Supplementary data

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Supplementary Methods

Ascertainment of various types of dementia

Participants were screened for dementia at baseline and subsequent center visits with the Mini-Mental State Examination and the Geriatric Mental Schedule organic level [1,2]. Those with a Mini-Mental State Examination score < 26 or Geriatric Mental Schedule score > 0 underwent further investigation and informant interview, including the Cambridge Examination for Mental Disorders of the Elderly. All participants also underwent routine cognitive assessment. In addition, the entire cohort was continuously under surveillance for dementia through electronic linkage of the study database with medical records from general practitioners and the regional institute for outpatient mental health care. This provided detailed information and was used for diagnosis of dementia and for accurately determining time of disease onset. Available information on cognitive testing and clinical neuroimaging was used when required for diagnosis of dementia subtype. A consensus panel led by a consultant neurologist established the final diagnosis according to standard criteria for dementia (DSM-III-R), Alzheimer disease (NINCDS–ADRDA) and vascular dementia (NINDS-AIREN).

AD+CVD and vascular dementia. Participants who have a clear description of (cortical) infarction in CT or MRI report retrieved from record linkage with general practitioner (GP) medical records, or (specialized) outpatient clinic letters. When more than one lacunar lesion is present or mentioned in the imaging report, this is regarded sufficient to consider the involvement of brain CVD lesions. Additionally, a clinical presentation must be present that matches the AD profile, according to official criteria. Finally, there should be no indication for a direct time relationship (>3 months) between the symptoms of the vascular brain lesions and the clinical onset of the dementia syndrome, then instead consider the diagnosis vascular dementia (VaD).

Parkinson disease. Participants were screened in the baseline and follow-up examinations for cardinal signs of parkinsonism (i.e., resting tremor, cogwheel rigidity, hypo- or bradykinesia, or impaired postural reflexes). Persons with at least one sign present are examined with the Unified Parkinson's Disease Rating Scale (UPDRS) and a further neurologic exam. Parkinson's Disease is diagnosed if two or more cardinal signs are present in a subject not taking antiparkinsonian drugs (ATC code N04), or if at least one sign has improved through medication, and when all causes of secondary

parkinsonism (dementia, use of neuroleptics, cerebrovascular disease, multiple system atrophy, or progressive supranuclear palsy) can be excluded.

Possible Alzheimer disease. Participants with a progressive, serious cognitive defect without other identifiable causes of potential (reversible) cognitive impairment, such as delirium, but with no objective test results in summary data that unanimously confirm AD (e.g., the lack of information on a formal neuropsychological assessment). Participants with possible AD diagnoses are continuously monitored to gather more follow-up data. As such, these cases can be re-coded over time based on newly gathered information which enables updating of the probability of the syndrome diagnosis to 'probable' and to additionally adjust the underlying disease subtype of the dementia syndrome.

Undetermined. Undetermined subtype of dementia is assigned when very little information is present, for instance only a brief GP report, but with clear indications of a severe cognitive defect that progresses over time and influences activities in daily living. For these cases, additional data might be gathered in follow-up to update the probability of the diagnosis and to improve dementia subtyping.

Other. Other (rare) disease subtypes of a dementia syndrome beyond the above-mentioned common dementia types include Lewy body dementia, Creutzfeld Jacob, and Frontotemporal dementia.

Ascertainment of mild cognitive impairment (MCI)

MCI was defined using the following criteria: 1) presence of subjective cognitive complaints, 2) presence of objective cognitive impairment and 3) absence of dementia for Rotterdam Study participants aged 60 years or more using official criteria, which has been described in detail elsewhere [3]. Subjective cognitive complaints were evaluated by interview. This interview included three questions on memory (difficulty remembering, forgetting what one had planned to do, and difficulty finding words), and three questions on everyday functioning (difficulty managing finances, problems using a telephone, and difficulty getting dressed). Subjective cognitive complaints were scored positive when a subject answered "yes" to at least one of these questions. Objective cognitive impairment derived from a cognitive test battery comprising letter-digit substitution

task, Stroop test, verbal fluency test, and 15-word verbal learning test based on Rey's recall of words. To obtain more robust measures, compound scores for various cognitive domains including memory function, information-processing speed and executive function were constructed. Compound scores for memory, information processing speed and executive function were calculated using Z-scores, and a person was classified as cognitively impaired if they scored below 1.5 SD of the age and education adjusted means of the study population. For MCI subtypes, Amnestic MCI was defined as persons with MCI who had an impaired test score on memory function (irrespective of other domains). Non-amnestic MCI was defined as persons with MCI having normal memory function, but an impaired test score on executive function or information-processing speed.

Confounders and risk factors

Smoking habits, use of medications and information on level of education were assessed during the home interview using a computerized questionnaire. Highest level of education was categorized in 4 groups: completed primary education, lower vocational training or general education, intermediate vocational training or intermediate and higher general education, and higher vocational training, college or university. Smoking habits was categorized as current, former and never smokers.

Clinical measurements were collected during the regular visit at the study center. Body mass index was calculated as weight in kilograms per height in meters squared. Concentrations of serum total cholesterol and high-density lipoprotein cholesterol were determined by using an automated enzymatic procedure (Boehringer Mannheim System, Mannheim, Germany). Blood pressure was measured twice at the right arm in sitting position at the research center and the average of 2 blood pressure readings was used. Diabetes mellitus type 2 was diagnosed as fasting blood glucose $\geq$ 7.00 mmol/l or the use of anti-diabetic medication was evaluated by interview and pharmacy records.[4] Cognitive score was assessed using the MMSE.[5]

Apolipoprotein E (*APOE*) genotype was assessed on coded DNA samples using polymerase chain reaction without knowledge of the dementia diagnosis. The *APOE* genotype was categorized to 0, 1 or 2 apolipoprotein E4 (*APOE*-4) alleles.

Coronary heart disease (CHD) was defined as a fatal or nonfatal myocardial infarction, a surgical or percutaneous coronary revascularization procedure (as a proxy for unstable or incapacitating angina), or death due to CHD.[6] Stroke was defined according to the World Health Organization criteria.[7]

History of stroke and CHD at entry into the study was assessed through interview and verified in medical records. Putative incident strokes and CHD get identified through the linkage of the study database with files from general practitioners, the municipality, and nursing home physicians' files, after which additional information (including brain imaging) is collected from hospital records.

Information on vital status was additionally obtained from the central registry of the municipality of the city of Rotterdam. Follow-up was complete until January 1, 2016.

Statistical analysis

Total-Tau, NfL, Aβ40 and Aβ42 plasma levels (pg/ml) have been log transformed based on the result of a Box Cox screening on the optimal transformation, both on the raw data and on data adjusted for the potential impact of age, gender and ApoE genotype. The optimal Lambda values were -0.14, -0.41, 0.10 and 0.31 for Tau, NfL, Aβ40 and Aβ42, respectively (-0.07, 0.11, 0.42 and 0.34 after adjustment). This indicates that a log transformation would be a reasonable transformation covering all four variables (and the one ratio) to obtain sufficient normality. The 2-base log scale was used to facilitate the interpretation of the results of the statistical evaluation in terms of impact of doubling steps of the AD related proteins. To account for potential non-normalities, and to allow easier interpretation and comparison between markers we have also analyzed the antigens categorized into 4 equally sized groups using quartiles.

Fig. S1. Flow diagram of the number of participants included in this study

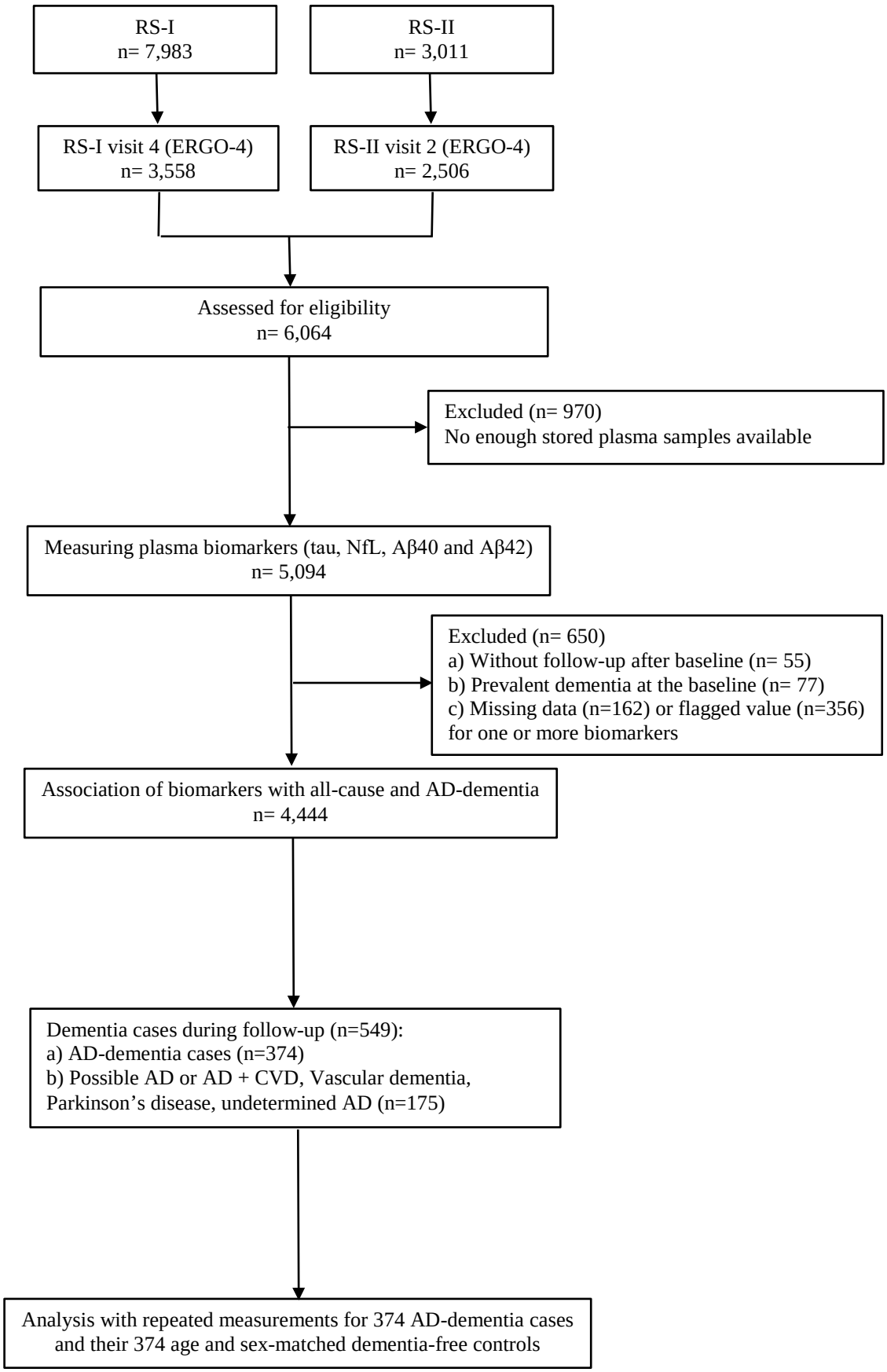
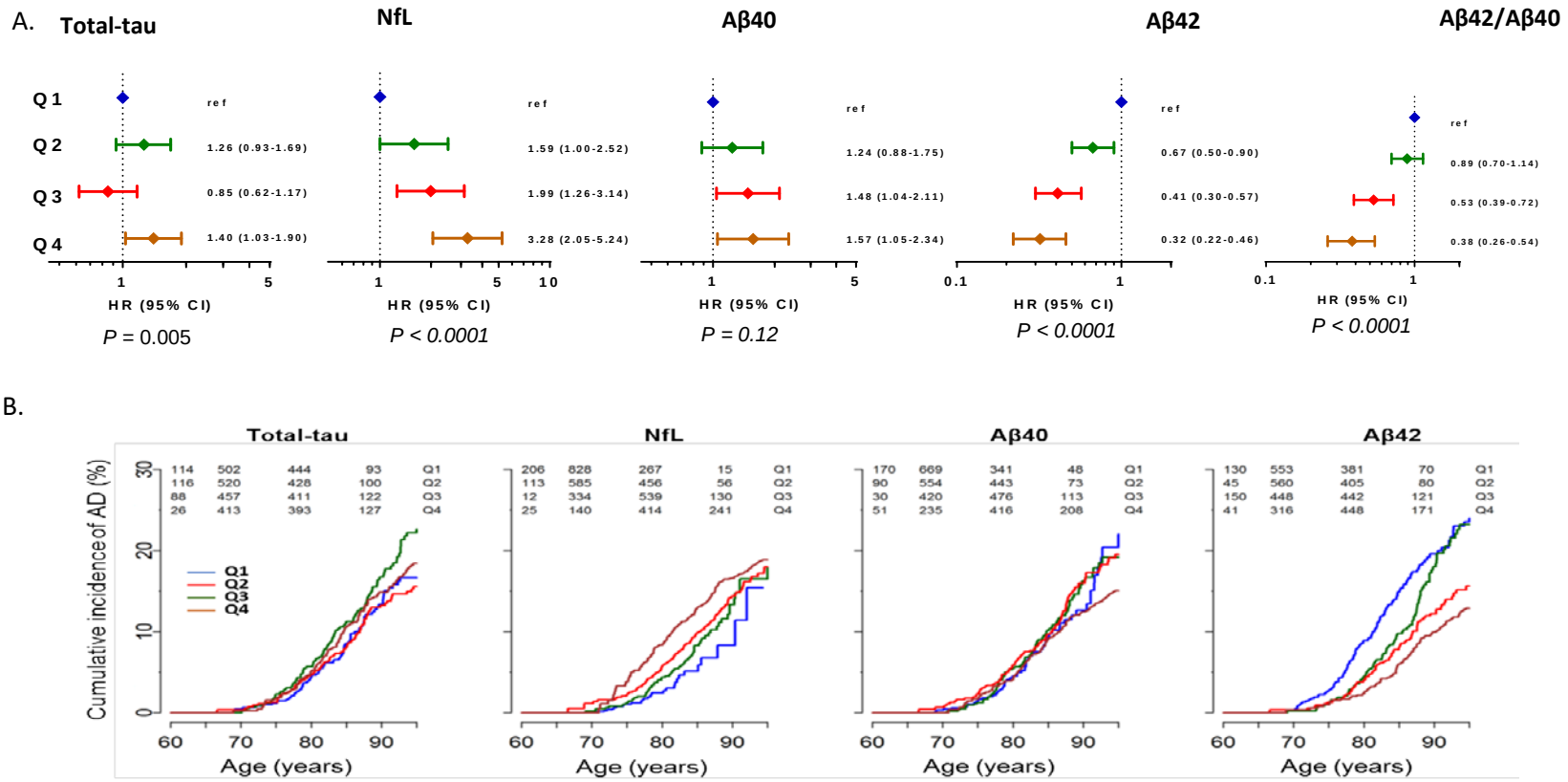


Fig. S2. Associations of plasma total-tau, NfL, Aβ40 and Aβ42 levels and Aβ42/Aβ40 ratio with AD-dementia



A) Forest plots showing hazard ratios for AD-dementia and 95%CI per quartile of plasma levels (pg/ml) of total-tau, NfL, Aβ40, Aβ42, and Aβ42/Aβ40 ratio, with the lowest quartile (Q1) as reference group. Hazard ratios were obtained with the Cox proportional hazard model adjusted for age, gender, assay batch number, systolic blood pressure, total and HDL cholesterol, smoking status, highest level of education, body mass index, *APOE-ε4* status, history of diabetes, stroke and coronary heart disease. **B**) Cause-specific incidence curves showing the incidence of AD-dementia with current age for total-tau (p-value for test for equality of the cause-specific cumulative incidence curve between the 4 groups: Total-tau (p=0.03)), NfL (p=0.009), Aβ40 (p=0.003), and Aβ42 (p<0.0001). Out of 4444 participants, 374 individuals had a diagnosis of AD-dementia, 175 had dementia other than Alzheimer's and 1229 individuals had death as a competing event. The remaining 2666 individuals were censored.

Fig. S3. Trajectories of the tau (A), Aβ40 (B) and Aβ40/Aβ42 ratio (C) on a time to AD-dementia diagnosis/index-date scale

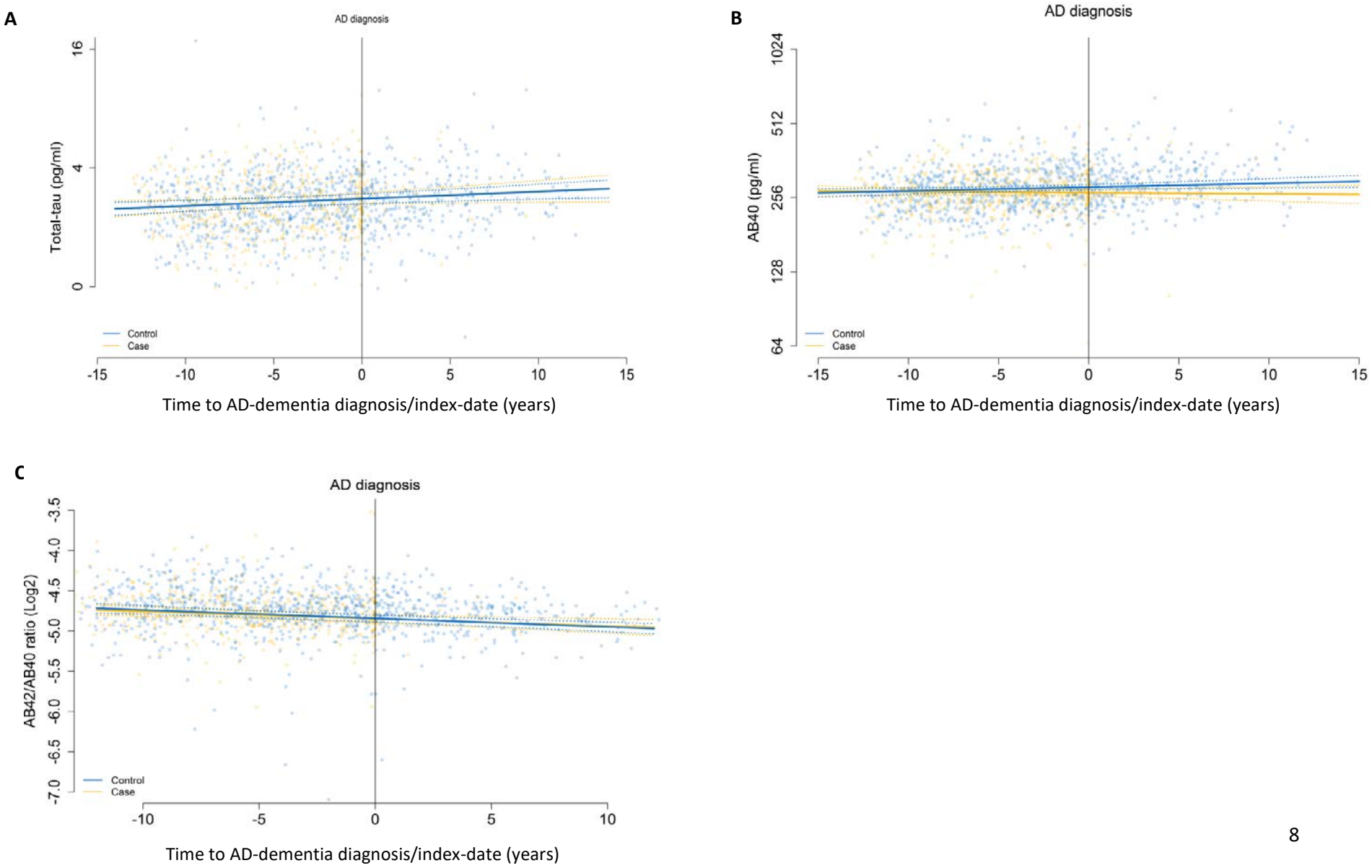
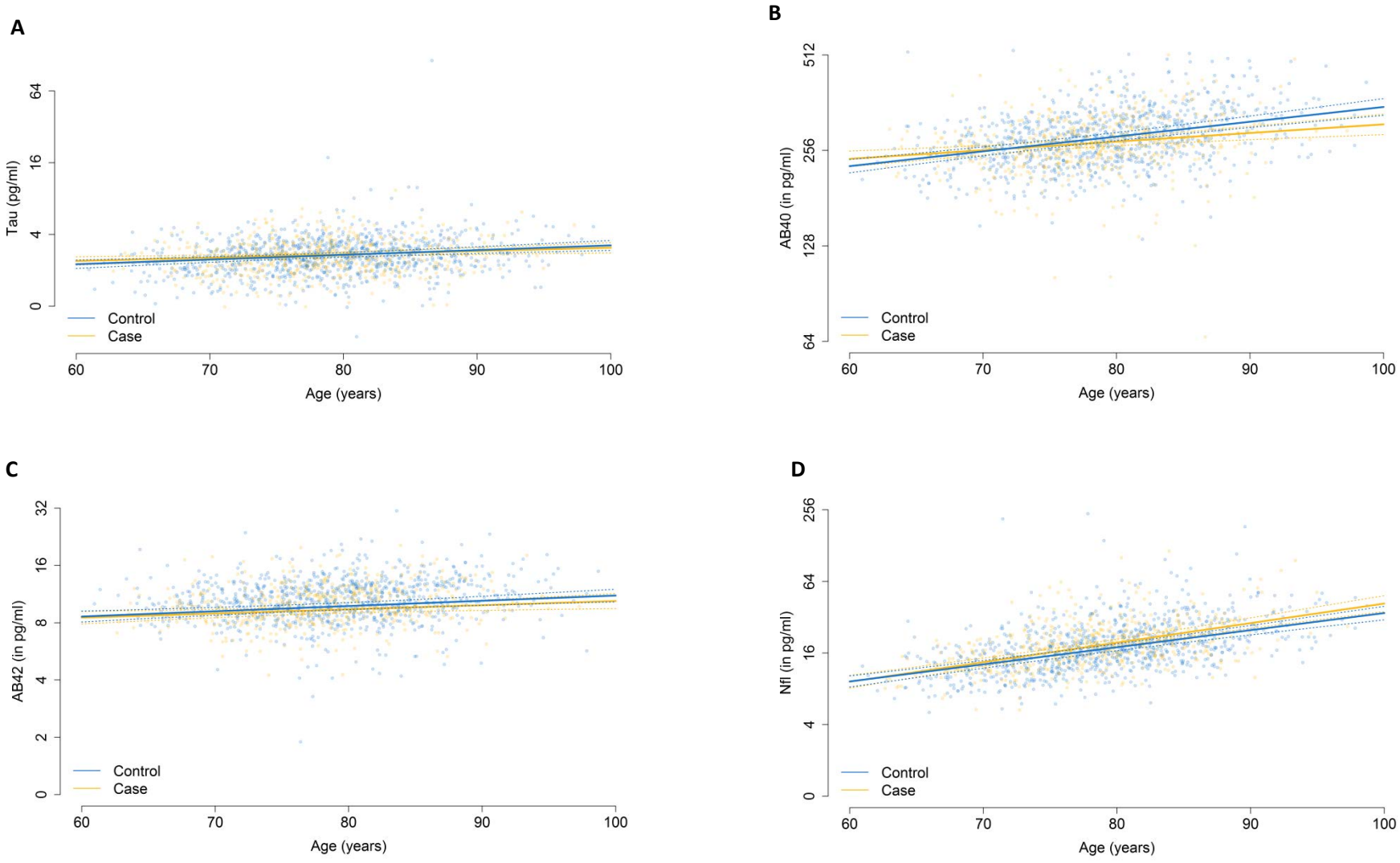


Fig. S4. Trajectories of the total-tau (A), Aβ40 (B), Aβ42 (C), and NfL (D) levels and Aβ42/Aβ40 ratio (E) in AD-dementia cases and dementia-free controls on an age scale



E

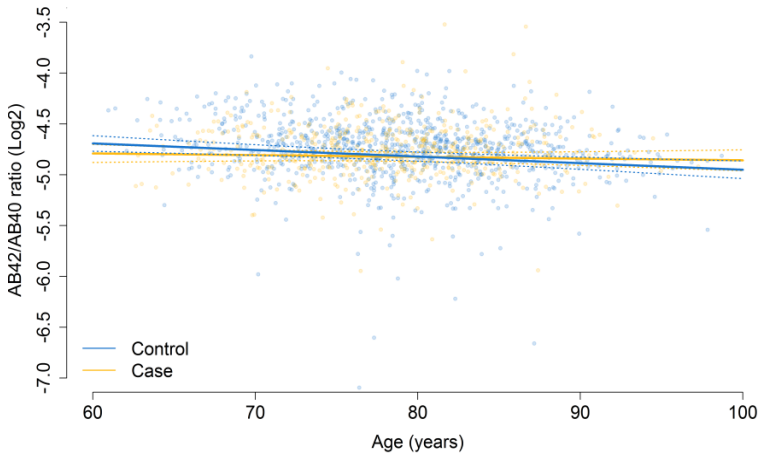

AB42/AB40 ratio (Log2)
-3.5
-4.0
-4.5
-5.0
-5.5
-6.0
-6.5
-7.0
Control
Case
60
70
80
90
100
Age (years)

Table S1. Baseline Characteristics of the fourth visit of RS-I and the second visit of RS-II

Variable	Stat	RS-I visit 4	RS-II visit 2	Combined RS-I-4 & RS-II-2	*p*-value*** RS-I vs RS-II
Total Population	N (%)	2518 (56.7)	1926 (43.3)	4444 (100)	
Age	Avg (sd)	75.2 (6)	67.7 (7)	71.9 (7.5)	<0.0001
Gender: Women	N (%)	1465 (58.2)	1090 (56.6)	2555 (57.5)	0.29
Cohort: RS-I	N (%)	2518 (100)	0 (0)	2518 (56.7)	
Cohort: RS-II	N (%)	0 (0)	1926 (100)	1926 (43.3)	
MMSE	Avg (sd)	27.8 (2)	27.8 (1.9)	27.8 (2)	0.49
<=20	N (%)	22 (0.9)	8 (0.4)	30 (0.7)	
21-25	N (%)	217 (8.6)	177 (9.2)	394 (8.9)	
26-29	N (%)	1918 (76.2)	1410 (73.2)	3328 (74.9)	
30	N (%)	357 (14.2)	295 (15.3)	652 (14.7)	
Missing	N (%)	4 (0.2)	36 (1.9)	40 (0.9)	
MCI**	N (%)	253 (10.8)	166 (9.1)	419 (9.4)	0.07
Education Level					<0.0001
Primary education	N (%)	355 (14.1)	138 (7.2)	493 (11.1)	
Lower-Intermediate education level	N (%)	1079 (42.9)	852 (44.2)	1931 (43.5)	
Intermediate-higher level	N (%)	787 (31.3)	555 (28.8)	1342 (30.2)	
Higher-university education level	N (%)	281 (11.2)	333 (17.3)	614 (13.8)	
education level missing	N (%)	16 (0.6)	48 (2.5)	64 (1.4)	
ApoE4					0.36
0 alleles	N (%)	1772 (70.4)	1311 (68.1)	3083 (69.4)	
1 allele	N (%)	614 (24.4)	452 (23.5)	1066 (24)	
2 alleles	N (%)	38 (1.5)	39 (2)	77 (1.7)	
Missing	N (%)	94 (3.7)	124 (6.4)	218 (4.9)	
Smoking					0.002
Never	N (%)	749 (29.7)	557 (28.9)	1306 (29.4)	
Former	N (%)	1386 (55)	993 (51.6)	2379 (53.5)	
Current	N (%)	336 (13.3)	337 (17.5)	673 (15.1)	
Missing	N (%)	47 (1.9)	39 (2)	86 (1.9)	
BMI***	Avg (sd)	27.4 (4.1)	27.8 (4.1)	27.6 (4.1)	0.0008
<18.5	N (%)	17 (0.7)	6 (0.3)	23 (0.5)	
18.5-25.0	N (%)	680 (27)	452 (23.5)	1132 (25.5)	
25.0-30.0	N (%)	1200 (47.7)	956 (49.6)	2156 (48.5)	
30.0-35.0	N (%)	442 (17.6)	385 (20)	827 (18.6)	
>35.0	N (%)	113 (4.5)	103 (5.3)	216 (4.9)	
Missing	N (%)	66 (2.6)	24 (1.2)	90 (2)	
Total cholesterol	Avg (sd)	5.6 (1)	5.7 (1)	5.6 (1)	<0.0001
HDL cholesterol	Avg (sd)	1.4 (0.4)	1.5 (0.4)	1.5 (0.4)	0.75
Systolic BP	Avg (sd)	152.3 (21.4)	145 (19.8)	149.1 (21.1)	<0.0001
Diastolic BP	Avg (sd)	79.3 (11.2)	80.1 (10.5)	79.7 (10.9)	0.03
CHD (of 2445/1926/4371 assessed)	N (%)	290 (11.9)	141 (7.3)	431 (9.9)	<0.0001
DM (of 2343/1926/4269 assessed)	N (%)	323 (13.8)	258 (13.4)	581 (13.6)	0.71
Stroke	N (%)	126 (5)	71 (3.7)	197 (4.4)	0.03

*Continuous variables have been tested by a t-test, binary variables by a binomial test and categorical variables by Chi2 test.

**See Supplemental information subsection Ascertainment mild cognitive impairment

***Body mass index: weight in kilograms divided by the square of the height in meters. SD: standard deviation.

We investigated whether there would be differences at baseline between RS-I-4 and RS-II-2 that potentially would impact on our analysis of the association between plasma total-tau, NfL, Aβ40 and Aβ42 plasma levels and incident AD. Continuous variables were tested by a t-test, binary variables by a binomial test and categorical variables by Chi2 test. There was a significant difference in age between participants in RS-I-4 and RS-II-2, which explains largely the differences between these two cohorts for the MCI prevalence at baseline, the overall education level, smoking, BMI, total cholesterol, blood pressure, CHD and stroke. Although the absolute differences were small, they show significance as a result of the power of the study because of its size. Since all our analyses were adjusted for age we decided to use the combined RS-I-4 and RS-II-2 data.

Table S2. Missing or invalid data for the plasma biomarkers

Reason for omission	Missing values
Instrument or technical failure	279
Insufficient volume	47
CV>20% for Tau	89
CV>20% for Aβ40	21
CV>20% for Aβ42	19
CV>20% for NfL	14

From the total of 5866 samples (in the cross-sectional and trajectory analyses), plasma biomarkers levels of 5540 samples (94%) were successfully analyzed. The majority of missing samples were due to system failures (n=279), and few because of insufficient volume (n=47). Of these samples, 14 to 87 samples were flagged due to coefficient of variation (CV) higher than 20% for any of the analytes. No samples were excluded based on controls out of range, and all samples were within the range of the assays.

Table S3. Coefficient of variations (CVs) for the plasma biomarkers in two batches

A. In the first batch including 2,000 samples

	NF-L		Tau		Ab40		Ab42	
	Mean Concentration, pg/mL	CV	Mean Concentration, pg/mL	CV	Mean Concentration, pg/mL	CV	Mean Concentration, pg/mL	CV
Control 1	12.2	7%	2.29	7%	23.3	12%	3.43	5%
Control 2	250	8%	94.7	6%	426	4%	74.8	5%
Expected Ranges, C1	9.83-14.6		1.00-3.01		17.4-26.0		2.88-3.96	
Expected Ranges, C2	190-306		43.0-129		315-482		54.2-85.4	

B. In the second batch including 3,866 samples

QC	NF-L		Tau		Ab40		Ab42	
	Mean Concentration, pg/mL	CV	Mean Concentration, pg/mL	CV	Mean Concentration, pg/mL	CV	Mean Concentration, pg/mL	CV
Control 1	12.0	9%	1.20	8%	25.4	5%	5.77	7%
Control 2	3.41	10%	154	8%	153	11%	84.9	6%
Acceptance range, C1	8.59-15.4		0.601-1.80		21.3-32.0		4.47-7.50	
Acceptance range, C2	2.37-4.44		77.3-232		121-189		69.5-104	

The coefficient of variations (CVs) were calculated from the Simoa data for each of the biomarkers. For each assay plate, 2 controls were included for each analyte, the Quanterix assays provide a high and low QC control for each marker. One control was an antigen spiked in control buffer, and another was a positive plasma control pool with endogenous antigen. For each antigen control, nominal values and acceptance ranges were pre-specified. From the total of 5866 samples, 5540 samples (94%) were successfully analyzed. As shown in Table S1, the majority of missing samples were due to system failures (n=279), and few because of insufficient volume (n=47). Of these, 14 to 87 samples were flagged due to CVs higher than 20% for any of the analytes. Because of limitations in available sample volume, no re-analysis could be done in case of assay failures or samples with >20% CV. For example, if we focused on the largest group with a CV >20% (total-tau with n=89), the total-tau levels ranging from 0.3 to 4.8 pg/ml were not biased towards the low values, considering the interquartile range of this study was between 1.9 and 3.0 pg/ml.

Table S4. Correlation between age, Tau, NfL, Aβ40, Aβ42, Aβ42/Aβ40 ratio and APOE

Variable	AGE	log2Tau	log2NfL	log2Aβ40	log2Aβ42	log2Aβratio	APOE ε4*
Pearson correlation matrix							
AGE	1.000	0.095	0.588	0.375	0.172	-0.135	-0.029
log2Tau	0.095	1.000	0.144	0.241	0.128	-0.065	0.005
log2NfL	0.588	0.144	1.000	0.491	0.334	-0.046	-0.017
log2Aβ40	0.375	0.241	0.491	1.000	0.588	-0.204	-0.014
log2Aβ42	0.172	0.128	0.334	0.588	1.000	0.672	-0.113
log2Aβratio	-0.135	-0.065	-0.046	-0.204	0.672	1.000	-0.135
APOE ε4*	-0.029	0.005	-0.017	-0.014	-0.113	-0.135	1.000
P-values for correlation matrix							
AGE	_	<.0001	<.0001	<.0001	<.0001	<.0001	0.02
log2Ttau	<.0001	_	<.0001	<.0001	<.0001	<.0001	0.68
log2NfL	<.0001	<.0001	_	<.0001	<.0001	0.002	0.16
log2Aβ40	<.0001	<.0001	<.0001	_	<.0001	<.0001	0.26
log2Aβ42	<.0001	<.0001	<.0001	<.0001	_	<.0001	<.0001
log2Aβratio	<.0001	<.0001	0.002	<.0001	<.0001	_	<.0001
APOE ε4*	0.02	0.68	0.16	0.26	<.0001	<.0001	_

*for APOE ε4 Kendall's Tau. Aβratio = Aβ42/Aβ40 ratio.

Table S5. Association of plasma total-tau, NfL, Aβ40 and Aβ42 levels and the Aβ42/Aβ40 ratio with AD-dementia

Biomarker	Association with AD-dementia					
	Model I		Model II		Model III	
	Hazard Ratio (95%CI)	*p*-value (Overall)	Hazard Ratio (95%CI)	*p*-value (Overall)	Hazard Ratio (95%CI)	*p*-value (Overall)
Tau (per $\log_2$ pg/ml increase)	1.17 (0.93, 1.46)	0.18	1.16 (0.91, 1.47)	0.23	1.07 (0.84, 1.35)	0.58
NfL (per $\log_2$ pg/ml increase)	1.49 (1.27, 1.74)	<0.0001	1.65 (1.39, 1.97)	<0.0001	1.50 (1.26, 1.78)	<0.0001
Aβ40 (per $\log_2$ pg/ml increase)	0.81 (0.55, 1.17)	0.27	0.77 (0.52, 1.14)	0.19	0.78 (0.48, 1.27)	0.31
Aβ42 (per $\log_2$ pg/ml increase)	0.60 (0.47, 0.76)	<0.0001	0.64 (0.49, 0.84)	0.001	0.59 (0.43, 0.79)	0.0006
***Aβ42/Aβ40 ratio**	0.52 (0.39-0.70)	< 0.0001	0.62 (0.45, 0.86)	0.003	0.62 (0.45, 0.85)	0.003

Model I: Adjusted for age, sex and assay batch number.
Model II: Model I + additional adjustment for highest level of education, smoking status, systolic blood pressure, total and high-density lipoprotein cholesterol (HDL), body mass index (BMI), history of diabetes, stroke and coronary heart disease (CHD), and *APOE*-ε4 status.
Model III: Model II + additional adjustment for all plasma biomarkers.
*****Model III for Aβ42/Aβ40 ratio: Model II + additional adjustment only for total-tau and NfL plasma levels.

Table S6. Association of plasma biomarkers levels with AD-dementia without adjustment for assay batch

	Alzheimer disease					
	Model I†		Model II‡		Model III*	
	Hazard Ratio (95% CI)	(Overall) p-value	Hazard Ratio (95% CI)	(Overall) p-value	Hazard Ratio (95% CI)	(Overall) p-value
Tau (per log_2 pg/ml increase)	1.15 (0.93, 1.43)	0.20	1.11 (0.89, 1.39)	0.35	1.11 (0.89, 1.40)	0.36
NfL (per log_2 pg/ml increase)	1.49 (1.27, 1.74)	<0.0001	1.51 (1.27, 1.78)	<0.0001	1.68 (1.41, 2.00)	<0.0001
Aβ40 (per log_2 pg/ml increase)	0.81 (0.55, 1.17)	0.27	0.77 (0.52, 1.14)	0.20	0.75 (0.46, 1.21)	0.24
Aβ42 (per log_2 pg/ml increase)	0.60 (0.47, 0.76)	<0.0001	0.67 (0.52, 0.87)	0.002	0.62 (0.46, 0.82)	0.001
****Aβ42 /Aβ40** (per log_2 increase)	0.55 (0.41, 0.74)	<0.0001	0.66 (0.48, 0.91)	0.01	0.65 (0.47, 0.89)	0.007

† Adjusted for only one plasma biomarker and further for age, gender;

‡ Adjusted for only one plasma biomarker and further for age, gender and systolic blood pressure, total and HDL cholesterol, smoking status, highest level of education, body mass index, APOE-ε4 status, history of diabetes, stroke and coronary heart disease

* Adjusted all plasma biomarkers and further for age, gender and systolic blood pressure, total and HDL cholesterol, smoking status, highest level of education, body mass index, APOE-ε4 status, history of diabetes, stroke and coronary heart disease.

**Model III for Aβ42/Aβ40 ratio: Model II + additional adjustment only for total-tau and NfL plasma levels.

Table S7. Associations of plasma levels of biomarkers with vascular and other non-AD dementia

Biomarker	Vascular dementia (n=27)		Other non-AD dementia (n=51)	
	HR (95% CI)	***p*-value**	**HR (95% CI)**	***p*-value**
Tau	0.89 (0.41-1.95)	0.77	0.91 (0.51-1.64)	0.75
NfL	1.95 (1.19-3.20)	0.008	1.78 (1.18-2.68)	0.006
Aβ40	0.46 (0.13-1.60)	0.22	0.66 (0.24-1.82)	0.42
Aβ42	0.49 (0.21-1.16)	0.10	0.57 (0.29-1.11)	0.10

Table shows the changes of the hazard ratio for non-AD dementia subtypes per log2 (pg/ml) higher concentrations of the biomarkers.
Ascertainment methods for various types of dementia have previously been described in detail elsewhere,[21,37] and are summarized in Supplementary Information.
Model: Adjusted for gender, age and *APOE*-ε4 status at baseline.

Table S8. Association of plasma biomarkers levels with vascular and other non-AD dementia without batch adjustment

	Vascular dementia		Other non-AD dementia	
	Hazard Ratio (95% CI)	(Overall) p-value	Hazard Ratio (95% CI)	(Overall) p-value
Tau (per $\log_2$ pg/ml increase)	0.69 (0.30, 1.62)	0.40	0.79 (0.42, 1.48)	0.46
NfL (per $\log_2$ pg/ml increase)	1.83 (1.09, 13.07)	0.02	1.86 (1.19, 2.91)	0.01
Aβ40 (per $\log_2$ pg/ml increase)	0.43 (0.10, 1.79)	0.24	0.61 (0.17, 2.21)	0.46
Aβ42 (per $\log_2$ pg/ml increase)	0.64 (0.23, 1.83)	0.41	0.66 (0.30, 1.44)	0.3
***Aβ42 /Aβ40** (per $\log_2$ increase)	0.72 (0.22, 2.32)	0.58	0.66 (0.29, 1.53)	0.34

Adjusted for all plasma biomarkers and further for age, gender and systolic blood pressure, total and HDL cholesterol, smoking status, highest level of education, body mass index, APOE-ε4 status, history of diabetes, stroke and coronary heart disease.
*For Aβ42/Aβ40 ratio: Adjusted only for total-tau and NfL plasma levels and further for all covariates mentioned above.

Table S9. Association per quartile of plasma levels of total-tau, NfL, Aβ40, Aβ42 and Aβ42/Aβ40 ratio with AD-dementia without batch adjustment

	Model I†		Model II‡		Model III*	
	Hazard Ratio (95% CI)	(Overall) p-value	Hazard Ratio (95% CI)	(Overall) p-value	Hazard Ratio (95% CI)	(Overall) p-value
Tau Q1	1.00	(0.45)	1.00	(0.46)	1.00	(0.60)
Tau Q2	1.26 (0.94, 1.70)	0.12	1.25 (0.93, 1.69)	0.14	1.18 (0.88, 1.60)	0.27
Tau Q3	1.11 (0.82, 1.50)	0.49	0.86 (0.78, 1.44)	0.71	1.00 (0.73, 1.36)	0.33
Tau Q4	1.19 (0.89, 1.61)	0.25	1.16 (0.86, 1.57)	0.34	1.11 (0.81, 1.51)	0.52
NfL Q1	0.40 (0.26, 0.62)	<0.0001	0.42 (0.27, 0.65)	<0.0001	0.34 (0.22, 0.54)	<0.0001
NfL Q2	0.51 (0.37, 0.71)	<0.0001	0.52 (0.38, 0.72)	<0.0001	0.44 (0.32, 0.62)	<0.0001
NfL Q3	0.74 (0.58, 0.94)	0.02	0.76 (0.59, 0.98)	0.03	0.68 (0.53, 0.88)	0.004
NfL Q4	1.00	(<0.0001)	1.00	(<0.0001)	1.00	(<0.0001)
Aβ40 Q1	1.00	(0.78)	1.00	(0.68)	1.00	(0.35)
AB40 Q2	1.09 (0.78, 1.52)	0.61	1.06 (0.76, 1.48)	0.64	1.12 (0.80, 1.58)	0.51
Aβ40 Q3	1.17 (0.85, 1.61)	0.33	1.20 (0.87, 1.65)	0.35	1.31 (0.92, 1.85)	0.13
Aβ40 Q4	1.06 (0.77, 1.47)	0.72	1.06 (0.76, 1.48)	0.73	1.38 (0.94, 2.04)	0.10
Aβ42 Q1	1.00	(<0.0001)	1.00	(0.004)	1.00	(<0.0001)
Aβ42 Q2	0.80 (0.61, 1.06)	0.12	0.87 (0.66, 1.16)	0.34	0.73 (55, 0.98)	0.03
Aβ42 Q3	0.74 (0.56, 0.98)	<0.0001	0.83 (0.63, 1.10)	0.20	0.62 (0.46, 0.85)	0.003
Aβ42 Q4	0.49 (0.36, 0.66)	<0.0001	0.57 (0.42, 0.78)	0.0004	0.37 (0.26, 0.53)	<0.0001
Aβ42/Aβ40 Q1	1.00	(<0.0001)	1.00	(0.004)	1.00	(0.001)
Aβ42/Aβ40 Q2	0.81 (0.62, 1.04)	0.10	0.86 (0.67, 1.12)	0.26	0.84 (0.65, 1.09)	0.19
Aβ42/Aβ40 Q3	0.70 (0.54, 0.93)	0.01	0.82 (0.62, 1.08)	0.16	0.77 (0.58, 1.01)	0.06
Aβ42/Aβ40 Q4	0.43 (0.31, 0.60)	<0.0001	0.54 (0.39, 0.75)	0.0003	0.50 (0.36, 0.70)	<0.0001

† Adjusted for only one plasma biomarker and further for age, gender;

‡ Adjusted for only one plasma biomarker and further for age, gender and systolic blood pressure, total and HDL cholesterol, smoking status, highest level of education, body mass index, APOE-ε4 status, history of diabetes, stroke and coronary heart disease.

*Adjusted for all plasma biomarkers and further for age, gender and systolic blood pressure, total and HDL cholesterol, smoking status, highest level of education, body mass index, APOE-ε4 status, history of diabetes, stroke and coronary heart disease.

For Aβ42/Aβ40 ratio: Adjusted only for total-tau and NfL plasma levels and further for all covariates mentioned above.

Table S10. Changes of the hazard ratio for all-cause dementia and AD-dementia per log2 pg/ml higher NfL and Aβ42 baseline plasma levels after 1, 2.5, 5, 7.5 and 10 years

	NfL		Aβ42	
Years after baseline	**HR (95% CI)**	**p-value**	**HR (95% CI)**	**p-value**
A. All-cause dementia				
0	2.21 (1.79-2.72)	<0.0001	0.58 (0.37, 0.90)	0.0226
1	2.07 (1.72-2.48)	<0.0001	0.59 (0.40, 0.86)	0.0107
2.5	1.88 (1.60-2.19)	<0.0001	0.60 (0.43, 0.82)	0.0023
5	1.59 (1.39-1.83)	<0.0001	0.62 (0.48, 0.80)	0.0001
7.5	1.35 (1.14-1.60)	0.0005	0.64 (0.48, 0.84)	0.0012
10	1.15 (0.91-1.44)	0.2368	0.66 (0.45, 0.97)	0.0238
B. AD-dementia				
0	2.29 (1.77-2.97)	<0.0001	0.50 (0.30, 0.82)	0.007
1	2.15 (1.71-2.70)	<0.0001	0.51 (0.33, 0.81)	0.004
2.5	1.94 (1.60-2.36)	<0.0001	0.54 (0.37, 0.79)	0.001
5	1.64 (1.38-1.95)	<0.0001	0.60 (0.44, 0.81)	0.0009
7.5	1.39 (1.13-1.72)	0.002	0.65 (0.46, 0.92)	0.02
10	1.18 (0.88-1.57)	0.26	0.71 (0.44, 1.15)	0.16

Table S11. Absolute numbers of participants and incident AD-dementia cases (%) of each combination of quartile group of NfL and Aβ42 plasma levels

	Q1 NfL			Q2 NfL			Q3 NfL			Q4 NfL		
	N	AD			AD			AD			AD	
	total	n	%	N	n	%	N	n	%	N	n	%
Q1 Aβ42	107	1	0.9	196	6	3.0	291	19	6.5	515	62	12.0
Q3 Aβ42	248	1	0.4	287	9	3.1	310	25	8.1	266	43	16.2
Q2 Aβ42	342	6	1.8	315	20	6.3	259	30	11.6	195	39	20.0
Q4 Aβ42	414	19	4.6	311	30	9.6	251	36	14.3	135	28	20.7

References

1. Licher S, Darweesh SKL, Wolters FJ, et al. Lifetime risk of common neurological diseases in the elderly population. J Neurol Neurosurg Psychiatry 2018.
2. de Bruijn RF, Bos MJ, Portegies ML, et al. The potential for prevention of dementia across two decades: the prospective, population-based Rotterdam Study. BMC Med 2015;13:132.
3. de Bruijn RF, Akoudad S, Cremers LG, et al. Determinants, MRI correlates, and prognosis of mild cognitive impairment: the Rotterdam Study. J Alzheimers Dis 2014;42 Suppl 3:S239-49.
4. Alberti KG, Zimmet PZ. Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation. Diabetic medicine : a journal of the British Diabetic Association 1998;15:539-53.
5. Folstein MF, Folstein SE, McHugh PR. "Mini-mental state". A practical method for grading the cognitive state of patients for the clinician. Journal of psychiatric research 1975;12:189-98.
6. Leening MJ, Kavousi M, Heeringa J, et al. Methods of data collection and definitions of cardiac outcomes in the Rotterdam Study. Eur J Epidemiol 2012;27:173-85.
7. Hatano S. Experience from a multicentre stroke register: a preliminary report. Bulletin of the World Health Organization 1976;54:541-53.