BRAIN
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Preclinical Alzheimer's disease biomarkers accurately predict cognitive and neuropathological outcomes

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Alzheimer's disease biomarkers are widely accepted as surrogate markers of underlying neuropathological changes. However, few studies have evaluated whether preclinical Alzheimer's disease biomarkers predict Alzheimer's neuropathology at autopsy. We sought to determine whether amyloid PET imaging or CSF biomarkers accurately predict cognitive outcomes and Alzheimer's disease neuropathological findings.

This study included 720 participants, 42–91 years of age, who were enrolled in longitudinal studies of memory and aging in the Washington University Knight Alzheimer Disease Research Center and were cognitively normal at baseline, underwent amyloid PET imaging and/or CSF collection within 1 year of baseline clinical assessment, and had subsequent clinical follow-up. Cognitive status was assessed longitudinally by Clinical Dementia Rating®. Biomarker status was assessed using predefined cut-offs for amyloid PET imaging or CSF p-tau181/amyloid-β42. Subsequently, 57 participants died and underwent neuropathologic examination. Alzheimer's disease neuropathological changes were assessed using standard criteria. We assessed the predictive value of Alzheimer's disease biomarker status on progression to cognitive impairment and for presence of Alzheimer's disease neuropathological changes.

Among cognitively normal participants with positive biomarkers, 34.4% developed cognitive impairment (Clinical Dementia Rating > 0) as compared to 8.4% of those with negative biomarkers. Cox proportional hazards modelling indicated that preclinical Alzheimer's disease biomarker status, APOE ε4 carrier status, polygenic risk score and centred age influenced risk of developing cognitive impairment. Among autopsied participants, 90.9% of biomarker-positive participants and 8.6% of biomarker-negative participants had Alzheimer's disease neuropathological changes. Sensitivity was 87.0%, specificity 94.1%, positive predictive value 90.9% and negative predictive value 91.4% for detection of Alzheimer's disease neuropathological changes by preclinical biomarkers. Single CSF and amyloid PET baseline biomarkers were also predictive of Alzheimer's disease neuropathological changes, as well as Thal phase and Braak stage of pathology at autopsy. Biomarker-negative participants who developed cognitive impairment were more likely to exhibit non-Alzheimer's disease pathology at autopsy.

The detection of preclinical Alzheimer's disease biomarkers is strongly predictive of future cognitive impairment and accurately predicts presence of Alzheimer's disease neuropathology at autopsy.

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Introduction

Alzheimer's disease is the most common cause of dementia in older adults and is characterized neuropathologically by the presence of extracellular plaques consisting of aggregated amyloid-β (Aβ) peptide and neurofibrillary tangles consisting of intraneuronal inclusions of aggregated, hyperphosphorylated tau protein.[1] Substantial evidence supports the observation that Alzheimer's disease neuropathologic changes (ADNC) accrue in the brain for years before the onset of symptomatic disease, representing a preclinical phase. This has been supported by: post-mortem studies demonstrating presence of ADNC in a substantial number of cognitively normal older adults[2–4]; detection of intracerebral accumulation of amyloid over time by PET imaging, which is associated with increased risk of progression to symptomatic Alzheimer's disease[5,6]; inverse correlation of CSF levels of $A\beta_{42}$ with amyloid imaging markers in cognitively normal individuals,[7] which is predictive of progression to symptomatic Alzheimer's disease and whole brain atrophy[8]; and reductions in $A\beta_{42}$ and elevated levels of total tau (t-tau) and tau phosphorylated at position 181 (p-tau181) that appear decades before the expected time of onset of symptomatic disease in autosomal dominant Alzheimer's disease cohorts.[9,10] Several longitudinal studies have assessed outcomes in older adults with preclinical Alzheimer's disease and demonstrated that risk of progression from preclinical to symptomatic Alzheimer's disease increases over time.[11–15]

Amyloid PET imaging and CSF p-tau181/$A\beta_{42}$ are accepted methods for diagnosing Alzheimer's disease in patients with established cognitive impairment and are widely viewed as valid modalities for detecting the presence of ante-mortem Alzheimer's disease neuropathology. However, the early neuropathologic correlation studies that validated these methods used cohorts with small participant numbers[16–19] or were based on post-mortem biomarker assessment.[20] Larger follow-up studies in autopsy-confirmed cohorts have either focused on terminally ill participants with short life expectancy[21] or cohorts primarily consisting of cognitively impaired participants.[22–25] Most studies evaluating performance of CSF or amyloid PET biomarkers in the preclinical phase of the disease have evaluated longitudinal changes in biomarkers over time,[26,27] or assessed the performance of such tests relative to one another[15,28,29] or in relation to the onset of cognitive impairment.[8,14] Few studies of preclinical Alzheimer's disease have validated these tests in relation to gold-standard neuropathologic diagnosis and have generally been limited by small cohort sample size prohibiting the assessment of diagnostic accuracy.[11] With the development of disease-modifying therapies, clinical trials are now moving towards primary prevention paradigms, enrolling cognitively normal participants based on the presence of preclinical Alzheimer's disease biomarkers.[30,31] Therefore, it is crucial to validate that biomarker testing in this population reliably detects individuals with confirmed Alzheimer's disease neuropathological change.

In this study, we have evaluated whether the aggregate use of amyloid PET imaging or CSF biomarker testing in cognitively normal participants in a large study of community-dwelling participants accurately predicts cognitive outcomes and ADNC at autopsy.

Methods

Participants

Participants were cognitively normal community-dwelling individuals, aged 42.5–91.2 years at baseline, who were enrolled in longitudinal studies of memory and aging at the Knight Alzheimer Disease Research Center (ADRC) at the Washington University School of Medicine in St Louis. All participants underwent annual clinical assessments, including Clinical Dementia Rating® (CDR®)[32] and Mini-Mental State Examination (MMSE).[33] The CDR is a well-validated instrument consisting of semi-structured interviews with the participant and their collateral source as conducted by an experienced clinician to identify intra-individual changes in participant cognitive and functional abilities. Assigned CDR scores describe dementia severity (CDR 0 = cognitively normal, CDR 0.5 = very mild dementia, CDR 1 = mild dementia, CDR 2 = moderate dementia, CDR 3 = severe dementia). Participants were included in this study if they were CDR 0 at baseline clinical assessment, underwent CSF collection by lumbar puncture (LP) and/or amyloid PET imaging [either ¹¹C Pittsburgh compound B (PIB) or ¹⁸F-AV-45 (AV45)] within 12 months of baseline clinical assessment, and had at least one additional clinical assessment after their baseline visit.

For participants who progressed to CDR > 0 during longitudinal follow-up, aetiologic diagnoses were assigned by experienced clinicians within the Knight ADRC following review of clinical history obtained at time of CDR assessment along with limited cognitive testing and based on standard research criteria for diagnosis of various dementia syndromes. Clinical diagnoses were determined independently of knowledge of biomarker status. Participant clinical assessments in this study were performed between 6 March 2002 and 1 January 2019.

Standard protocol approvals, registrations and patient consents

The Washington University Human Research Protection Office approved all study procedures. Written informed consent was obtained according to the Declaration of Helsinki from all participants before study enrolment.

Amyloid imaging

Participants underwent a PET scan with either PIB or AV45. PIB PET imaging was performed with a Siemens 962 HR + ECAT PET or

Biograph mCT scanner (Siemens/CTI). AV45 PET imaging was performed with a Siemens Biograph mMR scanner (Siemens/CTI). Data were processed using regions of interest derived from T_1-weighted MRIs using Freesufer.[34] Data from the 30–60 min post-injection window for PIB and the 50–70 min post-injection window for AV45 were converted to standardized uptake value ratios (SUVRs) relative to the cerebellar cortex. Data were partial volume corrected using a geometric transfer matrix.[35] Values from regions known to be involved in Alzheimer's diseaseee were averaged together to provide a summary measure of amyloid deposition. Amyloid PET positivity was defined by previously established SUVR cut-offs for PIB (>1.42)[26] and AV45 (>1.22).[36] For some analyses, PIB and AV45 tracer data were converted to centiloid units[37–39] and combined using previously established cut-offs (centiloid units > 16.4).

CSF collection and processing

Participants underwent CSF collection by LP. CSF was collected as previously described.[7] Participants underwent LP at ~8 AM following overnight fasting. CSF (20–30 ml) was collected in a 50-ml polypropylene tube via gravity drip using an atraumatic Sprotte 22-G spinal needle. The tube was inverted gently to disrupt potential gradient effects and centrifuged at low speed to pellet any cellular debris. The CSF was then aliquoted into polypropylene tubes and stored at –80°C. To limit issues related to assay drift[40] and lot-to-lot variability[41] associated with commercial enzyme-linked immunosorbent assays, only those participants whose CSF was processed using next-generation automated Roche Elecsys assays were included for analysis. CSF $A\beta_{42}$, t-tau and p-tau181 were measured with corresponding Elecsys immunoassays on the Roche cobas e601 analyser. CSF biomarker positivity was defined by the ratio of CSF p-tau181/Aβ42 using a cut-off of >0.0198. This biomarker ratio and this cut-off were chosen to yield high concordance and positive percent and negative percentage agreement with PIB status.[28]

APOE ε genotyping and computation of polygenic risk score

APOE ε genotype status of each participant was obtained using previously described methods[42] and provided by the Genetics core of the Knight ADRC. Polygenic risk score (PRS) for each participant derived using genotype status across 24 single nucleotide polymorphisms known to be associated with Alzheimer's disease risk were computed and provided by the Genetics core. PRS are presented as summed $\log_2$-transformed odds ratio (OR). Methods describing the computation of PRS have been described elsewhere.[43,44]

Neuropathological assessment

Eighty-two participants died during the study period. Of these, 57 (70%) provided autopsy consent and the brains of those participants were evaluated by experienced neuropathologists within the Knight ADRC Neuropathology Core. Autopsies in this study were performed between 6 September 2004 and 10 July 2020. Standard procedures[45] and consensus criteria[46–60] were used to establish neuropathological diagnoses. For purposes of this study, Alzheimer's disease pathology was scored as present or absent based on specimen categorization by either National Institute on Aging (NIA)-Reagan[61] or NIA-Alzheimer's Association (AA) criteria,[62] depending on when each autopsy was completed. For those specimens categorized using NIA-Reagan criteria, Alzheimer's disease pathology was considered 'present' if specimens were

classified as having either intermediate or high likelihood that Alzheimer's disease pathological changes underlie dementia. For those specimens categorized by NIA-AA criteria, Alzheimer's disease pathology was considered 'present' if specimens were classified as having either intermediate or high ADNC; by NIA-AA criteria, intermediate and high ADNC (but not low ADNC) are considered sufficient to account for observed dementia. Braak neurofibrillary tangle stage[46] and Thal amyloid phase[48] were recorded for each participant, when available. Comorbid neuropathological diagnoses, including Lewy body pathology, hippocampal sclerosis, TDP-43 inclusions, frontotemporal lobar degeneration, other primary tauopathies, cerebrovascular disease, infarctions and haemorrhages were also recorded.

Study design

In participants who were cognitively normal at the time of biomarker assessment, we assessed whether a 'positive' Alzheimer's disease molecular biomarker value predicted progression to cognitive impairment (CDR > 0) and/or the presence of ADNC at autopsy. For participants who completed amyloid PET imaging as described previously, this modality was used to classify biomarker positivity. For participants who underwent CSF collection, CSF p-tau181/Aβ_{42} ratio was used to establish biomarker positivity. For participants assessed by both modalities, amyloid PET was used to define biomarker status to maximize the number of longitudinal biomarker data points available for assessment. ADNC was defined as described previously.

Statistical analyses

Summary statistics for continuous variables (means/medians and standard deviations) and categorical variables (percentages) were calculated for all participants as well as the biomarker-negative and -positive subgroups. For biomarker-negative versus -positive comparisons, two-sample *t*-tests were used for continuous variables and z-score tests for two proportions were used for categorical variables. Wilcoxon rank sum tests for continuous variables and Chi-square tests and binomial tests for proportions were used if the parametric test assumptions were not met. Cox proportional hazards models with death as a competing risk were used to compare the probability of progression to CDR > 0 for biomarker-negative and -positive participants. Biomarker positivity was used as the main effect and centred age (normalized age used for statistical modelling), APOE ε4 carrier status (0 = non-carrier, 1 = carrier), gender, self-reported race, centred education (years), PRS, the time interval from baseline to last clinical assessment (years) and all two-way interaction terms for these variables were included as covariates. Centred age and centred education were calculated by subtracting overall means of each metric across the entire dataset from individual values. For these models, hazard ratios (HR) and their 95% confidence intervals with *P*-values were reported. Logistic regression models were used to assess the effect of baseline positive biomarkers on the odds of intermediate-high ADNC at autopsy for those participants with neuropathological data (*n* = 46). As with the Cox models, biomarker positivity, centred age, APOE ε4 carrier status, gender, race, centred education, PRS and the time interval from baseline to death were included in the models. Univariate logistic regression models were used to assess the effect of each baseline individual biomarker (CSF $A\beta_{42}$, p-tau181, t-tau, p-tau181/Aβ42, amyloid PET centiloid) on the odds of intermediate-high ADNC at autopsy. Univariate ordinal logistic regression models were used to assess the effect of each baseline individual

biomarker on the cumulative odds of less advanced Braak stage and Thal phase. For each ordinal regression model, assumption of proportional odds was met. For binomial and ordinal logistical regression models, the ORs and their 95% confidence intervals with P-values were reported.

Data availability

All data from analyses in this paper will be deposited in the Washington University Knight ADRC dataset and made available to qualified investigators by request following approval by the Knight ADRC data request committee.

Results

Participants

Table 1 summarizes baseline characteristics and cognitive outcomes for participants in this study. Race was self-reported by participants. A total of 720 participants met criteria for inclusion as described previously; 117 (16.3%) underwent CSF collection only, 252 (35.0%) underwent amyloid PET imaging only and 351 (48.7%) completed both CSF collection and amyloid PET (in these participants, amyloid PET was used for biomarker status determination). 22.5% of biomarker-positive and 14.4% of biomarker-negative participants were defined on the basis of CSF assessment, whereas 77.5% of biomarker-positive and 85.6% of biomarker-negative participants were defined on the basis of amyloid PET assessment.

A total of 160 participants (22.2%) were designated biomarker-positive at baseline assessment (91.9% age ≥ 65 years), while 560 participants (78.8%) were designated biomarker-negative (68.0% age ≥ 65 years). Participants defined as biomarker-positive tended to be older ($P < 0.0001$), were more likely $APOE$ ε4 allele carriers ($P < 0.0001$), and were more likely to self-report their race as White ($P = 0.0007$) as compared to biomarker-negative participants. The mean interval from baseline to final clinical assessment was 5.8 years (median 5.3 years; range 0.8–15.9 years) and this did not differ based on biomarker designation. Biomarker-positive participants had higher overall mortality compared to biomarker-negative participants ($P = 0.0058$).

Cognitive outcomes

A significantly higher proportion of biomarker-positive participants (34.4%) progressed from normal cognition (CDR = 0) to impaired cognition (CDR > 0) at last assessment as compared to biomarker-negative participants (8.4%) during the period of evaluation ($P < 0.0001$). This was reflected by a lower average MMSE at the time of last clinical assessment in biomarker-positive versus -negative participants ($P < 0.0001$). When evaluated by time interval of follow-up from initial biomarker to final assessment, a higher proportion of biomarker-positive (53.3%) compared to biomarker-negative (12.5%) participants progressed to CDR > 0 when followed for 10–15 years from baseline assessment ($P = 0.0003$). Among all biomarker-positive participants who progressed to CDR > 0 ($n = 55$) at last assessment, the average time interval from detection of positive biomarkers to development of cognitive impairment was 3.9 years (SD 2.5 years, range 0.9–10.9 years). There were no observed differences in these outcomes between African-American/Black participants and White participants, however the small number of biomarker-positive African-American/Black participants limited detection of differences due to lack of power.

To better assess the underlying clinical phenotypes associated with progression to CDR > 0 among biomarker-positive and biomarker-negative participants, the clinical diagnoses assigned at final assessment are listed in Table 1. Alzheimer's disease dementia was the assigned diagnosis in 80% of biomarker-positive CDR progressors compared to 55.3% of biomarker-negative CDR progressors ($P = 0.0073$). Uncertain dementia, reflecting either atypical pattern or questionable level of impairment, was the assigned diagnosis in 12.7% of biomarker-positive CDR progressors compared to 36.1% of biomarker-negative CDR progressors ($P = 0.0054$).

To determine the effect of biomarker status, age, sex, self-reported race, years of education, $APOE$ ε4 carrier status, PRS and interval from baseline assessment to final assessment on risk of progression to CDR > 0, multivariable Cox proportional hazard regression with death as a competing risk was used to model the effects of these covariates on CDR progression (Supplementary Table 1). Due to missing covariates, only 680 of 720 available participants were included in this analysis. In this model, preclinical Alzheimer's disease biomarker positivity (HR = 2.794; $P < 0.0001$) and $APOE$ ε4 + carrier status (HR = 1.941; $P = 0.0266$) had the strongest independent effect on risk of developing cognitive impairment. Age at baseline (independent of $APOE$ ε4 carrier status) and PRS also significantly contributed to risk of developing cognitive impairment. There was a statistically significant interaction between $APOE$ ε4 carrier status and age ($P = 0.0045$), highlighting a reduced effect of age on risk of CDR progression among $APOE$ ε4 carriers as compared to $APOE$ ε4 non-carriers. All other tested covariates in the model did not significantly contribute to increased risk of developing cognitive impairment. Cumulative incidence function curves derived from this model (Fig. 1A) demonstrate the increased probability of CDR progression over time in biomarker-positive compared to biomarker-negative individuals.

There was a surprisingly large number of biomarker-negative individuals with CDR progression ($n = 47$ of total $n = 560$), among which 55.3% were assigned a clinical diagnosis of Alzheimer's disease dementia (described previously). To better understand the relationship between longitudinal biomarkers, biomarker status and cognitive outcomes, participants from the original cohort with at least one additional biomarker assessment of the same modality used to define original biomarker status were separately analysed and presented in Supplementary Table 2. Nearly all of these participants with longitudinal biomarkers were assigned biomarker status based on amyloid PET imaging. The biomarker-positive cohort had significantly higher baseline ($P = 0.0448$) and final ($P = 0.0337$) amyloid PET centiloid units among CDR progressors compared to non-progressors, whereas there was no significant difference in baseline or final amyloid PET centiloid units between the CDR progressors and non-progressors in the biomarker-negative cohort. A total of 28 biomarker-negative participants in this cohort converted to biomarker-positive status over the course of longitudinal biomarker assessment, however only one of these progressed to CDR > 0. This is supported by plots of longitudinal amyloid PET trends over time (Fig. 1B). Among biomarker-negative CDR progressors, there is no trend towards increased amyloid accumulation.

Neuropathological outcomes

During the course of follow-up, 82 participants died and 57 (70%) came to autopsy. Clinical characteristics and neuropathological outcomes are detailed in Table 2. Average age at death was 85.5 years and was not significantly different between biomarker-negative and -positive participants. The average interval from

Table 1 Demographics and clinical characteristics for cognitively normal (CDR = 0) participants at baseline

	All participants		Biomarker negative		Biomarker positive		P-value[a]
	n	Mean (SD), or %	*n*	Mean (SD), or %	*n*	Mean (SD), or %	
Age, mean (SD), years	720	69.2 (9.2)	560	67.9 (9.3)	160	73.9 (6.8)	**<0.0001**
Male sex, %	302	41.9	231	41.2	71	44.4	0.4778
Race, %							
Non-Hispanic White	621	86.3	470	83.9	151	94.4	**0.0007**
African-American	96	13.3	87	15.5	9	0.6	**0.0012**
Asian	2	0.3	2	0.4	0	0	0.4237
More than one	1	0.1	1	0.2	0	0	0.5687
APOE ε4 carriers, %	241	33.5	147	26.2	94	58.7	**<0.0001**
PRS, mean (SD)	683	−0.00355 (0.00729)	531	−0.00405 (0.00711)	152	−0.00181 (0.00762)	**0.0007**
Education, mean (SD), years	720	16.0 (2.6)	560	16.0 (2.5)	160	16.0 (3.0)	0.9890
Baseline MMSE, mean (SD)	720	29.1 (1.2)	560	29.1 (1.1)	160	29.0 (1.3)	0.1954
Final MMSE, mean (SD)	718	28.7 (2.1)	560	28.9 (1.5)	158	27.7 (3.3)	**<0.0001**
Interval from baseline assessment to final assessment, mean (SD) [median], years	720	5.8 (3.4) [5.2]	560	5.8 (3.5) [5.2]	160	5.7 (3.1) [5.3]	0.8415
Interval from baseline biomarker to final assessment, mean (SD) [median], years	720	5.6 (3.4) [5.1]	560	5.6 (3.5) [5.1]	160	5.6 (3.1) [5.1]	0.8509
Interval from baseline biomarker to study conclusion[b] or death, mean (SD) [median], years	720	7.6 (3.7) [7.5]	560	7.7 (3.8) [7.3]	160	7.6 (3.4) [7.5]	0.9424
Mortality, %	82	11.4	54	9.6	28	17.5	**0.0058**
Progression to CDR > 0, %[c]	**102**	**14.17**	**47**	**8.4**	**55**	**34.4**	**<0.0001**
CDR at final assessment							
0	633	87.9	521	93.0	112	70.0	**<0.0001**
0.5	60	8.3	31	5.6	29	18.1	**<0.0001**
1	24	3.3	8	1.4	16	10.0	**<0.0001**
2	3	0.4	0	0	3	1.9	**0.0012**
3	0	0	0	0	0	0	–
Progression to CDR > 0 by time interval of follow-up from initial biomarker to final assessment, %							
0–5 years	39	11.0	17	6.1	22	28.6	**<0.0001**
5–10 years	45	16.2	20	9.5	25	36.8	**<0.0001**
10–15 years	17	19.5	9	12.5	8	53.3	**0.0003**
15–20 years	1	50.0	1	50.0	0	–	–
Clinical diagnoses assigned at time of final assessment for progressors (CDR > 0), %							
AD dementia[d]	70	68.6	26	55.3	44	80.0	**0.0073**
Parkinson's disease dementia	2	1.0	0	0	2	3.6	0.1868
DLB	1	1.0	1	2.1	0	0	0.2757
Vascular dementia	3	3.0	2	4.2	1	1.8	0.4654
Non-AD dementia	2	2.0	1	2.1	1	1.8	0.9124
Uncertain dementia[e]	24	23.5	17	36.1	7	12.7	**0.0054**
No. defined by CSF biomarker, %	117	16.3	81	14.4	36	22.5	**0.0151**
t-tau, mean (SD), pg/ml	117	234.8 (104.7)	81	194.0 (57.0)	36	326.8 (127.9)	**<0.0001**
p-tau181 mean (SD), pg/ml	117	21.5 (11.3)	81	16.6 (4.6)	36	32.6 (13.8)	**<0.0001**
Aβ42 mean (SD), pg/ml	117	1382.5 (623.6)	81	1622.4 (578.3)	36	842.8 (302.7)	**<0.0001**
p-tau181/Aβ42, mean (SD)	117	0.020 (0.018)	81	0.011 (0.003)	36	0.041 (0.020)	**<0.0001**
No. defined by amyloid PET biomarker	603	83.7	479	85.5	124	77.5	**0.0151**
PIB SUVR, mean (SD)	406	1.300 (0.610)	318	1.021 (0.095)	88	2.307 (0.624)	**<0.0001**
AV45 SUVR, mean (SD)	197	1.135 (0.459)	161	0.951 (0.160)	36	1.958 (0.457)	**<0.0001**

AD = Alzheimer's disease; DLB = dementia with Lewy bodies.

[a]P-values derived from tests comparing mean or % values between biomarker-negative and -positive subgroups. P-values < 0.05 are highlighted in bold.

[b]Study conclusion date 19 April 2019.

[c]Row highlighted in bold indicates main finding from this table.

[d]AD dementia category includes the following diagnostic subgroups containing only a single participant: Biomarker negative = CDR 0.5 in memory only; Biomarker-positive = AD dementia with other contributing aetiology, AD dementia with depression contributing, AD dementia with disturbed social comportment, AD dementia with language dysfunction, CDR 0.5 in memory only.

[e]Uncertain dementia category includes the following diagnostic subgroups: Uncertain dementia, Uncertain—possible non-AD dementia, Uncertain—questionable impairment.

time of initial biomarker assessment to death was 7.0 years (median 7.0 years; range 2.2–11.9 years) and also was not different between biomarker-negative or -positive participants. Amyloid PET imaging was used to categorize biomarker status for 37 participants, while 20 participants were categorized by CSF. 22 participants (38.6%) were biomarker positive at baseline assessment and

35 (61.4%) were biomarker negative. 68.2% of biomarker-positive participants who underwent autopsy had cognitive impairment at time of expiration compared to 48.6% of biomarker-negative participants (P = 0.1471).

Preclinical Alzheimer's disease biomarker positivity was a very strong predictor of underlying intermediate-to-high ADNC at
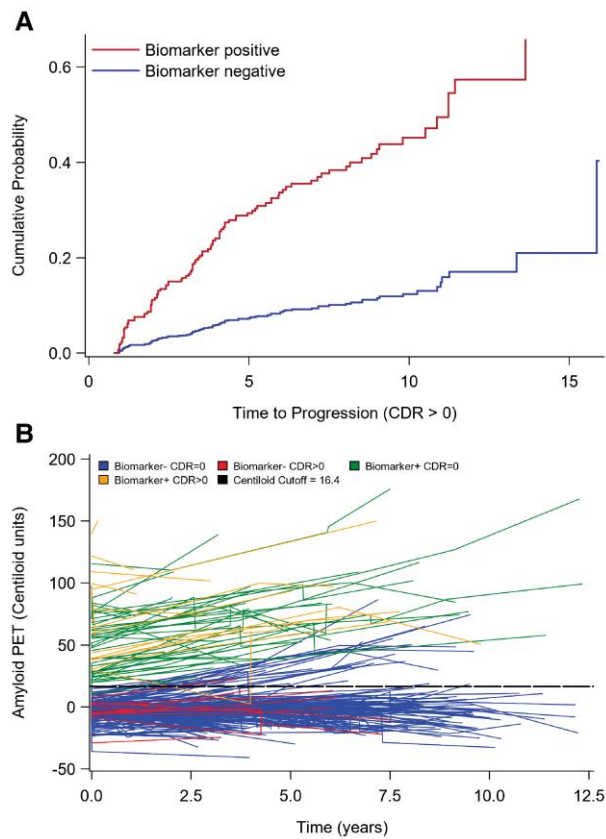
Figure 1 Survival analysis and longitudinal biomarker trends in biomarker-negative and -positive groups. (**A**) Model-based cumulative incidence function curves for probability of progression to CDR > 0 in biomarker-negative (blue; lower curve) and biomarker-positive (red; upper curve) populations. (**B**) Spaghetti plots of longitudinal amyloid PET biomarker trends in CDR progressors (CDR > 0) and non-progressors (CDR = 0). Amyloid PET PIB and AV-45 SUVR measurements were converted to Centiloid units to combine tracer data. Data were plotted and subcategorized by biomarker-positive and -negative assignment. Biomarker category assignment was based on SUVR cut-offs. The comparable converted Centiloid cut-off value (16.4) is presented for visualization but was not used in determining biomarker status. Fitted lines obtained by simple linear regression with 95% CI are also shown. Only participants with at least one additional biomarker assessment of the same modality used to define original biomarker status were included.

autopsy. 90.9% of participants with positive preclinical Alzheimer's disease biomarkers had intermediate-to-high ADNC on autopsy, whereas only 8.6% of participants negative for preclinical Alzheimer's disease biomarkers had intermediate-to-high ADNC (*P* < 0.0001). Preclinical Alzheimer's disease biomarker positivity was associated with high sensitivity (87.0%), specificity (94.1%), positive predictive value (PPV) (90.9%) and negative predictive value (NPV) (91.4%) for predicting intermediate-to-high ADNC at autopsy (Table 3). When participant cohorts categorized by either CSF or PET were evaluated separately, sensitivity, specificity, PPV and NPV remained high. Although CSF testing seemed to marginally outperform amyloid PET imaging for predicting ADNC at autopsy, this was not statistically significant when directly comparing diagnostic metrics between CSF and PET (sensitivity *P* = 0.824, specificity *P* = 0.3134, PPV *P* = 0.2622, NPV *P* = 0.971).

To evaluate the effect of preclinical biomarkers, centred age, *APOE* ε4 carrier status, sex, race, education, PRS and interval from baseline biomarker assessment to death on odds of harbouring

ADNC, a logistic regression model was employed using these covariates. The only variable significantly associated with an effect on odds of harbouring ADNC in this cohort was preclinical Alzheimer's disease biomarker positivity, which exhibited a very large effect size because so few individuals with positive biomarkers had absent or low ADNC (OR 886.15, 95% CI 23.85 >999.9, *P* = 0.0002). No other covariates demonstrated independent significant associations with diagnosis of ADNC.

In this cohort, baseline preclinical Alzheimer's disease biomarker evaluation resulted in three 'false-negative' and two 'false-positive' assignments when compared to gold-standard neuropathological assessment. To better evaluate these discordant biomarker assignments, baseline biomarker values were plotted after stratifying by baseline biomarker designation and final Alzheimer's disease neuropathological outcome (Fig. 2). In all cases, those participants with false-positive or -negative baseline biomarker assignments had baseline biomarker values that were closer to the threshold value compared to those participants who were correctly assigned.

To investigate this further, individual Alzheimer's disease neuropathological scores were evaluated in these discordant cases. Among the three biomarker-negative participants with underlying intermediate-high ADNC (false negatives), two cases were clearly discordant with substantial amyloid (Thal phase 4, CERAD score 3) and tau (Braak stage V) burden qualifying for a high ADNC designation despite negative biomarker assignment. The third 'false-negative' case had moderate amyloid (Thal phase 3, CERAD 1) and tau (Braak stage III) burden, qualifying for intermediate ADNC designation. There were two biomarker-positive participants lacking intermediate-high ADNC (false positives) and each had moderate amyloid burden (Thal phase 3–4, CERAD score 0) and low tau burden (Braak stage I-II), qualifying for low ADNC designation.

The association between biomarker levels and neuropathological outcomes was further explored by using univariate logistical regression models with baseline single biomarker levels as independent variable and probability of intermediate-high ADNC as dependent variable. OR from these analyses are summarized in Table 4. These models demonstrated that increased CSF $A\beta_{42}$ levels were associated with significantly lower odds of intermediate-high ADNC per unit change, whereas increased CSF p-tau181, t-tau, p-tau181/$A\beta_{42}$ and amyloid PET centiloid values were associated with significantly higher odds of intermediate-high ADNC per unit change. CSF $A\beta_{40}$ levels were not available with this specific Elecsys dataset, so $A\beta_{42}/A\beta_{40}$ ratios could not be calculated. Probability curves produced by these models are shown in Fig. 3A. Similarly, the effect of baseline single biomarker levels on the probability of having more advanced Braak stage and Thal phase pathology at autopsy was assessed using univariate ordinal logistic regression models. OR from these analyses are summarized in Table 5. The models similarly demonstrated that increased $A\beta_{42}$ levels were associated with significantly lower odds, whereas p-tau-181, t-tau, p-tau-181/$A\beta_{42}$ and amyloid PET centiloid values were associated with significantly higher odds of more advanced Braak stage and Thal phase pathology at autopsy per biomarker unit change. Cumulative probability curves from the ordinal logistic regression models are shown in Fig. 3B and C.

As described previously, there were a large number of biomarker-negative CDR progressors (48.6%) in this autopsy cohort but only a low percentage with underlying ADNC (8.6%). To better understand the underlying neuropathologies associated with each subcategory, the cohort was analysed separately by biomarker

Table 2 Clinical characteristics and neuropathological diagnoses of deceased participants with completed autopsy

	Total		Biomarker negative		Biomarker positive		P-value[a]
	n	Mean (SD) or %	n	Mean (SD) or %	n	Mean (SD) or %	
Age at baseline, mean (SD), years	57	78.8 (6.9)	35	79.0 (7.3)	22	78.6 (6.3)	0.8310
Age at death, mean (SD), years	57	85.5 (7.5)	34	85.9 (8.3)	19	86.3 (6.9)	0.8546
Male sex, %	25	43.9	17	48.6	8	36.4	0.3681
Race, %							
Non-Hispanic White	53	93.0	32	91.4	21	95.4	0.5619
African-American	4	7.0	3	8.6	1	4.6	0.5619
Asian	0	0	0	0	0	0	–
More than one	0	0	0	0	0	0	–
APOE ε4 carriers, %	14	24.6	4	11.4	10	45.4	**0.0036**
Education, mean (SD), years	57	16.2 (3.1)	35	15.8 (2.7)	22	16.7 (3.6)	0.3158
Interval from baseline assessment to death, mean (SD) [median], years	57	7.2 (3.3) [7.2]	35	6.9 (3.3) [7.2]	22	7.6 (3.3) [7.2]	0.4434
Interval from baseline biomarker to death, mean (SD) [median], years	57	7.0 (3.2) [7.0]	35	6.8 (3.2) [7.3]	22	7.4 (3.2) [6.9]	0.4657
Progression to CDR > 0 at expiration, %	32	56.1	17	48.6	15	68.2	0.1471
CDR at expiration, %							
0	25	43.9	18	51.4	7	31.8	0.1471
0.5	11	19.3	9	25.7	2	9.1	0.1211
1	4	7.0	3	8.6	1	4.6	0.5619
2	5	8.8	2	5.7	3	13.6	0.3030
3	12	21.0	3	8.6	9	40.9	**0.0035**
Intermediate-high ADNC[b], %	**23**	**40.3**	**3**	**8.6**	**20**	**90.9**	**<0.0001**
ADNC by severity, %							
None	7	12.3	7	20.0	0	0	**0.0251**
Low	27	47.4	25	71.4	2	9.1	**<0.0001**
Intermediate	9	15.8	1	2.9	8	36.4	**0.0007**
High	14	24.5	2	5.7	12	54.5	**<0.0001**
No. defined by CSF biomarker, %	20	35.1	12	60.0	8	40.0	0.2077
Intermediate-high ADNC, % CSF group	9	45.0	1	8.3	8	100.0	**<0.0001**
Elecsys t-tau, mean (SD), pg/ml	20	253.556 (135.095)	12	188.152 (66.145)	8	351.662 (155.978)	**0.0213**
Elecsys p-tau181, mean (SD), pg/ml	20	24.238 (15.961)	12	15.579 (5.263)	8	37.227 (18.071)	**0.0113**
Elecsys Aβ42, mean (SD), pg/ml	20	1277.65 (619.144)	12	1622.28 (552.519)	8	760.700 (227.481)	**0.0002**
p-tau181/Aβ42 Ratio, mean (SD)	20	0.0264 (0.0262)	12	0.0099 (0.0024)	8	0.0512 (0.0262)	**0.0029**
No. defined by amyloid PET biomarker, %	37	64.9	23	62.2	14	37.8	**0.0366**
Intermediate-high ADNC, % PET group	14	37.8	2	8.7	12	85.7	**<0.0001**
PIB SUVR, mean (SD)	32	1.605 (0.935)	20	1.025 (0.122)	12	2.572 (0.899)	**<0.0001**
AV45 SUVR, mean (SD)	5	1.393 (0.589)	3	1.109 (0.196)	2	1.818 (0.840)	0.2258

[a]P-values derived from tests comparing mean or % values between biomarker-negative and -positive subgroups. P-values < 0.05 are highlighted in bold.
[b]Defined by NIA-Regan intermediate-to-high likelihood of Alzheimer's diseaseee or NIA-AA intermediate-to-high ADNC. Row highlighted in bold indicates main finding from this table.

Table 3 Diagnostic performance of preclinical Alzheimer's disease biomarkers for predicting intermediate-high ADNC at autopsy

	n	Sensitivity	95% CI	Specificity	95% CI	PPV	95% CI	NPV	95% CI
Total, %	57	87.0	73.2–100.0	94.1	86.2–100.0	90.9	78.9–100.0	91.4	82.1–100.0
CSF p-tau181/Aβ42, %	20	88.9	68.4–100.0	100.0	100.0–100.0	100.0	100.0–100.0	91.7	76.0–100.0
Amyloid PET, %	37	85.7	67.4–100.0	91.3	79.8–100.0	85.7	67.4–100.0	91.3	79.8–100.0

status and CDR progressor status (Supplementary Table 3). Cerebrovascular disease was highly prevalent across all subcategories, with only two participants lacking any vascular pathology. When vascular disease was limited to pathology more likely to influence cognition (infarcts, haemorrhages, severe arteriolosclerosis or severe white matter changes), pathological changes remained prevalent (~40% across the whole cohort) but were significantly less prevalent amongst biomarker-positive CDR non-progressors (n = 0; 0%) as compared to biomarker-positive CDR progressors (n = 9; 60%). Primary tau pathologies, which include typical

frontotemporal lobar degeneration pathologies as well as common age-related primary tauopathies such as argyrophilic grain disease, primary age-related tauopathy and ageing-related tau astrogliopathy, were commonly observed across the cohort (43.8%) but noticeably absent in any biomarker-positive CDR non-progressor. Overall, 12 of 17 biomarker-negative CDR progressors had neuropathological diagnoses expected to influence cognition, including discordantly assigned ADNC, cerebral Lewy bodies, hippocampal sclerosis, frontotemporal lobar degeneration-TDP43 and prion disease. In comparison, only 3 of 18 biomarker-negative CDR non-progressors
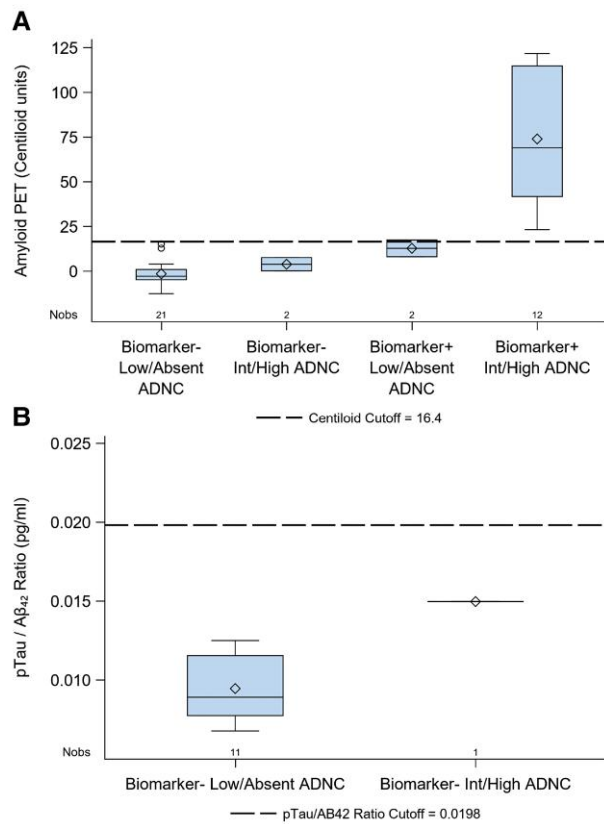
Figure 2 Baseline biomarker levels in participants stratified by biomarker status and final Alzheimer's disease neuropathological diagnosis. (**A**) Boxplots of baseline amyloid PET biomarker levels stratified by biomarker positivity and low/absent or intermediate/high ADNC at autopsy. Amyloid PET PIB and AV-45 SUVR measurements were converted to Centiloid units to combine tracer data. Biomarker category assignment was based on SUVR cut-offs. The comparable converted Centiloid cut-off value (16.4) is presented for ease of visualization but was not used as a threshold for biomarker positivity. (**B**) Boxplots of baseline CSF p-tau181/Aβ_{42} levels for biomarker-negative participants stratified by low/absent or intermediate/high ADNC at autopsy. Among the participants whose biomarker status was based on CSF, there were no biomarker-positive participants without ADNC in the autopsy cohort. So, biomarker-positive participants were excluded from this plot. In both **A** and **B**, the box size defines the interquartile range, the horizontal line indicates the median, the diamond indicates the mean, the whiskers indicate maximum and minimum range of data points and open circles indicate outliers.

had similar diagnoses, including cerebral Lewy bodies and medial temporal lobe TDP43 pathology.

Discussion

This prospective study evaluated clinical and neuropathological outcomes in a community-dwelling cohort of cognitively normal participants stratified by the presence or absence of preclinical Alzheimer's disease. Preclinical Alzheimer's disease biomarker positivity, as defined by either CSF p-tau181/Aβ_{42} or amyloid PET imaging in individuals with normal cognition, was associated with significantly increased risk of developing cognitive impairment over time and was highly predictive of intermediate-high ADNC at autopsy. This study adds to the base of evidence supporting the use of these biomarkers to define Alzheimer's disease in its

Table 4 Effect of baseline individual biomarkers on odds of intermediate-high ADNC present at autopsy

Biomarker	Odds ratio	95% CI	P-value[a]
Elecsys Aβ_{42}[b] (per 100 pg/ml increase)	0.513	0.326–0.807	**0.0039**
Elecsys p-tau181[b] (per 1 pg/ml increase)	1.086	1.015–1.161	**0.0161**
Elecsys t-Tau[b] (per 10 pg/ml increase)	1.077	1.007–1.152	**0.0297**
p-tau181/Aβ42 ratio[b] (per 0.001 unit increase)	1.134	1.031–1.248	**0.0099**
Centiloid[c] (per 5 unit increase)	2.248	1.219–4.145	**0.0095**

[a]P-values < 0.05 are highlighted in bold
[b]Total *n* limited to 33 of 57 due to missing CSF values.
[c]Total *n* limited to 37 of 57 due to missing amyloid PET Centiloid values.

preclinical phase by providing crucial neuropathological validation. Indeed, in this study, 20 out of 22 participants with positive preclinical Alzheimer's disease biomarkers who went to autopsy were found to have intermediate-high ADNC.

Most recent Alzheimer's disease clinical trials have added detection of either PET- or CSF-based Alzheimer's disease biomarkers as inclusion criteria to enroll only those cognitively impaired participants with anticipated ADNC. With the development of Alzheimer's disease disease-modifying therapies,[1] treatment trials are beginning to focus on prevention paradigms in preclinical Alzheimer's disease populations.[30,31] In studies focused on prevention of sporadic Alzheimer's disease, this requires screening strategies to identify possible participants. These data support the use of these modalities for screening cognitively normal individuals in the community. The recent discovery of promising plasma-based Alzheimer's disease biomarkers[63,64] should enable even broader screening of the general population. Neuropathological correlation will be required for thorough validation of plasma-based preclinical Alzheimer's disease biomarkers.

CSF- and PET-based preclinical Alzheimer's disease diagnosis in this study was associated with high sensitivity, specificity, PPV and NPV for detection of intermediate-high ADNC. Although there were a significant number of biomarker-negative participants who developed cognitive impairment prior to death, 53% of these participants had underlying non-Alzheimer's disease neuropathological diagnoses that could influence cognition, whereas only 17% had 'false-negative' ADNC.

The strong association of preclinical Alzheimer's disease biomarker positivity with subsequent development of cognitive impairment reported here is well-established and has been presented previously in a number of prior studies, both from the Washington University Knight ADRC cohort[5,8,11,14] as well as other cohorts of ageing.[12,65–68] A unique feature of this cohort is the size and duration of prospective longitudinal cognitive follow-up, with 87 participants (12.1% of total cohort) being followed for 10–15 years from time of initial biomarker assessment. In this study, though the proportion of biomarker-positive individuals with progression to CDR > 0 increased significantly with interval of follow-up, only 53.3% of biomarker-positive participants progressed when followed for 10–15 years from baseline assessment, suggesting an extended period between development of ADNC and onset of cognitive impairment. This finding is consistent with previous studies that have evaluated time to incident cognitive impairment in relation to biomarker positivity and found that a proportion of biomarker-positive participants persist beyond 15 years without progression to CDR > 0, supporting a preclinical stage of Alzheimer's disease that can extend beyond 15 years.[14]
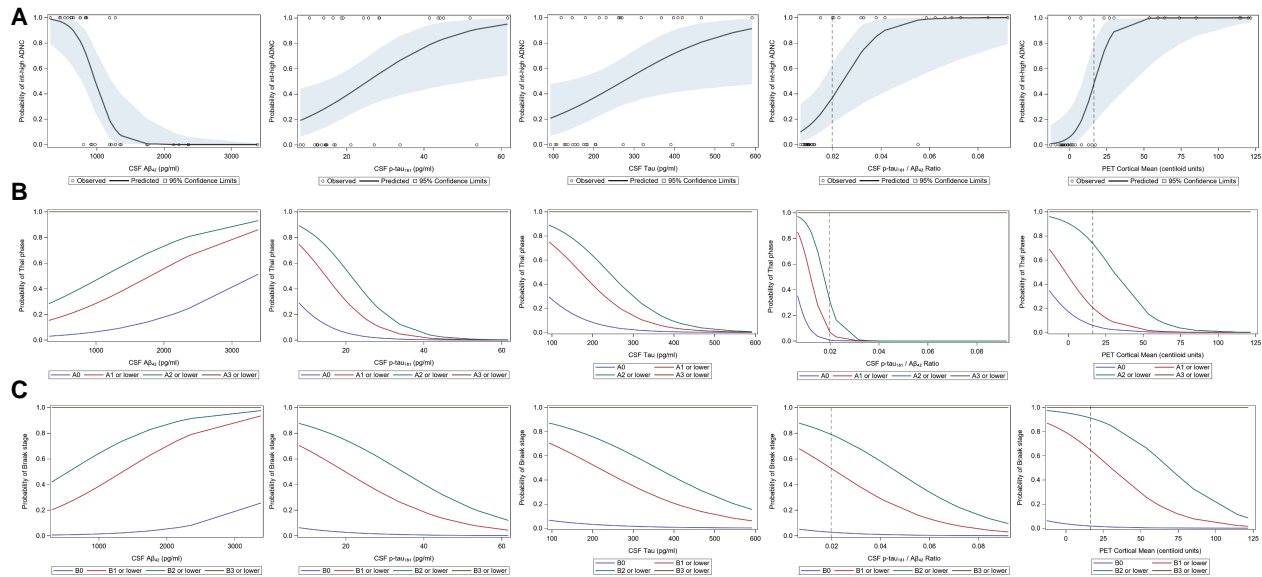
Figure 3 Probability curves of ADNC pathology as a function of baseline single biomarker values. (A) Probability curves (with shaded 95% confidence intervals) of intermediate-high ADNC at autopsy as a function of baseline levels of $A\beta_{42}$, p-tau181, t-tau, p-tau181/$A\beta_{42}$ ratio and amyloid PET Centiloid units. Probability curves are generated from univariate logistic regression models with biomarker level as independent variable and probability of intermediate-high ADNC as dependent variable. **(B)** Cumulative probability curves of Thal phase pathology at autopsy as a function of baseline levels $A\beta_{42}$, p-tau181, t-tau, p-tau181/$A\beta_{42}$ ratio and amyloid PET Centiloid units. Thal phases are grouped in this model consistent with NIA-AA criteria[62]: A0 = Thal phase 0, A1 = Thal phase 1 or 2, A2 = Thal phase 3, A3 = Thal phase 4 or 5. **(C)** Cumulative probability curves of Braak stage pathology at autopsy as a function of baseline levels $A\beta_{42}$, p-tau181, t-tau, p-tau181/$A\beta_{42}$ ratio and amyloid PET Centiloid units. Braak stages are grouped in this model consistent with NIA-AA criteria[62]: B0 = Braak stage 0, B1 = Braak stage I or II, B2 = Braak stage III or IV, B3 = Braak stage V or VI. **(B and C)** Probability curves were generated from univariate ordinal logistic regression models with biomarker level as independent variable and cumulative probability of pathologic group as dependent variable. Modelled probabilities are cumulated over lower pathology groups, such that each curve delineates the probability of a given pathologic group or lower. Vertical dashed lines highlight p-tau181/$A\beta_{42}$ ratio and Centiloid cut-off thresholds for reference.

In this study, there were two 'false-positive' and three 'false-negative' cases, as determined by discordance between biomarker status and presence or absence of intermediate-high ADNC. One possible interpretation is that these participants were falsely assigned at baseline due to the imprecision of the biomarker cut-off threshold, supported by the fact that all discordant cases had biomarker levels near the cut-off threshold. An alternative explanation is that the false-positive participants had low levels of ADNC below the threshold for intermediate-to-high ADNC designation on autopsy. Indeed, the two false-positive cases showed low ADNC at autopsy with moderate amyloid burden but insufficient tau pathology to qualify as intermediate-to-high ADNC. By analogy, the false-negative participants could have relatively modest amyloid burden at autopsy with ADNC stage driven primarily by high levels of tau pathology. However, in reality, two of the false-negative cases harboured substantial amyloid and tau pathology consistent with high ADNC, whereas the third case had moderate amyloid and tau burden consistent with intermediate ADNC. Therefore, it is also possible these false-negative participants may have developed more substantial ADNC in the interval between baseline biomarker assessment and autopsy.

Tau PET imaging was not assessed in this cohort, as its inclusion would have significantly limited cohort size. However, tau PET has been shown to accurately predict high ADNC in a terminally ill population[69] and seems to reflect underlying Alzheimer's disease tau pathology.[70,71] Although the biomarker false-positive rate was low in this study, it is likely that assessment of tau PET status in a preclinical population would further lower this false-positive rate, potentially at the expense of a higher false-negative rate.

Previous studies have demonstrated an association between Thal amyloid phase, but not Braak stage, on the level of ante-mortem PIB

PET binding.[72,73] However, these prior studies have not evaluated whether baseline Alzheimer's disease biomarker levels in cognitively normal participants influence the level of Thal phase and Braak stage pathology at autopsy. In this study, baseline individual biomarker levels (CSF $A\beta_{42}$, p-tau181, t-tau, p-tau181/$A\beta_{42}$ and amyloid PET centiloid) were all significantly associated with the probability of harbouring intermediate-high ADNC and having more or less advanced Thal phase and Braak stage pathology at autopsy

Neuropathological assessment also demonstrated frequent occurrence of non-Alzheimer's disease comorbid pathological diagnoses, although these did not appear evenly distributed across groups when stratified by progression to CDR > 0. These data indicate that among biomarker-positive CDR non-progressors, non-Alzheimer's disease neuropathological diagnoses are infrequent, whereas among biomarker-negative CDR progressors, non-Alzheimer's disease neuropathologic diagnoses more commonly underlie cognitive impairment, with some contribution from limited numbers of false-negative ADNC diagnoses.

One concern about a cross-sectional screening approach for detecting preclinical Alzheimer's disease in the general population is the possibility for progression from negative to positive biomarkers after the initial screening test and the theoretical risk for developing Alzheimer's disease dementia after a negative screening test. Indeed, in this cohort there was a surprisingly large number of biomarker-negative individuals who progressed to CDR > 0, most of which were assigned a clinical diagnosis of Alzheimer's disease dementia. In the longitudinal biomarker clinical cohort of this study, conversion from biomarker negative to positive was observed among ~10% of biomarker-negative participants. However, the rate of progression to CDR > 0 among this group was 3.5%,

Table 5 Effect of baseline individual biomarkers on cumulative odds of less advanced Braak stage or Thal phase pathology

Biomarker	Thal phase[a]			Braak stage[b]		
	Odds ratio[h]	95% CI	P-value	Odds ratio[h]	95% CI	P-value[c]
Elecsys Aβ_{42}[a] (per 100 pg/ml increase)	1.121[d]	1.010–1.245	**0.0321**	1.140[e]	1.019–1.277	**0.0227**
Elecsys p-tau181[a] (per 1 pg/ml increase)	0.854[d]	0.763–0.956	**0.0061**	0.929[e]	0.873–0.978	**0.0083**
Elecsys t-Tau[a] (per 10 pg/ml increase)	0.867[d]	0.787–0.955	**0.0040**	0.930[e]	0.877–0.986	**0.0152**
p-tau181/Aβ_{42} Ratio[a] (per 0.001 unit increase)	0.721[d]	0.536–0.873	**0.0081**	0.952[e]	0.917–0.980	**0.0033**
Centiloid[b] (per 5 unit increase)	0.799[f]	0.707–0.903	**0.0003**	0.691[g]	0.506–0.848	**0.0039**

[a]Thal phases grouped as follows for ordinal logistic regression model: phase 0 = A0, phase 1–2 = A1, phase 3 = A2, phase 4–5 = A3.
[b]Braak stages grouped as follows for ordinal logistic regression model: stage 0 = B0, stage I–II = B1, stage III–IV = B2, stage V–VI = B3.
[c]P-values < 0.05 are highlighted in bold.
[d]Total n limited to 31 of 57 due to missing CSF values or missing Thal phase.
[e]Total n limited to 33 of 57 due to missing CSF values or missing Braak stage.
[f]Total n limited to 34 of 57 due to missing Centiloid values or missing Thal phase.
[g]Total n limited to 36 of 57 due to missing Centiloid values or missing Braak stage.
[h]For the ordinal logistic regression, probabilities modelled are cumulated over lower Thal and Braak stages such that the OR reflect change in odds of being at a lower stage or phase of pathology per unit change of biomarker.

significantly less than the base rate of CDR progression in the full biomarker-negative cohort (8.4%). Therefore, in this cohort with a mean interval follow-up of ~6 years, conversion from negative to positive preclinical Alzheimer's disease biomarkers was not highly predictive of onset of cognitive impairment, and, it seems unlikely that CDR progression among biomarker-negative participants in this dataset can be attributed to rapid development of substantial Alzheimer's disease neuropathologic change. While future strategies for screening populations for preclinical Alzheimer's disease may require longitudinal screening tests performed at regular intervals, the appropriate time interval for screening will need to be determined on the basis of more in-depth longitudinal analyses of cohorts of biomarker converters.

This study has a number of strengths. First, unlike most published biomarker-autopsy validation studies, this study by design evaluated a community-based cohort of cognitively normal participants, thereby exclusively focusing on the validity of these markers in the preclinical phase of disease. Since this cohort consists of participants living in the local community and not recruited from tertiary referral memory clinics with potential referral biases, the spectrum of comorbid neuropathology observed on autopsy is less likely to be skewed compared to that observed in other studies.[22] This is supported by the near universal presence of cerebrovascular disease noted across all categories of brain specimens in the autopsy cohort, a highly prevalent diagnosis in the general population of older adults.[74]

This study also has important limitations. The participant population included individuals highly motivated to participate in research, with many agreeing to serial LP. This population is unlikely to be representative of the general community. The total number of autopsied participants with positive preclinical Alzheimer's disease biomarkers was relatively low (38.6%). This limited sample size restricts the extent of neuropathological assessment. Also, a significant proportion of participants at autopsy with positive preclinical Alzheimer's disease biomarkers had not yet developed cognitive impairment prior to autopsy (31.8%), meaning they did not meet clinical criteria for Alzheimer's disease dementia. However, this limitation would not be expected to influence the accuracy of detecting ADNC. Another limitation of this study is that it included participants defined by either CSF or amyloid PET in combination and did not limit analysis to only one method for biomarker detection. This choice was made to broaden the number of participants with longitudinal assessments and the number available for inclusion in the autopsy cohort. There is risk that discordance in

positivity between these two methods might bias the data. We tried to limit this risk by using predefined cut-offs to establish positivity that have been shown previously by Knight ADRC investigators to demonstrate high concordance between CSF and PET modalities.[28] One final limitation of this study is that it does not definitively demonstrate whether detection of preclinical Alzheimer's disease biomarkers or ADNC inevitably leads to cognitive impairment in affected participants. Although the data in this study and many others strongly support the association between detection of preclinical Alzheimer's disease biomarkers and onset of cognitive impairment, it remains an open question as to what proportion of these participants, with sufficient longitudinal follow-up, will ultimately develop cognitive impairment.

In summary, preclinical detection of Alzheimer's disease biomarkers by CSF or amyloid PET is associated with significantly elevated risk of developing cognitive impairment and is highly predictive of intermediate-to-high ADNC (considered sufficient to account for dementia) at autopsy. This study provides neuropathological validation supporting the use of these modalities for preclinical Alzheimer's disease screening. Further longitudinal follow-up will be required to establish the long-term clinical outcomes among individuals with positive preclinical Alzheimer's disease biomarkers and among biomarker converters.

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Competing interests

Supplementary material

Supplementary material is available at *Brain* online.

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