediate, n (%)	8/70 (11)	0/12 (0)	-	-
High, n (%)	3/70 (4)	12/12 (100)	-	-
Positive AD pathophysiology†, n (%)	12/101 (12)[b]	30/33 (91)[ac]	1/12 (8)[b]	***p*<.001**
Plasma biomarkers				
Tau, pg/mL*, median (Q1, Q3)	2.2 (1.8, 2.9)[ns]	2.5 (1.9, 3.2)[ns]	2.2 (1.8, 2.7)[ns]	*p*=.427 η^2=.01§
NfL, pg/mL*, median (Q1, Q3)	43.4 (28.9, 60.7)[b,c]	26.0 (20.5, 35.9)[a,c]	11.1 (8.1, 15.1)[a,b]	***p*<.001** η^2=.32§
CSF biomarkers				
Tau, pg/mL*, median (Q1, Q3)	45.1 (25.8, 69.5)[b]	68.0 (47.2, 123.6)[ac]	44.6 (30.9, 65.1)[a]	*p*=.02 η2=.04§
NfL, pg/mL*, median (Q1, Q3)	2325.5 (1412, 3433.5)[bc]	1015.5 (790.5, 1314)[ac]	429 (352, 640)[ab]	***p*<.001** η^2=.40§

Table 1 – Footnotes: Values reported are mean ± standard deviation. Statistically significant results are **bold**.

Ψ: data available in 237 (89%) of the participants: 161 (94%) FTLD, 33 (83%) AD and 43 (78%) HC.

†: Comorbid AD was defined as presence of positive amyloid PET or intermediate or high likelihood of AD pathology by NIA-Reagan criteria.

§: ANCOVA adjusted for age at plasma sampling, sex and CDR-sb.

*: These variables were not normally distributed across groups and were log-transformed to achieve normality before the statistical analyses.

**: 3 AD, 1 FTLD-UPS, 1 Argyrophilic grain disease, 1 Lewy body disease, 1 No neurodegeneration.

a: different from FTLD-S

b: different from AD

c: different from HC

Abbreviations: ANCOVA = analysis of covariance; FTLD = frontotemporal lobar degeneration; HC= healthy control; η2= partial eta square;

Table 2. Cox proportional hazard models with plasma biomarkers associated with Survival

	FTLD-S		AD	
Covariates	Hazard ratio [95% CI]	*p*-value	Hazard ratio [95% CI]	*p*-value
Age	1.007 [.980 to 1.034]	.633	.989 [.940 to 1.041]	.679
Sex	.935 [.696 to 1.679]	.729	1.063 [.324 to 3.489]	.920
CDR-sb	1.056 [.976 to 1.142]	.176	1.271 [1.016 to 1.590]	**.036**
Phenotype[a]		**.028**	NA	NA
Plasma biomarkers				
Plasma tau	.649 [.330 to 1.278]	.211	.808 [.111 to 5.870]	.833
Plasma NfL	2.019 [1.301 to 3.134]	**.002**[b]	.796 [.071 to 8.923]	.853

Table 3 – Footnotes: Age, sex and CDR-sb at baseline were introduced as covariates. In FTLD-S group, the phenotype at diagnosis (bvFTD, SD, PSP, CBS and ALS-FTD) was also added as a covariate. Statistically significant results ($p<.05$) are **bold**.

a: bvFTD group was set as reference group. Diagnosis of bvFTD was associated with decreased survival compared to svPPA diagnosis. Additionally, diagnosis of FTD-ALS was associated with decreased survival when compared to bvFTD diagnosis.

b: The addition of plasma NfL in the model containing age, sex, CDR-sb and phenotype significantly improve the model (Chi-square=9.785, $p=.002$).

Key: CDR-sb = Clinical dementia rating sum of boxes; CI = confidence interval; HR = Hazard ratio;

Figures

Figure 1. Group differences in plasma tau and NfL concentrations.

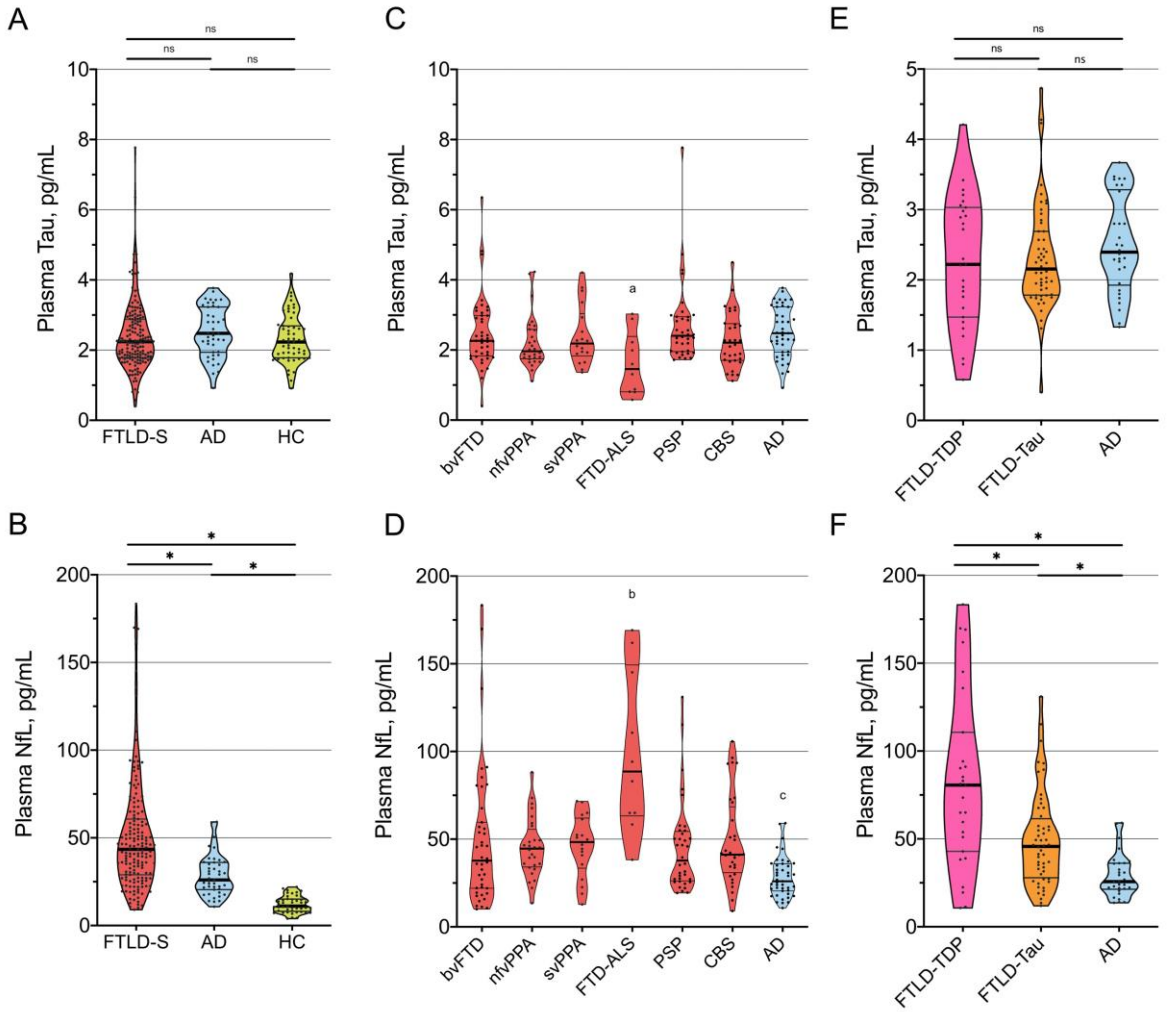

Figure 1 – footnotes: Group differences in the plasma levels of **A**) plasma tau and **B**) plasma NfL, between the main clinical groups. Groups differences in the plasma levels of **C**) tau and **D**) NfL between FTLD-S subgroups and AD group. Differences in the plasma levels of **E**) tau and **F**) NfL, between major neuropathological subtypes. In the panels **E-F**, participants with *C9orf72* (n=11) or *GRN* (n=7) mutations where included in the FTLD-TDP group (n=27) while participants with a *MAPT* mutation (n=4) were included in the FTLD-Tau group (n=52). The AD group in panels **E-F** included all AD participants with pathological confirmation of AD or a positive amyloid PET (n=30).

*: p<.001, Bonferroni's post-hoc test.

a: inferior to all other groups (p<.05, Bonferroni's post-hoc test) expect nfvPPA (p=.08).

b: superior to all other groups (p<.05, Bonferroni's post-hoc test).

c: inferior to all other groups (p<.05, Bonferroni's post-hoc test).

ns: no statistically significant differences between groups (p>.05).

Abbreviations: AD = Alzheimer's disease; FTLD-S = frontotemporal lobar degeneration-related syndromes; HC = Healthy controls.

Figure 2. Diagnostic value of plasma NfL for the differentiation of FTLD-S, AD and HC.

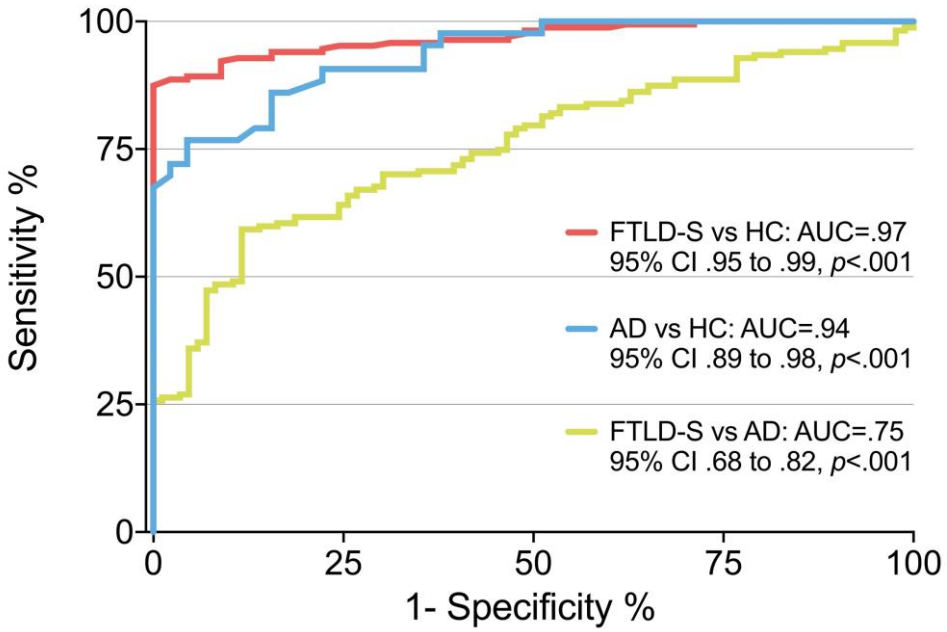

Figure 2 – footnotes: Diagnostic value of plasma NfL for the differentiation of FTLD-S, AD and HC.

Abbreviations: AUC= Area under the curve; CI = Confidence interval; FTLD-S = Frontotemporal lobar degeneration-related syndromes: AD= Alzheimer's disease; HC= Healthy controls: NfL = Neurofilament light chain.

Figure 3. Association of plasma biomarkers with clinical deterioration in FTLD-S and AD.

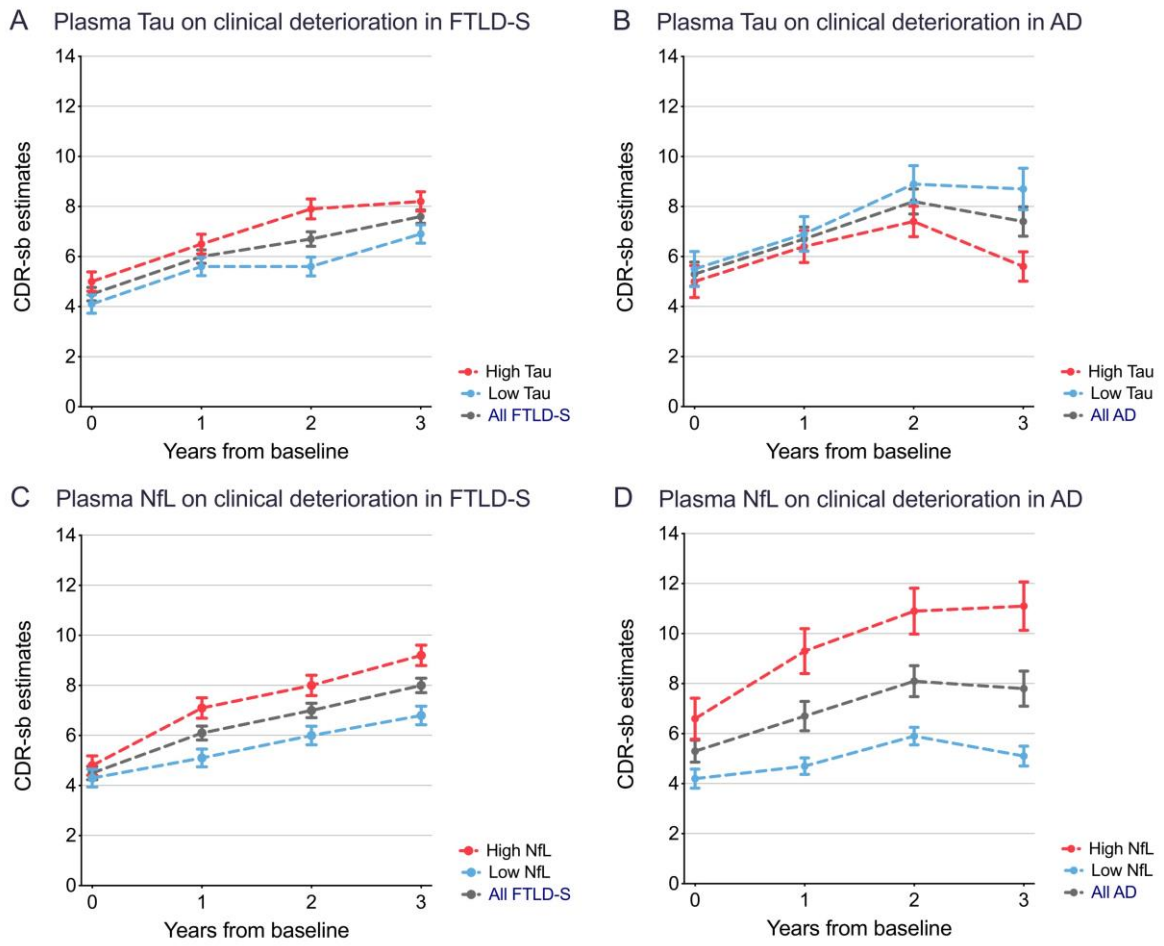

Figure 3 – footnotes: CDR-sb estimates were obtained from linear mixed-effects models adjusted for age, sex, basal CDR-sb and diagnosis clinical phenotype (in the FTLD-S group). For illustrative purposes, we show the groups with high levels of plasma biomarker (higher than the median) and low levels of plasma biomarker (lower than the median). Error bars represent 95% confidence intervals.

Key: CI = Confidence interval; FTLD = Frontotemporal lobar degeneration: NfL = Neurofilament light.

Figure 4. Relationship between plasma biomarkers and cortical thickness in FTLD-S and AD groups.

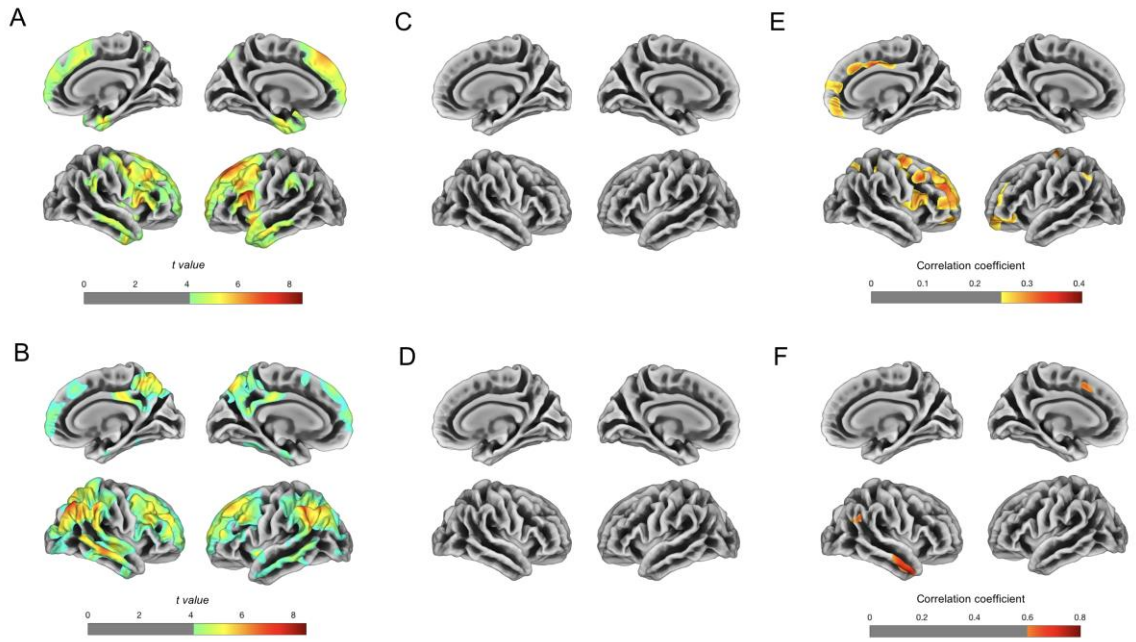

Figure 4 – Footnotes: Group comparison of cortical thickness between HC and FTLD-S (A) and AD (B); Correlation between basal plasma levels of tau and cortical thickness in FTLD-S group (C); Correlation between basal plasma levels of tau and cortical thickness in FTLD-S group (D); Correlation between basal plasma levels of tau and cortical thickness in AD group (E); Correlation between basal plasma levels of NfL and cortical thickness in FTLD-S group (F) Correlation between basal plasma levels of NfL and cortical thickness in AD group

For group comparisons only FDR ($p < 0.05$) are shown. For correlation analyses (C-F) the threshold for statistically significant correlation was set at $p < 0.001$.

Abbreviations: FDR = false discovery rate;

Figure 5. Kaplan-Meier Survival Curves for NfL in FTLD-S

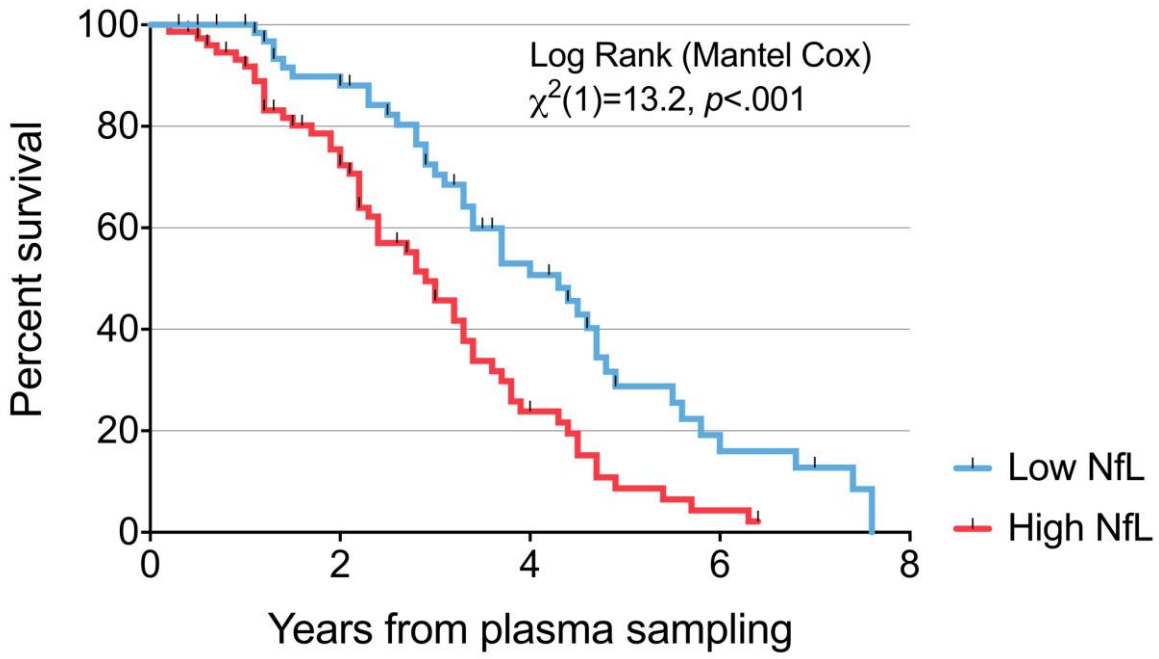

Figure 5 – footnotes: Kaplan-Meier Survival Curves for neurofilament light in FTLD-S. High NfL represent plasma NfL levels superior to 42 ng/mL (median split).

Abbreviations: FTLD = Frontotemporal lobar degeneration: NfL = Neurofilament light.