Title: Practical application of ADNI Plasma P-tau181 reference data to support diagnosis of Alzheimer's disease

Running Head: The application of Plasma P-tau181 to support Alzheimer's disease diagnosis

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Abstract

Objectives:

To assess plasma phosphorylated tau181 (p-tau181) levels in Alzheimer's disease (AD), cognitively impaired non-AD participants (CI non-AD) and Control participants in the Alzheimer's Disease Neuroimaging Initiative (ADNI) dataset that could potentially act as reference data for clinic diagnoses of AD.

Methods:

Data from 1558 participants (649 AD participants, 445 CI non-AD participants and 464 controls) were examined, comparing p-tau181 levels between Controls, A and other dementias, stratified by age.

Results:

There were significant differences in plasma p-tau181 values between Controls and those with AD at all ages up to 85 years. There were also significant differences between AD and CI non-AD participants up to the age of 85 years.

Conclusions:

Plasma P-tau181 may be a useful tool in the diagnosis of AD in those clinical settings where biomarkers have traditionally been less used. P-tau181 may be less useful as an aid to diagnosis in the very oldest-old. Further work is needed to establish the feasibility and utility of this biomarker within dementia diagnosis services not led by Neurologists, such as UK National Health Service Memory Services.

Keywords: p-tau181; Alzheimer's disease; diagnosis; biomarker

Key Points:

- In the ADNI dataset there were significant differences in plasma p-tau181 between controls and those with AD across all included age ranges, although there was overlap in 95% confidence intervals in the highest age ranges
- There were significant differences in plasma p-tau181 between AD participants and cognitively impaired participants without AD pathology.
- P-tau181 may be a useful tool to aid clinicians in the diagnosis of AD in situations where AD biomarkers have not been traditionally used.

Introduction

Blood-based biomarkers for Alzheimer's disease (AD) offer the potential to improve diagnostic accuracy within memory clinics where cerebrospinal fluid (CSF) biomarkers and Amyloid-Positron Emission Tomography (Amyloid-PET) are very rarely utilised[1,2]. Blood tests are more accessible and less invasive than CSF sampling and do not require staff who can perform lumbar punctures. They are less expensive than a PET scan and avoid radiation[3]. A blood immunoassay such as P-Tau181, which is a cheap and simple to test, offers a potential means of expanding the accessibility of AD biomarkers to a community clinical setting[4,5].

Concerns have been raised, however, about the choice of appropriate diagnostic cut-offs and potential high false negative rates for amyloid biomarkers in patients over the age of 70 years[6]. We have investigated whether plasma p-tau181 could distinguish between AD and healthy individuals and those with cognitive impairment who do not have AD pathology across different age ranges in the ADNI dataset. We wanted to look particularly at data from those individuals who were aged over 70, as they represent the majority of patients seen in NHS memory clinics where the novel availability of blood-based AD markers could potentially be valuable in making dementia diagnoses.

Methods

Study Design

We used anonymised data available from The Alzheimer's Disease Neuroimaging Initiative (ADNI-LONI[7]) database, downloaded in June 2021. ADNI is a longitudinal multicentre study within which participants with dementia and healthy control subjects, aged 55 to 90, have been followed for several years with serial clinical, imaging and biomarker assessments. The ADNI study is approved by regional ethical committees.

Participants

Inclusion and exclusion criteria for ADNI participants have been previously outlined[8]. We compiled data for all participants who had completed Clinical Dementia Rating (CDR) global scores, Amyloid-PET scan, and a plasma p-tau-181 test at a matched time point (within 90 days). Amyloid-PET scans in ADNI all used the AV45 tracer and a standardized uptake value ratio (SUVR) cut-off of 1.11 was considered 'Amyloid-PET-positive'[9]. Participants were considered as clinically affected AD cases if CDR-global >0 and Amyloid-PET was positive. Participants were considered as cognitively impaired (CI) non-AD participants if CDR >0 and Amyloid-PET was negative. Participants were classed as Controls if CDR-global=0 and Amyloid-PET was negative.

Plasma phosphorylated tau 181

Plasma p-tau181 concentrations were measured by the Single Molecule array (Simoa) technique. This assay was developed in the Clinical Neurochemistry Laboratory, University of Gothenburg, Sweden[4]. Two monoclonal antibodies (Tau12 and AT270) were used to measure N-terminal to mid-domain forms of P-tau181 (further assay information available in Karikari et al[4]).

Amyloid imaging

Amyloid PET imaging in ADNI was performed using 18F-florbetapir Aβ-PET (AV45): 70 MBq (10.0 mCi)±10%, 20 min (4×5 min frames) acquisition at 50–70 min post-injection. The ADNI images are pre-processed using a standardised pipeline[10]. The ADNI PET image acquisition is available online at adni-info.org. SUVR values were calculated as a ratio of mean uptake in six predefined cortical regions of interest (ROIs) to the mean cerebellum uptake[11].

Measurement of cognition

Cognitive performance was measured using the CDR Sum of Boxes (CDR-SB)[12]

Statistical Analysis

95% Confidence intervals were generated for plasma p-tau-181 data. Significance testing was undertaken using non-paired, 2-tailed student-t tests. All statistical analysis was undertaken in Microsoft Excel.

Results

A total of 649 AD participants and 464 Controls were identified; 54% were male. Mean (+/- SD) age was 74.9 (+/-7.2) years (range 55-95). There were 445 CI non-AD participants. The mean (+/- SD) age was 72.5 (+/-8.4). Age and gender composition is shown in Table 1 (Table 1).

Plasma p-tau181 in AD vs Control and CI non-AD groups stratified by age

Mean (+/-SD) plasma P-tau181 levels were higher in the AD group than in the Control group 23.2 (+/-11.0) vs. 15.0 (+/-11.5) pg/ml ($p<0.0001$) with no significant difference between sexes in disease or control groups (male vs female disease mean (SD) 23.2 (+/-11.8) vs 23.1 (+/-9.7), $p=0.86$; male vs female control mean (SD) 15.9 (+/-13.0) vs 14.0 (+/-9.7) pg/ml, $p=0.08$). Mean (+/-SD) plasma P-tau181 levels were significantly higher in the AD group than in the CI non-AD group 23.2 (+/-11.0) vs. 14.5 (+/-9.7) pg/ml ($p<0.0001$).

P-tau181 values provided a means of differentiating AD participants versus Controls and CI non-AD individuals on a plotted chart, showing non-overlapping 95% confidence intervals, and this difference was robust up to age 85. In the ages 85-95 there was overlap of the 95% confidence intervals for AD, Control and CI non-AD participants (Figures 1 and 2).

Discussion

Within ADNI participants, there were significant differences in p-tau181 levels between AD and controls and between AD and CI non-AD participants and these differences robustly

separated the diagnostic groups up to the age of 85 years. Beyond 85 years, the measure was less robust in differentiating between both AD and control participants and AD and CI non-AD participants. This larger cohort dataset shows consistency with a metanalysis of pooled p-tau181 levels of AD and controls, where the weighted mean difference was 11.7 pg/ml and pooled sensitivity and sensitivity using SIMOA was 0.89 CI (0.8-0.93) and 0.86 CI (0.79-0.91) respectively[13].

The data suggest that Plasma p-tau181 was effective in differentiating participants a) with an impairment on CDR score who were Amyloid PET positive (AD) from b) participants with an impairment on CDR score who were Amyloid PET negative (CI non-AD) and c) those who had a normal CDR score and were Amyloid PET negative (Controls) in the ADNI dataset. If this observation were to hold true in other populations of patients who are investigated for suspected dementia, this blood biomarker would have utility in clinical practice.

There was some overlap of confidence intervals in the 85–95-year-old age range. This may be due to a smaller sample size of controls and participants at this age range or a consequence of rising plasma p-tau-181 levels in the ageing population or possible confounding co-morbidities including cerebrovascular disease [14–16].

Possible factors that may reduce applicability of these data as reference values to help with diagnosis in individual cases within NHS memory services might include differences in plasma p-tau181 sample collection and storage in clinical practice and the robustness of assays in clinical laboratories. Encouragingly, such assays are now highly automated and have less potential for operator error although they are reliant on the quality of the sample taken[17].

ADNI provides a longitudinal dataset including older participants (including large numbers of participants in their 80s). Limitations of the ADNI dataset include that it is a research sample which may limit the extent to which the results can be generalised into clinical practice where co-morbidities (e.g. poor overall medical health and cerebrovascular disease) may be more common.

Further studies are needed to validate the plasma P-tau181 levels against established CSF measures using UK laboratory-based assays and to establish cut-offs in a clinical setting.

Future research directions include investigating the feasibility and acceptability of the introduction of use of the Plasma p-tau-181 biomarker in UK Memory Services and whether clinicians in UK NHS Memory Services find the marker helpful in making AD diagnoses.

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Table 1: Control and AD and CI non-AD participants categorised by age and gender

	Control (N=464)				Alzheimer's Disease (N=649)				CI Non-AD (N=445)			
Age(y)	N	Gender		Mean age	N	Gender		Mean age	N	Gender		Mean age
		Male	Female			Male	Female			Male	Female	
55-64	15	4 (27%)	11 (73%	62	59	23 (39%	36 (61%)	61	94	46 (49%	48 (51%)	61
65-69	107	49 (46%	58 (54%	67	83	41 (49%	42 (51%)	67	74	38 (51%	36 (49%)	67
70-74	120	56(47%	64 (53%	72	146	81 (55%	65 (45%)	72	95	54 (57%	41 (43%)	72
75-79	117	67 (57%	50 (43%	77	171	113 (66	58 (34%)	77	87	48 (55%	39 (45%)	77
80-84	60	29 (48%	31 (52%	82	124	81 (65%	43 (53%)	82	57	31 (54%	26 (46%)	82
85-95	45	28 (62%	17 (38%	88	66	37 (58%	27 (42%)	88	38	25 (66%	13 (34%)	88

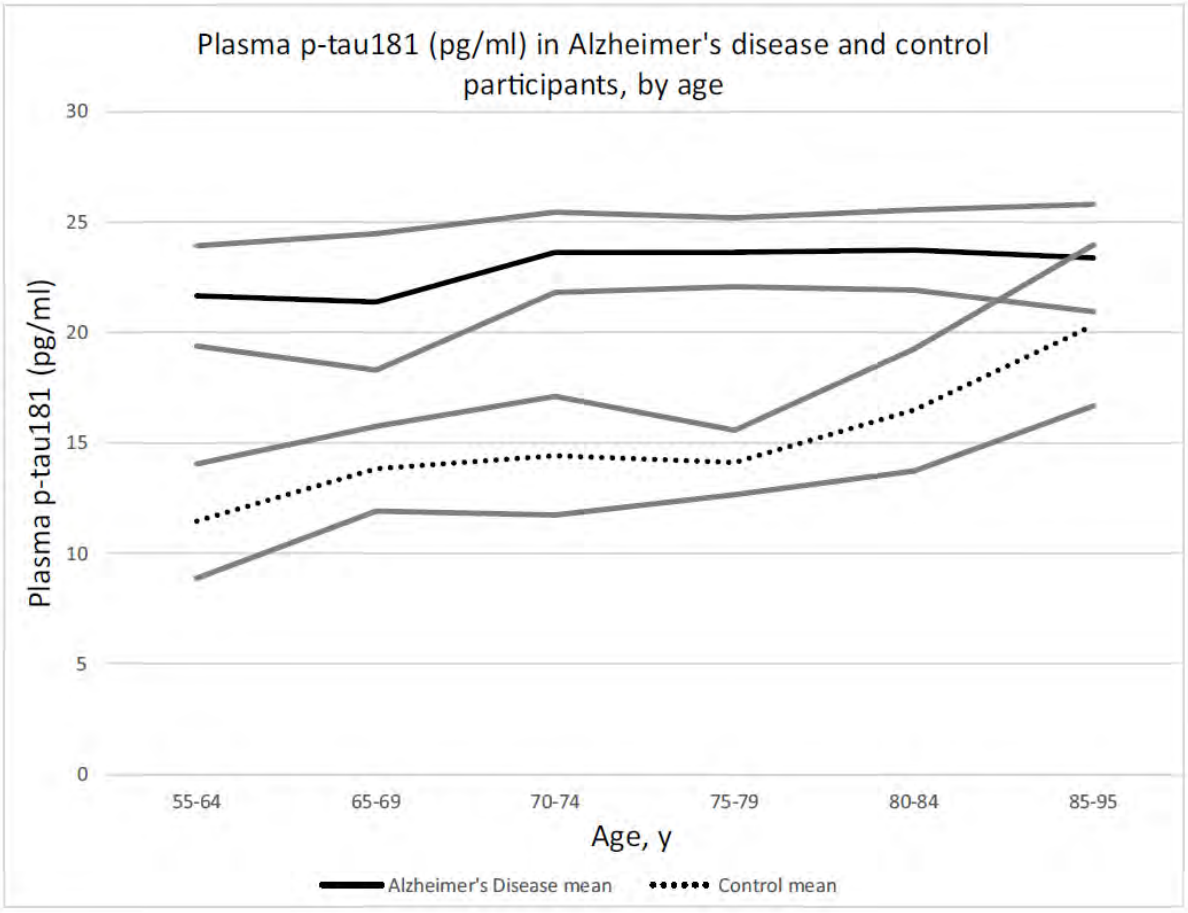

Figure 1: Concentrations of plasma p-tau181 (pg/ml) in Alzheimer's disease participants (black) and control participants (dotted) stratified by age, from the ADNI dataset. Data shown as mean plasma p-tau181 with 95% confidence intervals (grey). There were differences in P-tau181 concentration between AD participants versus controls, with non-overlapping 95% confidence intervals up to age 85 and an overlap of confidence intervals in the 85–95-year age range.

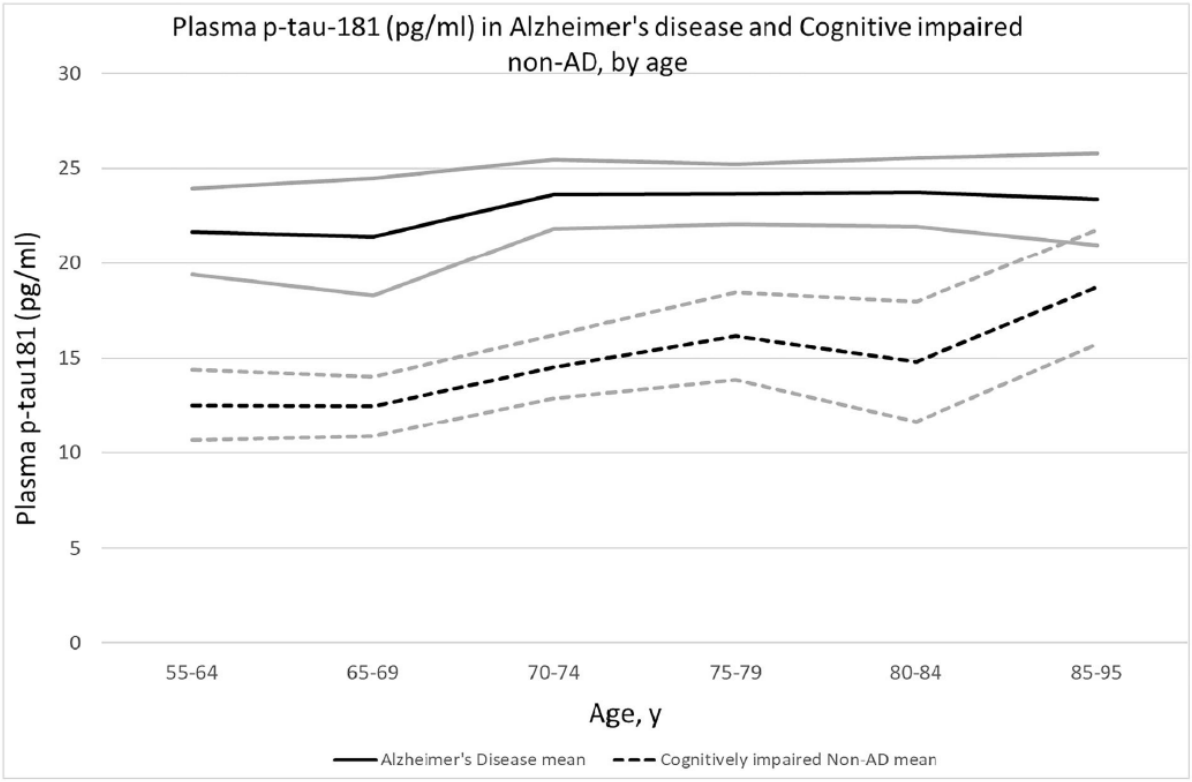

Figure 2: Concentrations of plasma p-tau181 (pg/ml) in Alzheimer's disease participants (black) and CI non-AD participants, defined as CDR>0 and "amyloid PET negative" (dotted) stratified by age, from the ADNI dataset. Data shown as mean plasma p-tau181 with 95% confidence intervals (grey). There were differences in P-tau181 concentration between AD participants versus CI non-AD participants, with non-overlapping 95% confidence intervals up to age 85 and an overlap of confidence intervals in the 85–95-year age range.