Plasma Amyloid, Inflammatory Markers, and Risk of Dementia in the Ginkgo Evaluation of Memory Study

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Abstract

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Most individuals with dementia have mixed neurodegenerative and vascular pathologies that may interact to accelerate cognitive decline. Prior studies have shown that two vascular inflammatory markers (pentraxin 3 and serum amyloid P) and plasma amyloid are associated with dementia, though results have been inconsistent. Using weighted Cox proportional hazards regression, we evaluated the joint associations of these inflammatory markers and plasma amyloid (Aβ1-42 and Aβ1-42/Aβ1-40 ratio) with risk of all-cause dementia. All four biomarkers were significantly associated with risk of dementia, and a composite of biomarker z-scores showed substantially higher risk for those with high levels of both inflammation and amyloid. Results for inflammatory markers and the composite z-score were stronger for individuals with MCI at baseline. Assessing inflammatory biomarkers in combination with plasma amyloid may lead to better prediction of symptomatic dementia onset than plasma amyloid alone.

Introduction

Dementia is currently the sixth-leading cause of death in the United States, and the annual number of new cases of dementia is projected to double by 2050.[1] Therapeutic interventions for individuals with dementia have so far been ineffective, and many believe this is due to targeting pathology too late in disease development.[1] For drug therapies to succeed, we must be able to determine when individuals are in the earliest asymptomatic stages of disease. Alzheimer's neuropathology can be identified *in vivo* using CSF biomarkers and PET imaging,[2,3] but these methods are invasive and costly. Blood-based biomarkers would enable alternatives that can be applied widely in public health practice. In addition, successfully identifying and treating those most likely to develop symptoms, regardless of their level of neuropathology, will result in the greatest reduction in dementia cases. Blood biomarkers may play an important role in identifying those most likely to develop symptoms.

Levels of plasma amyloid (Aβ42 and Aβ42/Aβ40 ratio) are weakly but significantly correlated with CSF levels.[4,5] Recently, three large population-based studies have shown that plasma amyloid is significantly associated with incident dementia.[6–8] Prior studies using data from the Ginkgo Evaluation of Memory Study (GEMS) indicate that two vascular inflammatory biomarkers in the pentraxin family, pentraxin 3 (PTX3) and serum amyloid P (SAP), may be associated with cognitive decline, but need further investigation.[9,10] Other studies of these inflammatory biomarkers and association with dementia have found mixed results.[11–14]

The prototypic long pentraxin PTX3 is an acute phase response protein produced in a variety of cell types, including vascular endothelial cells, smooth muscle cells, epithelial cells, fibroblasts, monocytes, and macrophages.[15] It is closely related to the inflammatory state of the vasculature and is highly expressed in advanced atherosclerotic lesions,[16] which are associated with

cognitive decline and dementia.[17] PTX3 may inhibit phagocytosis of damaged neurons by macrophages, which may further exacerbate neuronal cell death, providing a link between inflammation and Alzheimer's pathology.[18]

SAP is a short pentraxin primarily produced in the liver as a systemic response to inflammation. In addition to its possible association with dementia through inflammation, SAP is connected to, but distinct from, the amyloid-β protein associated with Alzheimer's disease. It binds to all types of amyloid fibrils, including amyloid-β in the brain. It then promotes amyloid plaque formation by preventing proteolysis of amyloid-β fibrils, and also induces neuronal apoptosis.[14] There is evidence it is able to cross the blood-brain barrier,[19] and decreased levels of SAP in plasma may indicate Alzheimer's pathology due to binding to amyloid-β plaques.

Since most individuals with dementia have mixed neurodegenerative and vascular pathologies that may interact to accelerate cognitive decline,[20] combining plasma amyloid and vascular inflammatory markers could lead to better prediction of onset of cognitive symptoms than amyloid alone, and may help determine the most appropriate targets for drug therapy in the preclinical stage.

In this study, we evaluated the joint associations of these two inflammatory markers (PTX3 and SAP) and plasma amyloid (Aβ1-42 and Aβ1-42/Aβ1-40 ratio) with time to all-cause dementia in GEMS, a large prospective cohort study of older adults. In this cohort, plasma was collected at baseline and dementia was evaluated at follow-up over many years, providing a valuable opportunity to study longitudinal associations of these biomarkers and incident dementia.

Methods

Study Design and Population

GEMS was a double-blind, placebo-controlled trial to evaluate the effectiveness of *Ginkgo biloba* as prevention of dementia in older adults.[21,22] Individuals aged 75 or older who were free of neurological or neurodegenerative diseases were recruited and followed for an average of 6 years. Other exclusions included taking cholinesterase inhibitors; currently taking tricyclic antidepressants, antipsychotics, or other psychotropic or central cholinergic effecting drugs; refusal to stop taking *G. biloba* or reduce vitamin E intake; having a history of bleeding disorders or thrombocytopenia, taking anticoagulants or other similar risks; or abnormal lab values or disabling medical conditions.[23] Individuals with mild cognitive impairment (MCI) at baseline were not excluded. MCI was defined as having a CDR global score of 0.5 and being in the lowest decile of Cardiovascular Health Study normative data on selected neuropsychological test scores.[23]

Participants were recruited from 2000-2002 using voter registration and other purchased mailing lists from four US communities with academic medical centers: Hagerstown, MD (Johns Hopkins University); Pittsburgh, Pennsylvania (University of Pittsburgh); Sacramento, CA (University of California-Davis); and Winston-Salem and Greensboro, NC (Wake Forest University).[23] *G. biloba* was not found to have a protective effect for any primary outcomes including mortality and dementia, and loss to follow-up was low (6.3%).[23] Therefore, this cohort provides a valuable resource for studying the etiology of dementia.

This ancillary study used a case-cohort design, including data from all 523 GEMS participants classified with incident dementia during follow-up and a random sample of 796 participants who

remained dementia-free (total n=1,319). The case-cohort design is particularly useful for follow-up studies on cohorts where new exposures are difficult or expensive to ascertain. Like nested case-control studies with risk-set sampling, the resulting odds ratios can be interpreted as relative risks, but selection of controls in the case-cohort design is simpler.

Outcome Ascertainment

We evaluated time to incident all-cause dementia over a maximum follow-up of 7.3 years. To establish cognitive status at baseline, all participants underwent a comprehensive neuropsychological test battery and were also administered the Modified Mini-Mental State Examination (3MSE), the Clinical Dementia Rating Scale (CDR), and the Alzheimer's Disease Assessment Scale-cognitive portion (ADAS-cog). Participants and their identified proxy returned for follow-up visits every six months to evaluate cognitive decline and onset of dementia. Participants were required to repeat the neuropsychological test battery if new memory or other cognitive problems were reported by the participant or their proxy; they had received a dementia diagnosis by a private physician; they were prescribed a dementia medication; or they had pre-specified declines in at least two of their 3MSE, CDR, or ADAS-cog scores. Neuropsychological test battery results were reviewed alongside all clinical assessments by an expert panel of study physicians blinded to treatment assignment. If these test results suggested dementia, they were followed by additional neurological examination and brain magnetic resonance imagining. Final classification was made by an expert consensus panel of neurologists using DSM-IV criteria. Those found to have dementia at follow-up were no longer followed for cognitive decline. For participants who died between follow-up visits, families and health care providers were contacted and medical records were reviewed to determine whether dementia had occurred after the last visit prior to death.[23]

Biomarker Exposures

Primary exposures include vascular inflammatory markers PTX3 and SAP and plasma Aβ1-42 and Aβ-42/Aβ-40, all measured at baseline. Stored blood samples collected at baseline were previously assessed for inflammatory biomarkers and plasma amyloid at the University of Vermont Laboratory for Clinical Biochemistry Research. Inflammatory biomarkers were measured using commercially available assays. PTX3 was measured using the Human Pentraxin 3/TSG-14 Immunoassay (R&D Systems, Inc.; Minneapolis, MN). SAP was measured using the single panel Milliplex MAP Human CVD2 Panel (Millipore; Billerica, MA). Inter-assay coefficients of variation ranged from 5.8-9.4% for PTX3 and 8.7-9.4% for SAP.

As described in the Honolulu Aging Study, plasma amyloid levels were measured using a sandwich ELISA initially developed by Eli Lilly and optimized by the University of Vermont Laboratory for Clinical Biochemistry Research.[24] The monoclonal antibodies 21F12 and 2G3 were used as capture antibodies for Aβ1-42 and Aβ1-40, respectively. Both assays used biotinylated 3D6 as the detection antibody. Streptavidin-horseradish peroxidase conjugate was added to provide enzyme activity using tetramethylbenzidine as a substrate. Inter-assay coefficients of variation ranged from 3.1-7.9% for Aβ1-40 and 12.0-20.0% for Aβ1-42.[24]

Potential Confounders

Potential confounders were identified *a priori*. Key covariates include demographic characteristics (age, race/ethnicity, sex, education, and clinic), presence of an APOE ε4 allele, and cardiovascular disease (CVD) risk factors (hypertension, diabetes mellitus, BMI, smoking status, alcohol use [number of drinks per week], and history of heart disease [heart attack, angina pectoris, stroke, transient ischemic attack, heart failure, atrial fibrillation, deep vein

thrombosis, coronary bypass surgery, balloon angioplasty, heart valve replacement, pacemaker implant, or defibrillator implant]). Hypertension was defined as ≥140 mm Hg systolic or ≥90 mm Hg diastolic blood pressure or use of an antihypertensive medication. History of heart disease components were collected from self-report at baseline, based on responses to the question, "Has a doctor ever told you that you had…[disease/condition]?" APOE-ε4 status was tested for participants who provided genetic consent and had sufficient deoxyribonucleic acid for analyses (n = 1,032). We did not adjust for MCI status at baseline since we believed it was on the causal pathway between exposures and outcome.

Statistical Analysis

We evaluated associations of plasma amyloid and inflammatory markers with time to dementia using weighted Cox proportional hazards regressions with robust standard errors, censoring patients at death or loss-to-follow-up. Models were weighted using Lin and Ying estimates based on guidance from Therneau and Li to account for the case-cohort structure.[25] We compared unadjusted models to models adjusted for baseline demographics, CVD risk factors, and presence of an APOE ε4 allele. As an exploratory analysis, we also stratified by MCI status at baseline.

Exposures were log-transformed and standardized to evaluate risk associated with change of one standard deviation. No participants were excluded for having extreme levels of PTX3 indicating acute infection.[26] To assess the joint association of inflammation and plasma amyloid with dementia, we assessed a composite summary z-score of the standardized inflammatory and amyloid markers (PTX3, SAP, and Aβ1-42/Aβ1-40), reverse-coding where decreased levels are associated with increased risk. We did not apply adjustments for multiple testing.

Covariate data was missing for only a small number of participants, except for APOE genotype, which was missing for 22% of participants. Participants with missing APOE status had lower dementia incidence than those with APOE status, and therefore it was not missing completely at random. Missing completely at random (MCAR) is missingness with no relation to any covariates. In contract, missing at random (MAR) is missingness with relation to covariates other than the missing variable. If missingness is related to the missing variable (i.e., APOE carriers declined genetic testing for APOE status more often than non-carriers), the data is said to be missing not at random (MNAR) and is inherently biased. MCAR data can be estimated without bias using normal statistical methods, but MAR requires a method such as imputation to remove bias. Since we believe APOE status was missing at random (due mostly to technical problems with samples), we therefore imputed missing covariates to remove bias in our estimates. We used multiple imputation with chained equations, which does not rely on the assumption that variables are normally distributed. All other model variables were used as predictors in the imputation process, plus blood pressure, MMSE score, and global CDR. Ten imputed values were generated for each subject with missing data, yielding ten complete datasets. Results of the parallel analyses using the ten imputed datasets were combined according to Rubin's rules[27] to obtain summary estimates. Data were analyzed using R version 3.5.1.

This research was considered exempt from review by the University of Washington Division of Human Subjects Institutional Review Board because all personal identifying information was removed.

Results

The 1,319 participants included in this analysis had a mean baseline age of 79 years, were primarily white (95%), and were highly educated (mean of 14 years of education; Table 1). At baseline, participants who developed dementia over the follow-up period were slightly older, more likely to be female, more likely to have a history of heart disease, and had slightly lower BMI. Unsurprisingly, APOE-ε4 carriers and those with MCI at baseline were much more likely to develop dementia. Dementia rates by clinic ranged from 35% at Pittsburgh to 47% at Johns Hopkins; differences were not explained by different rates of MCI or APOE by clinic.

Individuals who developed dementia had higher mean PTX3 levels ($p<0.001$) and lower mean SAP, Aβ1-42, and Aβ1-42/Aβ1-40 ratio ($p=0.013$ for all) at baseline (Table 2). The combined z-score of all four biomarkers was higher in participants with dementia ($p<0.001$). Baseline Aβ1-40 levels did not differ by dementia status ($p=0.627$). Similar differences in exposure levels were observed when comparing those with MCI to those who were cognitively normal at baseline.

In unadjusted models, all four biomarkers (SAP, PTX3, Aβ1-42, and Aβ1-42/Aβ1-40 ratio) were univariately associated with onset of dementia and remained significantly associated after adjustment for covariates (Table 3a). The relationship with PTX3 was the strongest, with a 19% increased risk of dementia for each standard deviation increase in log PTX3 (aHR 1.19; 95% CI: 1.05-1.35). When evaluating the summary z-score, participants in the highest quartile had an 81% higher risk of dementia compared to those in lowest quartile (aHR: 1.81; 95% CI: 1.30-2.52), and a test for trend was highly significant ($p<0.001$).

Stratifying by MCI status resulted in stronger estimates for the group with MCI at baseline (n=282) for PTX3, SAP, and the summary z-score (Table 3b). Associations between plasma

amyloid and dementia were slightly stronger for the group with normal cognition at baseline (Table 3c). Summary z-scores remained strongly associated for both MCI and cognitively normal groups. Within the MCI group, the highest quartile had a 119% higher risk of dementia compared to the lowest quartile (aHR: 2.19; 95% CI: 1.02-4.68). For cognitively normal participants, the HR comparing the highest to lowest quartile was 1.71 (95% CI: 1.14-2.56). Summary z-score tests for trend in both MCI and cognitively normal groups were statistically significant (p=0.007 for MCI, p=0.003 for cognitively normal).

Discussion

SAP**,** PTX3, Aβ1–42, and Aβ1–42/Aβ1–40 blood biomarkers are associated with incident dementia in this large cohort study of older adults. A composite of individual biomarker z-scores showed substantial increased risk of dementia for those with levels of inflammation and amyloid above the median. While the risk of dementia associated with plasma amyloid was similar for those with and without MCI at baseline, high levels of the composite z-score incorporating inflammatory markers was associated with a higher risk for those with MCI, indicating that inflammatory markers may aid in prediction of progression from MCI to dementia. Considering inflammation and amyloid biomarkers in combination may result in more accurate assessment of dementia risk than using either type of biomarker alone.

Prior studies evaluating SAP and PTX3 have shown inconsistent results. A study of 241 individuals found no differences in SAP levels between normal, MCI, and AD groups at baseline, and no associations of serum SAP with risk of progression to AD.[14] A postmortem study found that SAP levels were significantly increased in the brains of individuals with Alzheimer's dementia compared to normal controls.[11] Since SAP binds to amyloid-β in the brain, we would expect higher concentrations in the brains of individuals with AD. The mechanisms

between SAP in the brain, CSF, and blood are unclear. It's possible that plasma levels may be decreased due to increased binding in the brain. The authors also reported that asymptomatic individuals with Alzheimer's pathology had SAP levels similar to normal controls, suggesting that higher SAP levels in the brain reduces resilience to dementia.[11] This supports the use of a combined SAP/amyloid marker to evaluate risk of dementia, as those with AD pathologic burden may be at increased risk of symptoms when their SAP levels change.

A study investigating associations of plasma PTX3 with Parkinson's Disease also included AD subjects, and found no difference in PTX3 levels between individuals with AD and normal controls, although it was probably under-powered to detect this association with only 116 non-Parkinson's subjects.[12] A cross-sectional study of elderly hypertensive patients without dementia symptoms found that PTX3 was inversely associated with Mini-Mental State Examination (MMSE) score and was associated with carotid atherosclerosis,[13] providing a link between PTX3 and vascular dementia.

Our plasma amyloid results are consistent with prior studies. An analysis of Framingham Study participants found increased plasma Aβ1-42 and Aβ1-42/Aβ1-40 ratio were associated with lower risk of dementia (Aβ1-42 HR: 0.80, 95% CI: 0.71–0.90; Aβ1-42/Aβ1-40 HR: 0.86, 95% CI: 0.76–0.98).[6] Individuals in the Rotterdam Study also had increased risk of dementia per standard deviation decrease in Aβ1-42 (HR: 1.27; 95% CI: 1.02–1.58), but the risk associated with Aβ1-42/Aβ1-40 was not significant.[7] The connections between CSF and PET amyloid levels and dementia are well-established, and recent studies have confirmed the link between plasma and brain amyloid. A cross-sectional study of plasma Aβ1-42/Aβ1-40 ratio found consistent associations of plasma amyloid with CSF, FDG-PET, and amyloid-PET biomarkers.[28] Multiple studies have shown that plasma amyloid is highly correlated with amyloid-PET,[29] and combining plasma amyloid with age and APOE improves this correlation.[30]

Strengths of this study include the prospective, longitudinal design, the large sample size and low loss to follow-up, and the frequent and rigorous assessment of dementia. We acknowledge several limitations. Like many clinical trials, external generalizability may be limited because GEMS participants were not a representative sample of the population. The GEMS cohort had a much higher proportion of white, highly educated individuals than seen in the general population. Given that the cohort consisted of adults aged 75 or older without neurological disease at baseline, participants may have had better cognitive health than average, and results may not extend to those under age 75. Biomarkers were only measured at baseline, so we cannot investigate the association of dementia with changes in biomarker levels. More studies using longitudinal measures of inflammation are needed.

In summary, this analysis showed that SAP, PTX3, and amyloid blood biomarkers are associated with dementia onset, and using a combination of biomarkers leads to stronger associations. As plasma amyloid assays continue to improve, they may soon be used as screening tests to determine if most costly or invasive tests are warranted. It is likely this screening test would incorporate age and APOE into results, but prediction for symptomatic dementia may be improved by also incorporating inflammatory markers.

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Tables

Table 1. Characteristics of Study Population at Baseline by Dementia Status at Follow-up

	No Dementia	**Dementia**	**Total**
Characteristic	*Mean or N (SD or %)*	*Mean or N (SD or %)*	*Mean or N (SD or %)*
n	796	523	1,319
Age (years)	78.3 (3.1)	79.9 (3.6)	79.0 (3.4)
Gender			
Male	455 (57.2)	267 (51.1)	722 (54.7)
Female	341 (42.8)	256 (48.9)	597 (45.3)
Race			
White	763 (95.9)	491 (93.9)	1,254 (95.1)
Non-White[1]	33 (4.1)	32 (6.1)	65 (4.9)
Education (years)	14.4 (2.9)	14.2 (3.2)	14.3 (3.1)
BMI	27.5 (4.3)	26.6 (4.2)	27.1 (4.3)
Alcohol (drinks per week)	4.0 (6.8)	3.0 (6.2)	3.6 (6.6)
Smoking status			
Never smoked	316 (40.6)	222 (42.9)	538 (41.5)
Former smoker	433 (55.6)	276 (53.3)	709 (54.7)
Current smoker	30 (3.9)	20 (3.9)	50 (3.9)
Hypertension[2]	440 (55.3)	281 (53.7)	721 (54.7)
Diabetes[3]	71 (9.0)	49 (9.6)	120 (9.2)
Cystatin C (mg/L)	0.82 (0.21)	0.85 (0.22)	0.83 (0.21)
History of heart disease[4]	264 (35.1)	197 (40.5)	461 (37.2)
Clinic			
Johns Hopkins	116 (14.6)	101 (19.3)	217 (16.5)
Pittsburgh	240 (30.2)	132 (25.2)	372 (28.2)
UC-Davis	230 (28.9)	166 (31.7)	396 (30.0)
Wake Forest	210 (26.4)	124 (23.7)	334 (25.3)
MCI at baseline	83 (10.4)	199 (38.0)	282 (21.4)
APOE-ε4 carrier[6]	117 (18.3)	144 (36.6)	261 (25.3)

(1) Includes 43 African-Americans, 12 Asian, and 10 other race/ethnicity.
(2) Defined as $\geq$ 40 mgHg systolic or $\geq$ 90 mgHg diastolic blood pressure or treatment with anti-hypertensive medications.
(3) Self-reported response to question “Has a doctor ever told you that you had…[disease/condition]?”
(4) APOE genotype was completed on 1,032 participants (21.8% missing due to lack of informed consent or technical problems with sample).

Table 2. Inflammatory Biomarker and Plasma Amyloid Levels at Baseline by Dementia Status at Follow-up

		At Follow-Up			At Baseline		
	Total	**No Dementia**	**Dementia**		**Cognitively Normal**	**MCI**	
Characteristic	*Mean or N (SD or %)*	*Mean or N `(SD or %)*	*Mean or N (SD or %)*	*p-value*[1]	*Mean or N (SD or %)*	*Mean or N (SD or %)*	*p-value*[1]
n	1,319	796	523		1,037	282	
PTX3 (mean ng/mL)	1.00 (0.67)	0.94 (0.52)	1.09 (0.85)	<0.001	0.98 (0.68)	1.08 (0.64)	0.024
SAP (mean ng/mL)	44,936 (12,678)	45,636 (12,465)	43,869 (12,935)	0.013	45,192 (12,361)	43,996 (13,761)	0.160
Aβ1-42 (mean pg/mL)	14.98 (24.86)	16.38 (29.25)	12.86 (15.85)	0.013	15.30 (27.30)	13.83 (12.36)	0.382
Aβ1-40 (mean pg/mL)	191.82 (92.68)	192.83 (106.27)	190.29 (67.04)	0.627	192.05 (100.71)	190.99 (53.88)	0.866
Aβ1-42/Aβ1-40	0.07 (0.09)	0.08 (0.10)	0.07 (0.08)	0.013	0.08 (0.10)	0.07 (0.07)	0.461
Composite z-score quartile[2]							
Q1	323 (25.0)	218 (28.1)	105 (20.4)		265 (26.2)	58 (20.9)	
Q2	322 (25.0)	215 (27.7)	107 (20.8)		260 (25.7)	62 (22.3)	
Q3	322 (25.0)	174 (22.5)	148 (28.7)		250 (24.7)	72 (25.0)	
Q4	323 (25.0)	168 (21.7)	155 (30.1)		237 (23.4)	86 (30.9)	
Test for trend				<0.001			0.036

(1) Based on t-tests for continuous and chi-square tests for categorical variables.
(2) Composite z-score of PTX3, SAP, and Aβ1-42/Aβ1-40 ratio

Table 3a. Associations of Inflammatory Biomarkers and Plasma Amyloid with Dementia (All Subjects)

		Unadjusted		Adjusted[1]	
	n	**Hazard Ratio (95% CI)**	**p-value**	**Hazard Ratio (95% CI)**	**p-value**
PTX3	1318	1.20 (1.08-1.34)	<0.001	1.19 (1.05-1.35)	0.005
SAP	1313	0.88 (0.80-0.97)	0.009	0.88 (0.79-0.99)	0.036
Aβ1-42/ Aβ1-40	1297	0.85 (0.77-0.94)	<0.001	0.82 (0.73-0.92)	<0.001
Aβ1-42	1297	0.86 (0.78-0.94)	0.002	0.82 (0.72-0.92)	<0.001
Composite z-score quartile[2]					
Q1	323	1.00 (reference)		1.00 (reference)	
Q2	322	1.09 (0.82-1.45)	0.551	0.95 (0.69-1.32)	0.773
Q3	322	1.60 (1.21-2.11)	0.001	1.48 (1.07-2.06)	0.018
Q4	323	1.76 (1.34-2.32)	<0.001	1.81 (1.30-2.52)	<0.001
Test for trend			<0.001		<0.001

(1) Adjusted for age, race/ethnicity, sex, education, clinic, BMI, smoking status, alcohol consumption, hypertension, diabetes, history of heart disease, and presence of at least one copy of APOE-ε4 allele.
(2) Composite z-score of PTX3, SAP, and Aβ1-42/Aβ1-40 ratio

Table 3b. Associations of Inflammatory Biomarkers and Plasma Amyloid with Dementia (Subjects with MCI at Baseline)

		Unadjusted		Adjusted[1]	
	n	**Hazard Ratio (95% CI)**	**p-value**	**Hazard Ratio (95% CI)**	**p-value**
PTX3	282	1.32 (1.08-1.62)	0.007	1.35 (1.03-1.77)	0.028
SAP	281	0.88 (0.73-1.06)	0.180	0.85 (0.67-1.07)	0.172
Aβ1-42/Aβ1-40	279	0.85 (0.68-1.07)	0.157	0.83 (0.65-1.06)	0.142
Aβ1-42	279	0.86 (0.69-1.07)	0.174	0.83 (0.65-1.06)	0.134
Composite z-score quartile[2]					
Q1	68	1.00 (reference)		1.00 (reference)	
Q2	55	0.98 (0.55-1.76)	0.956	0.99 (0.48-2.04)	0.978
Q3	74	1.78 (0.99-3.20)	0.055	2.42 (1.09-5.40)	0.031
Q4	81	1.68 (0.97-2.89)	0.062	2.19 (1.02-4.68)	0.044
Test for trend			0.016		0.007

(1) Adjusted for age, race/ethnicity, sex, education, clinic, BMI, smoking status, alcohol consumption, hypertension, diabetes, history of heart disease, and presence of at least one copy of APOE-ε4 allele.
(2) Composite z-score of PTX3, SAP, and Aβ1-42/Aβ1-40 ratio

Table 3c. Associations of Inflammatory Biomarkers and Plasma Amyloid with Dementia (Subjects with Normal Cognition at Baseline)

		Unadjusted		Adjusted[1]	
	n	**Hazard Ratio (95% CI)**	**p-value**	**Hazard Ratio (95% CI)**	**p-value**
PTX3	1,036	1.13 (1.00-1.29)	0.054	1.12 (0.96-1.30)	0.146
SAP	1,032	0.91 (0.82-1.02)	0.114	0.91 (0.79-1.04)	0.165
Aβ1-42/Aβ1-40	1,018	0.82 (0.74-0.92)	<0.001	0.77 (0.67-0.89)	<0.001
Aβ1-42	1,018	0.83 (0.74-0.93)	<0.001	0.77 (0.67-0.88)	<0.001
Composite z-score quartile[2]					
Q1	255	1.00 (reference)		1.00 (reference)	
Q2	267	1.15 (0.81-1.62)	0.429	0.98 (0.66-1.46)	0.926
Q3	248	1.51 (1.08-2.12)	0.017	1.39 (0.94-2.07)	0.103
Q4	242	1.62 (1.16-2.27)	0.005	1.71 (1.14-2.56)	0.009
Test for trend			0.001		0.003

(1) Adjusted for age, race/ethnicity, sex, education, clinic, BMI, smoking status, alcohol consumption, hypertension, diabetes, history of heart disease, and presence of at least one copy of APOE-ε4 allele.
(2) Composite z-score of PTX3, SAP, and Aβ1-42/Aβ1-40 ratio.