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Inflammatory biomarkers are associated with total brain volume:

The Framingham Heart Study

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Abstract

Background—Systemic inflammation is associated with ischemia and Alzheimer disease (AD). We hypothesized that inflammatory biomarkers would be associated with neuroimaging markers of ischemia (i.e., white matter hyperintensities [WMH]) and AD (i.e., total brain volume [TCB]).

Methods—MRI WMH and TCB were quantified on 1,926 Framingham Offspring participants free from clinical stroke, TIA, or dementia (mean age 60 ± 9 years; range 35 to 85 years; 54% women) who underwent measurement of a circulating inflammatory marker panel, including CD40 ligand, C-reactive protein, interleukin-6 (IL-6), soluble intracellular adhesion molecule-1, monocyte chemoattractant protein-1, myeloperoxidase, osteoprotegerin (OPG), P-selectin, tumor necrosis factor-alpha (TNFα), and tumor necrosis factor receptor II. To account for head size, both TCB (TCBV) and WMH (WMH/TCV) were divided by total cranial volume. We used multivariable linear regression to relate 10 log-transformed inflammatory biomarkers to brain MRI measures.

Results—In multivariable models, inflammatory markers as a group were associated with TCBV ($p < 0.0001$) but not WMH/TCV ($p = 0.28$). In stepwise models adjusted for clinical covariates with backwards elimination of markers, IL-6 and OPG were inversely associated with TCBV; TNFα was inversely related to TCBV in a subset of 1,430 participants. Findings were similar in analyses excluding individuals with prevalent cardiovascular disease. The relations between TCBV and inflammatory markers were modified by both sex and age, and generally were more pronounced in men and in older individuals.

Conclusions—Although our observational cross-sectional data cannot establish causality, they are consistent with the hypothesis that higher inflammatory markers are associated with greater atrophy than expected for age.

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Cerebrovascular disease and Alzheimer disease (AD) are associated with inflammation. Inflammatory cytokines are elevated during acute stroke[1,2] and in the CSF of individuals with subcortical vascular dementia.[3] Furthermore, inflammation is a well-documented feature[4] and purported risk factor[5] for AD. Amyloid deposition stimulates neuroinflammatory processes with neurotoxic effects that exacerbate the pathogenesis of AD and increase neuronal damage.[6,7]

The examination of inflammatory markers in relation to cerebrovascular disease and AD has led to critical insights; however, diagnosed neurologic disease reflects the extreme clinical presentation of the pathologic processes involved. One method for evaluating early preclinical changes of AD and cerebrovascular disease is MRI. Accumulating evidence suggests that MRI white matter hyperintensities (WMH) represent cerebrovascular brain injury, as evidenced by their association with increased risk of stroke,[8] independent of major vascular risk factors.[9] Furthermore, MRI measures of cerebral atrophy reliably differentiate patients with AD from age-matched controls[10–16] and are associated with accelerated cognitive deterioration.[13,17,18] Thus, findings on brain MRI may serve as early biologic markers of AD and cerebrovascular disease.

Though CRP has been related to WMH in a previous community-based sample,[19] to our knowledge the relations of a panel of inflammatory markers to WMH and total brain volume (TCB) have not been examined in community-based samples. We hypothesized that increased systemic inflammatory biomarkers would be associated with increased WMH and decreased TCB in a community-based cohort of adults without clinical dementia or stroke. We also sought to examine whether sex or age modified the relations between inflammatory markers and brain MRI findings.

Methods

Participants

The design and selection criteria of the Framingham Offspring Study have been described elsewhere.[20] Briefly, 5,124 participants were recruited in 1971 and have been examined every 4 to 8 years since. The current sample was derived from the 3,539 participants attending the seventh examination cycle (1998 through 2001). Participants underwent routine physical examination, medical history, and laboratory assessment of cardiovascular risk factors. From March 1999 to December 2004, 2,254 participants attending the seventh Offspring examination cycle consented to undergo brain MRI. As previously reported, the Offspring participants who refused or were unable to undergo MRI were older and generally less healthy than those who received MRI.[8] Of the 2,254 participants who underwent MRI, 328 were excluded from the present study if they had a neurologic condition that could substantively alter our brain MRI measures (e.g., dementia, clinical stroke, MS; n = 49); underwent physical examination off-site (n = 77); were missing blood (n = 11) or covariate data (n = 30); or did not have all nine circulating inflammatory markers measured ([n = 161]; a tenth marker, tumor necrosis factor-α [TNFα], was measured in a smaller sample [496 fewer] as part of another protocol [PI: J.B.M.], which started later in the examination cycle). After exclusions, a total of 1,926 participants were included in the current investigation, 1,430 of whom had measures of TNFα. The study protocol was approved by the Institutional Review Board of Boston University Medical Center, and all participants provided written informed consent.

Clinical covariates were defined at the seventh examination cycle. Systolic and diastolic blood pressures were the mean of the two measurements obtained by a Framingham Heart Study physician. Current smoking (i.e., yes/no within the year prior to examination[7]) and medication use were ascertained by self-report. The clinical definition of diabetes mellitus included fasting blood glucose ≥126 mg/dL or the use of oral hypoglycemic or insulin. Information about prior

cardiovascular disease (i.e., coronary heart disease, heart failure, stroke, and intermittent claudication) was obtained via medical histories and physical examinations conducted at the Framingham Heart Study, as well as hospitalization and personal physician records. A panel of three experienced investigators determined cardiovascular disease diagnoses using previously described criteria.[21]

MRI acquisition

The Framingham MRI acquisition protocol has been reported in detail elsewhere.[8] Briefly, the majority of participants were imaged on a Siemens Magnetom 1 T field strength MR machine using a T2-weighted double spin-echo coronal imaging sequence of 4 mm contiguous slices from nasion to occiput with a repetition time of 2,420 msec, echo time (TE) of TE1 20/TE2 90 msec; echo train length 8 msec; field of view 22 cm; and an acquisition matrix of 182 × 256 interpolated to 256 × 256 with one excitation. After acquisition of the MR scans, the digital information was transferred to a central laboratory directed by one of the authors (C.D.) for postprocessing and analysis. All analyses were performed blinded to demographic and clinical information. MRI quantification was performed with a custom-written computer program operating on a UNIX, Solaris platform. Image evaluation was based on a semiautomatic segmentation analysis that involves operator-guided removal of non-brain elements as previously described.[22] In brief, non-brain elements were manually removed from the image by operator guided tracing of the dura matter within the cranial vault including the middle cranial fossa but above the posterior fossa and cerebellum. The resulting measure of the cranial vault was defined as the total cranial volume (TCV) and served as an estimate of head size to account for recognized sex differences. Quantification of TCB and WMH required a multistep process that began with image segmentation to define brain matter from CSF.[23] For segmentation of brain from CSF, a difference image was created by subtracting the second from the first echo image. Image intensity non-uniformities were removed from the difference image,[24] and the resulting corrected image was modeled as a mixture of two Gaussian probability functions with the segmentation threshold determined at the minimum probability between these two distributions.[22] The protocol for WMH segmentation from brain matter has been described elsewhere,[23,24] as has the inter-rater reliabilities for these methods.[22,25,26] For the present study, repeat analysis of intra- and inter-rater reliabilities were consistently above 0.90.

Circulating inflammatory markers

At examination cycle 7, we measured 10 biomarkers representing a variety of pathways and phases of the inflammatory process.[27–29] The biomarkers included plasma CD40 ligand (CD40L), osteoprotegerin (OPG), P-selectin, tumor necrosis factor receptor II (TNFr2), and TNFα; and serum C-reactive protein (CRP), interleukin-6 (IL-6), soluble intracellular adhesion molecule-1 (sICAM-1), monocyte chemoattractant protein-1 (MCP-1), and myeloperoxidase (MPO). Fasting morning samples were collected and plasma and serum aliquots were stored at −70 °C. We measured all biomarkers, except CRP, in duplicate with commercially available ELISA kits from R&D Systems (sICAM-1, IL-6, MCP-1, P-selectin, TNFr2, TNFα), Bender MedSystems (CD40L), Oxis (MPO), and BIOMEDICA (OPG). High sensitivity CRP was measured using the Dade Behring BN100 nephelometer. The reproducibility of the biomarkers was good. The intra-assay coefficients of variation were as follows: CD40L 4.4%, IL-6 3.1%, sICAM-1 3.1%, MCP-1 4.1%, MPO 3.0%, OPG 3.7%, P-selectin 3.0%, TNFα 8.8%, and TNFr2 2.3%. The Kappa statistic for 146 CRP samples run in duplicate was 0.95.[29]

Statistical analysis

For analytical purposes, TCV was used to adjust for head size for both TCB (i.e., TCBV, or TCB/TCV) and WMH (i.e., WMH/TCV) and subsequently multiplied by 100. Because the

WMH/TCV ratio and the inflammatory markers had skewed distributions, they were log-transformed to normalize their distributions for analyses. For each dependent MRI measure (TCBV and log-WMH/TCV), a global omnibus test based on covariate-adjusted linear regression assessed if at least one inflammatory marker (excluding TNFα) was significantly related to the MRI measure. If the omnibus test was significant at the 0.05 level for the given MRI measure, we conducted backwards regression forcing in all covariates and inflammatory markers and then removing inflammatory markers one at a time until only markers with covariate-adjusted significance levels below 0.05 remained in the regression model. Covariates included in the multivariable model were selected based on prior work,[30,31] and included age, sex, systolic blood pressure, diastolic blood pressure, body mass index, height, total/high density lipoprotein cholesterol, current smoking, fasting glucose, triglycerides, diabetes, hypertension treatment, hormone replacement therapy, lipid lowering treatment, aspirin (at least 3 days per week), atrial fibrillation, electrocardiographic left ventricular hypertrophy, and prevalent cardiovascular disease. Secondarily, we repeated the analysis above with TNFα included in the set of markers, which resulted in a smaller dataset (n = 1,430).

Our primary model was to examine which subset of inflammatory markers was associated significantly with the brain MRI measures. However, multiple investigative groups have different sets of markers available, so we analyzed the covariate-adjusted linear relations of each log-transformed inflammatory marker, one marker at a time, to the dependent MRI measures, log-WMH/TCV and TCBV. To account for multiple testing in the individual marker analyses we used $p < 0.01$ to declare a significant linear relationship. Regression analyses with interaction terms were conducted to assess effect modification of these relationships by sex and age (<60 vs ≥60 years), again using $p < 0.01$ for each marker to declare significant effect modification. All data were analyzed using SAS version 8.1.[32]

Results

Participant characteristics

The participants' clinical characteristics are provided in table 1. The mean age of the participants was 60 years (range 35 to 85 years). The untransformed systemic inflammatory marker and MRI median and 25th to 75th percentile data are provided in table 2. The regressions were based on the log-transformed inflammatory values. Among men, log-transformed WMH/TCV ranged from −6.75 to 0.66, untransformed WMH/TCV ranged from 0.00117% to 1.93%, and TCBV ranged from 64.89% to 85.52%. Among the women, log transformed WMH/TCV ranged from −8.29 to 0.68, untransformed WMH/TCV ranged from 0.00025% to 1.98%, and TCBV ranged 67.84% to 90.38%. Many of the inflammatory markers were significantly correlated with age. Specifically, sex-adjusted correlations between log-transformed inflammatory markers and age were as follows: CD40L (r = −0.05, $p = 0.09$), CRP (r = 0.15, $p < 0.0001$), IL-6 (r = 0.23, $p < 0.0001$), sICAM-1 (r = 0.07, $p = 0.01$), MCP-1 (r = 0.18, $p < 0.0001$), MPO (r = −0.04, $p = 0.10$), OPG (r = 0.39, $p < 0.0001$), P-selectin (r = 0.12, $p < 0.0001$), TNFr2 (r = 0.36, $p < 0.0001$), and TNFα (r = 0.17, $p < 0.0001$).

Inflammatory markers and WMH/TCV

The inflammatory markers as a group were not related to WMH/TCV ($p = 0.28$ for the multimarker covariate-adjusted global test of significance). In analyses excluding individuals with prevalent cardiovascular disease (n = 195), findings were essentially unchanged ($p = 0.25$). Analyzing each marker separately, we observed that none of the inflammatory markers examined were related to WMH/TCV at the 0.01 or 0.05 level of significance (table 3). At an alpha of 0.05 we had 90% power to detect a marker that would increase the R squared by at least 0.007 (on a scale of 0 to 1) if added to the linear model containing the above-mentioned covariates.

Inflammatory markers and TCBV

The covariate-adjusted regression model relating the inflammatory markers as a group to TCBV was significant ($p < 0.0001$ for the global test of nine markers). Backward regression retained IL-6 ($p = 0.002$) and OPG ($p = 0.001$) in the model as being associated with TCBV. No other markers were significant after adjustment for OPG and IL-6. From this multimarker model, comparing individuals in the 25th percentile to the 75th percentile of the log-transformed IL-6 the estimated mean decrease in TCBV was 0.24%, whereas for log-transformed OPG the estimated mean decrease in TCBV was 0.28%. A decrease in brain volume of 0.25% was comparable to that associated with 1.5 years of advancing age (i.e., the multivariable non-marker adjusted beta coefficient for age was −0.16, indicating a 0.16% decline in TCBV per 1 year of advancing age). In secondary analyses excluding individuals with prevalent cardiovascular disease (n = 195), findings were not materially altered ($p = 0.0006$ for the global test of significance). Similarly, findings were not substantively changed ($p = 0.0002$ for the global test of significance) in a secondary analysis including nonsteroidal anti-inflammatory drug (NSAID) use as a covariate, nor did NSAID use modify the effect of the markers on TCBV (NSAID-by-marker interaction $p > 0.15$ for IL-6 and OPG in covariate-adjusted individual marker models; $p > 0.05$ for all other markers).

To gain a sense of the magnitude of variability explained by the markers we calculated the R-squared for various covariate models. Age alone, without the inflammatory markers or clinical covariates, explained more than 30% of the interindividual variability in TCBV (R-squared for age alone = 0.306). The R-squared for age plus all of the nonmarker clinical covariates was 0.364. Hence after accounting for age and sex, clinical covariates only explained an additional 0.04 of the variability in TCBV; adding the significant markers increased the R-squared further by approximately 0.01. Of interest is that the R-squared of age and sex plus only the significant markers (i.e., no risk factor covariates) was 0.341, which is approximately only 0.02 less than the R-squared of age plus the non marker risk factor covariates. To better understand the relation between the significant inflammatory markers (i.e., OPG, IL-6), the covariates, and TCBV, exploratory analyses involving a stepwise regression model was conducted in which OPG and IL-6 were entered first into the model. As expected, the initial amount of variability explained by the markers (i.e., partial R-squared values) decreased after age was included in the model. Specifically, for IL-6, the partial R-squared values decreased from 0.0297 to 0.0079, and, for OPG, the values decreased from 0.0696 to 0.0058. The significant reduction of the markers' partial R-squared values was due to the high correlation between advancing age and the inflammatory markers as well as TCBV.[8] However, as discussed above, the inflammatory markers remained significantly associated with TCBV after adjustment for age and additional clinical covariates.

In covariate-adjusted models examining each marker individually (vs the stepwise multimarker model presented above, which retained both IL-6 and OPG in the model), IL-6 and OPG were significantly inversely associated with TCBV at a more stringent $p < 0.01$; CRP, sICAM-1, and P-selectin were marginally inversely associated with TCBV (p values between 0.01 and 0.05; table 3). For example, individuals with log-transformed OPG levels at the 75th percentile, compared to individuals with log-transformed OPG levels at the 25th percentile, would be expected to have a 0.30% lower level of mean TCBV ($p = 0.003$). This estimate would translate into a decrease in brain volume of at least 3 cm^3.

There were several significant interactions between sex and age and inflammatory biomarkers on TCBV (table 4 and table 5). For TCBV, we observed significant interactions ($p < 0.01$) between sex and OPG, P-selectin, and TNFr2, and between age and IL-6, MCP1, P-selectin, and TNFr2. In general, the sex and age interactions were related to the magnitude rather than the direction of the association between the inflammatory markers and TCBV; the markers usually were more strongly associated with TCBV in men and in older participants than in

female and younger individuals. Examining the markers individually, men and older individuals had associations for IL-6, P-selectin, and TNFα with TCBV ($p < 0.01$).

Secondary analyses with TNFα

Secondary analyses were performed utilizing a smaller dataset limited to those participants with measured TNFα data (n = 1,430). In the smaller dataset, the multimarker covariate-adjusted regression model was not associated with WMH/TCV ($p = 0.18$) but was associated with TCBV ($p < 0.0001$). Backward elimination identified that TNFα was associated with TCBV ($p = 0.0001$). Unlike the analysis on the larger set of participants, OPG and IL-6 were not retained in this backward model at the 0.05 level of significance. This finding is due to a 50% reduction in the OPG coefficient in the smaller dataset even without TNFα in the model, which accounts for the loss significance. However, the removal of IL-6 from the backward model appeared to relate to its correlation to TNFα. Specifically, the covariate-adjusted correlations with TNFα are 0.22 ($p < 0.001$) for IL-6 and 0.08 ($p = 0.003$) for OPG.

Exploratory analyses

Exploratory analyses were performed post hoc to further examine the null findings between the inflammatory markers and WMH/TCV. If WMH was adjusted for total brain volume (i.e., WMH/TBV) instead of TCV, the inflammatory markers as a group were not related to natural log-transformed WMH/TBV ($p = 0.27$ for the multimarker covariate-adjusted global test of significance). Findings were similar if inflammatory markers as a group were related to the unadjusted natural log-transformed WMH (i.e., WMH not indexed to TBV or TCV; $p = 0.27$ for the multimarker covariate-adjusted global test of significance). Last, findings between the inflammatory markers and WMH/TCV were not substantively altered if we included individuals with prevalent dementia and stroke (n = 42) in the analyses ($p = 0.29$ for the multimarker covariate-adjusted global test of significance).

Discussion

In our community-based cohort, we examined a large panel of inflammatory markers in relation to brain MRI findings. Though we did not observe a significant association between the inflammatory markers and WMH/TCV, IL-6, OPG, and TNFα were modestly associated with TCBV in stepwise multivariable-adjusted models. In analyses of each inflammatory marker separately, the same markers (IL-6, OPG, and TNFα) were significantly associated with TCBV. The relations between inflammatory markers and TCBV were modified by both sex and age, such that the association was strongest among men and individuals aged 60 and older. These data are among the first to report an association between TCBV and circulating inflammatory markers in a dementia-free, community-based sample. Though the clinical magnitude of these associations was heavily influenced by age, the decrease in brain volume observed when the inflammatory marker increased from the 25th to the 75th percentile was comparable to the decrease in brain volume associated with every 1.5 years of advancing age.

Prior research has linked inflammatory markers, such as OPG,[33] with AD, which is characterized in part by global cerebral atrophy. Due to the observational nature of our study, we are unable to conclude if the association between certain markers of inflammation (i.e., IL-6, OPG, and TNFα) and TCBV reflects a unique pathophysiologic insight specific to these markers or a nonspecific manifestation of inflammation. Furthermore, inflammatory cytokines, such as IL-6 and TNFα, are activated in many conditions associated with advancing age,[34] and inflammatory markers purportedly contribute to the pathophysiology of cardiovascular disease.[35] We acknowledge that circulating biomarkers of inflammation may not adequately represent inflammatory events at the tissue level in the CNS. Though we selected a broad panel of inflammatory markers to represent various phases of the inflammatory cascade, we did not

measure all inflammatory biomarkers that previously have been related to neurologic phenotypes.

Prior research utilizing animal models has related hippocampal and entorhinal cortex atrophy to chronic neuroinflammation.[36] We extend the previous work by reporting an association between inflammatory markers and TCBV in a nondemented, community-based sample, and the association was most robust among individuals over age 60 years. The finding of effect modification by age coincides with a period in the life cycle when both health risk factors and cognitive impairment are of emerging relevance. We hypothesize, but cannot examine in a cross-sectional study, that the presence of systemic inflammation accelerates age-related changes in the brain, which may either act as a susceptibility factor or possibly an associated marker for the early evolution of AD neuropathology.[4]

We did not find an association between biomarkers of inflammation and WMH/TCV, which contrasts with prior epidemiologic evidence.[19] In an older cohort, the Rotterdam Scan Study documented that higher CRP concentrations were associated with both increased WMH cross-sectionally and WMH progression longitudinally,[19] which supports the well-established association between inflammation and the atherosclerotic process.[35] There are several potential explanations for our null finding, including the fact that our sample was predominantly healthy, younger, and had less WMH burden. Another reason may be the technique we employed for measuring WMH (i.e., we digitized and quantified WMH in contrast to semiquantitative estimates). A third explanation for the discrepancy is that we had adequate statistical power to detect modest associations between inflammatory markers and WMH cross-sectionally. However, we cannot exclude the possibility that the inflammatory markers had a very small association with WMH, which might have been detected in a larger dataset. Longitudinal study of our cohort may yield different relations between WMH and inflammatory markers that are more consistent with the previous findings.

Our study has a number of strengths, including the core MRI reading laboratory blinded to clinical characteristics, a large community-based cohort free from clinical dementia and stroke, simultaneous examination of 9 or 10 inflammatory biomarkers, comprehensive ascertainment of potential confounding variables, and stringent quality control procedures for measurement of both inflammatory biomarkers and brain MRI. However, the present findings must be tempered by several caveats. First, the age and racial makeup of the Framingham Offspring Study is predominantly white, of European descent, and middle-aged to elderly; the generalizability to other races, ethnicities, and age groups is unknown. The exclusion of institutionalized individuals and participants with clinical stroke and the inclusion of individuals willing to undergo MRI resulted in a generally healthy sample thereby reducing the likelihood of finding relations that may be present in the general population, which includes individuals with cognitive impairment or stroke.

Because we observed effect modification by age and sex and we present several models, our data might in part represent false positives due to multiple testing. To address multiple testing concerns we used an omnibus test (i.e., examining the markers as a group in relation to the MRI phenotypes) as the primary analysis. In addition, for the specific marker analyses (tables 3 through 5), we used a p value threshold of < 0.01. We cannot exclude the possibility that only very high values of some of the markers may be related to MRI phenotypes; we did not pursue threshold analyses because we sought to limit the number of statistical tests. Nevertheless, our findings will need to be replicated in another cohort.

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Table 1

Study sample clinical characteristic

Characteristics	Women (n = 1,032)	Men (n = 894)
Age, y	60.3 (9.4)	60.5 (9.3)
Systolic blood pressure, mm Hg	124 (19)	127 (17)
Diastolic blood pressure, mm Hg	72 (9)	75 (9)
Body mass index, kg/m^2	27.4 (5.8)	28.6 (4.4)
Total/HDL cholesterol, ratio	3.7 (1.2)	4.6 (1.4)
Fasting glucose, mg/dL	99 (22)	108 (29)
Triglycerides, mg/dL	126 (72)	143 (106)
Cigarette smoking	13	12
Diabetes	9	15
Hypertension treatment	27	34
Lipid lowering medication	15	22
Hormone replacement therapy	35	—
Aspirin treatment	25	41
Prevalent cardiovascular disease	7	14

Values are percentages or mean (SD); cardiovascular disease does not include clinical stroke or TIA.

Table 2
Inflammatory marker and MRI data

Characteristic	Women (n = 1,032), median (25th, 75th percentile)	Men (n = 894), median (25th, 75th percentile)
Untransformed inflammatory markers		
CD40L, ng/mL	1.4 (0.6, 4.1)	1.2 (0.5, 3.5)
CRP, mg/L	2.4 (1.0, 5.5)	1.7 (0.9, 4.1)
s-ICAM-1, ng/mL	238 (210, 280)	240 (209, 280)
IL-6, pg/mL	2.6 (1.7, 4.1)	2.7 (1.8, 4.2)
MCP-1, pg/mL	305 (250, 379)	316 (257, 388)
MPO, ng/mL	38.2 (27.1, 56.6)	42.7 (30.0, 63.4)
OPG, pmol/L	5.6 (4.6, 6.6)	5.1 (4.3, 6.1)
P-selectin, ng/mL	34.1 (26.7, 42.3)	38.4 (30.3, 48.4)
TNFr2, pg/mL	1,915 (1604, 2310)	2,005 (1695, 2454)
TNF-α*, pg/mL	1.2 (0.9, 1.6)	1.2 (0.9, 1.6)
MRI data		
Untransformed		
TCBV × 100, %	78.8 (76.6, 80.5)	77.9 (75.6, 80.0)
WMH/TCV × 100, %	0.05 (0.02, 0.09)	0.04 (0.02, 0.08)
Log-transformed		
WMH/TCV	−3.1 (−3.7, −2.5)	−3.2 (−3.8, −2.6)

* TNF-α data are based on a subset of male (n = 676) and female (n = 754) participants.

CRP = C-reactive protein; sICAM-1 = soluble intracellular adhesion molecule-1; IL = interleukin; MCP-1 = monocyte chemoattractant protein-1; MPO = myeloperoxidase; OPG = osteoprotegerin; TNF = tumor necrosis factor; WMH = white matter hyperintensities; TCV = total cranial volume.

Table 3

Multivariable-adjusted regression of each inflammatory marker on brain MRI measure

	TCBV*100			Log-transformed WMH/TCV*100		
	Estimate	**95% CI**	***p* Value**	**Estimate**	**95% CI**	***p* Value**
CD40L	0.11	−0.08, 0.30	0.25	−0.003	−0.07, 0.06	0.94
CRP	−0.25	−0.45,−0.05	0.02	−0.05	−0.012,0.03	0.21
s-ICAM-1	−0.17	−0.32,−0.02	0.03	0.05	−0.01, 0.10	0.09
IL-6	−0.26	−0.41,−0.10	0.001*	0.01	−0.04, 0.06	0.70
MCP-1	−0.01	−0.16, 0.13	0.86	−0.05	−0.10, 0.001	0.06
MPO	−0.15	−0.31, 0.005	0.06	−0.03	−0.08, 0.03	0.32
OPG	−0.30	−0.46, −.014	0.0003*	0.02	−0.03, 0.08	0.42
P-selectin	−0.19	−0.34,−0.03	0.02	−0.02	−0.07, 0.04	0.56
TNFr2	−0.15	−0.31, 0.005	0.06	0.01	−0.05, 0.06	0.81
TNF-α†	−0.36	−0.52,−0.20	<0.0001*	−0.03	−0.09, 0.02	0.26

Total n = 1,926; estimate = estimated average difference in MRI volume as log-transformed inflammatory marker increases from the 25th to the 75th percentile; 95% CI = 95% CI for change value. Models adjusted for age, sex, systolic blood pressure, diastolic blood pressure, body mass index, height, total/high density lipoprotein cholesterol, current smoking, fasting glucose, triglycerides, diabetes, hypertension treatment, hormone replacement therapy, lipid lowering treatment, aspirin, atrial fibrillation, electrocardiographic left ventricular hypertrophy, and prevalent cardiovascular disease.

*Significant.

†TNF-α data based on a participant subset (n = 1,430).

WMH = white matter hyperintensities; TCV = total cranial volume; CRP = C-reactive protein; sICAM-1 = soluble intracellular adhesion molecule-1; IL = interleukin; MCP-1 = monocyte chemoattractant protein-1; MPO = myeloperoxidase; OPG = osteoprotegerin; TNF = tumor necrosis factor.

Table 4

Sex interactions for multivariable adjusted regression models for each inflammatory marker separately in relation to TCBV*100

		Female			Male		
	Interaction *p* value	**Estimate**	**95% CI**	***p* Value**	**Estimate**	**95% CI**	***p* Value**
CD40L	0.57	0.19	−0.06, 0.45	0.14	0.02	−0.26, 0.29	0.91
CRP	0.78	−0.36	−0.64,−0.08	0.012	−0.08	−0.37, 0.21	0.59
s-ICAM-1	0.09	−0.10	−0.31, 0.11	0.34	−0.24	−0.46, −0.02	0.03
IL-6	0.015	−0.17	−0.38, 0.04	0.10	−0.32	−0.55, −0.09	0.006*
MCP-1	0.0104	0.09	−0.11, 0.28	0.40	−0.14	−0.36, 0.07	0.20
MPO	0.95	−0.12	−0.33, 0.09	0.26	−0.19	−0.43, 0.04	0.10
OPG	0.006*	−0.25	−0.44, −0.05	0.014	−0.31	−0.57, −0.06	0.02
P-selectin	0.007*	−0.03	−0.24, 0.18	0.77	−0.33	−0.55, −0.10	0.005*
TNFr2	0.0007*	−0.06	−0.29, 0.16	0.57	−0.26	−0.50, −0.02	0.03
TNF-α†	0.013	−0.27	−0.49, −0.04	0.02	−0.44	−0.67, −0.21	0.0002*

N = 1,926; estimate = estimated average difference in MRI volume as log-transformed inflammatory marker increases from the 25th to the 75th percentile; 95% CI = 95% CI for change value; model adjusted for the covariates listed in the table 3 legend.

*Significant.

†TNF-α data based on a subset of participants (n = 1,430).

CRP = C-reactive protein; sICAM-1 = soluble intracellular adhesion molecule-1; IL = interleukin; MCP-1 = monocyte chemoattractantprotein-1; MPO = myeloperoxidase; OPG = osteoprotegerin; TNF = tumor necrosis factor.

Table 5

Age interactions for multivariable adjusted regression models for each inflammatory marker separately in relation to TCBV*100

		<60 years			≥60 years		
	Interaction *p* value	**Estimate**	**95% CI**	***p* Value**	**Estimate**	**95% CI**	***p* Value**
CD40L	0.02	0.07	−0.18, 0.33	0.58	0.16	−0.10, 0.42	0.23
CRP	0.25	−0.19	−0.48, 0.09	0.18	−0.24	−0.52, 0.03	0.08
s-ICAM-1	0.14	−0.11	−0.32, 0.10	0.30	−0.23	−0.43,−0.03	0.02
IL-6	0.0001*	−0.08	−0.27, 0.11	0.41	−0.34	−0.56,−0.12	0.002*
MCP-1	0.001*	0.08	−0.13, 0.29	0.46	−0.12	−0.33, 0.08	0.24
MPO	0.45	−0.06	−0.26, 0.14	0.57	−0.27	−0.50,−0.03	0.03
OPG	0.02	−0.28	−0.49,−0.07	0.0096	−0.17	−0.38, 0.03	0.10
P-selectin	0.0002*	0.05	−0.14, 0.24	0.61	−0.37	−0.59,−0.16	0.0007*
TNFr2	0.0001*	0.01	−0.19, 0.21	0.89	−0.22	−0.44, 0.00	0.05
TNF-α†	0.001*	−0.20	−0.41, 0.02	0.07	−0.39	−0.61,−0.18	0.0004*

N = 1,926; estimate = estimated average difference in MRI volume as log-transformed inflammatory marker increases from the 25th to the 75th percentile; 95% CI = 95% CIs for change value; for model covariates please see Table 3 legend.

*Significant.

†TNF-alpha data is based on a subset of participants (n = 1,430).

CRP = C-reactive protein; sICAM-1 = soluble intracellular adhesion molecule-1; IL = interleukin; MCP-1 = monocyte chemoattractant protein-1; MPO = myeloperoxidase; OPG = osteoprotegerin; TNF = tumor necrosis factor.