Stroke

CLINICAL AND POPULATION SCIENCES

Clinical Relevance of Cortical Cerebral Microinfarcts on 1.5T Magnetic Resonance Imaging in the Late-Adult Population

Saima Hilal, PhD; Arwin Doolabi, BSc; Henri Vrooman, PhD; M. Kamran Ikram, PhD; M. Arfan Ikram, PhD; Meike W. Vernooij, PhD

BACKGROUND AND PURPOSE: Cortical cerebral microinfarcts (CMIs) have been linked with dementia and impaired cognition in cross-sectional studies. However, the clinical relevance of CMIs in a large population-based setting is lacking. We examine the association of cortical CMIs detected on 1.5T magnetic resonance imaging with cardiovascular risk factors, cerebrovascular disease, and brain tissue volumes. We further explore the association between cortical CMIs with cognitive decline and risk of stroke, dementia, and mortality in the general population.

METHODS: Two thousand one hundred fifty-six participants (age: 75.7±5.9 years, women: 55.6%) with clinical history and baseline magnetic resonance imaging (January 2009–December 2013) were included from the Rotterdam Study. Cortical CMIs were graded based on a previously validated method. Markers of cerebrovascular disease and brain tissue volumes were assessed on magnetic resonance imaging. Cognition was assessed using a detailed neuropsychological test at baseline and at 5 years of follow-up. Data on incident stroke, dementia, and mortality were included until January 2016.

RESULTS: Two hundred twenty-seven individuals (10.5%) had ≥1 cortical CMIs. The major risk factors of cortical CMIs were male sex, current smoking, history of heart disease, and stroke. Furthermore, presence of cortical CMIs was associated with infarcts and smaller brain volume. Persons with cortical CMIs showed cognitive decline in Stroop tests (color-naming and interference subtasks; β for color-naming, 0.18 [95% CI, 0.04–0.33], *P* interaction ≤0.001 and β for interference subtask, 1.74, [95% CI, 0.66–2.82], *P* interaction ≤0.001). During a mean follow-up of 5.2 years, 73 (4.3%) individuals developed incident stroke, 95 (5.1%) incident dementia, and 399 (19.2%) died. People with cortical CMIs were at an increased risk of stroke (hazard ratio, 1.18 [95% CI, 1.09–1.28]) and mortality (hazard ratio, 1.09 [95% CI, 1.00–1.19]).

CONCLUSIONS: Cortical CMIs are highly prevalent in a population-based setting and are associated with cardiovascular disease, cognitive decline, and increased risk of stroke and mortality. Future investigations will have to show whether cortical CMIs are a useful biomarker to intervene upon to reduce the burden of stroke.

Key Words: cognitive dysfunction ■ cortical microinfarcts ■ dementia ■ magnetic resonance imaging ■ stroke

Cortical cerebral microinfarcts (CMI) are typically defined as sharply delimited microscopic regions of cellular death, which were first identified on autopsy studies.[1] These lesions are reported to be highly prevalent not only in demented individuals but also among cognitively normal elderly. Pathological studies suggest that CMIs may exist in hundreds and thousands in the affected brain likely disrupting structural and functional brain connections.[2,3] Previously considered as invisible lesions during life, CMIs are increasingly made visible in

Correspondence to: Saima Hilal, PhD, Saw Swee Hock School of Public Health, National University of Singapore, Tahir Foundation Bldg, 12 Science Dr 2, Singapore 117549. Email saimahilal@nus.edu.sg

This article was sent to Mayank Goyal, Consulting Editor, for review by expert referees, editorial decision, and final disposition.

The Data Supplement is available with this article at https://www.ahajournals.org/doi/suppl/10.1161/STROKEAHA.120.032085.

For Sources of Funding and Disclosures, see page 930.

Stroke is available at www.ahajournals.org/journal/str

Nonstandard Abbreviations and Acronyms

APOE-ε4	apolipoprotein ε4
CeVD	cerebrovascular disease
CMI	cortical cerebral microinfarcts
EDIS	Epidemiology of Dementia in Singapore
FLAIR	fluid-attenuated inversion recovery
MRI	magnetic resonance imaging
PD	proton-density weighted
RS	Rotterdam Study
WMH	white matter hyperintensities

vivo using conventional imaging techniques, thus enabling the study of longitudinal impact of these lesions on brain damage and cognitive dysfunction.[4,5]

Previous studies have shown that the cortical CMIs detected on 3T magnetic resonance imaging (MRI) are associated with prevalent dementia and worse performance on visuoconstruction and language domains in a memory-clinic setting.[6] These findings echo in a subsample of population-based study where cortical CMIs were related to severe cognitive impairment and cognitive functioning independent of other markers of cerebrovascular disease (CeVD).[4] However, these data have been restricted to cross-sectional studies, which limits assessment of the temporal relationship between cortical CMIs and cognitive impairment. Thus far, limited data have shown that cortical CMIs at baseline predict cognitive decline in the domains of memory and language over 2 years of follow-up in a memory-clinic setting as well as on Montreal Cognitive Assessment in a stroke cohort.[5,7] Moreover, no study has yet examined the effects of cortical CMIs as a potential risk indicator of subsequent neurological diseases such as incident stroke, dementia, and mortality. Thus, there is a need to study the impact of cortical CMIs on more subtle brain changes and explore the potential clinical implications of these lesions in a large population-based setting consisting of cognitively normal individuals.

We examined the prevalence of cortical CMIs on 1.5T MRI scans and their association with cardiovascular risk factors, CeVD, and brain tissue volumes. We also explored the association of cortical CMIs with cognitive decline and their link with risk of stroke, dementia, and mortality in the RS (Rotterdam Study).

METHODS

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Study Population

The RS is a population-based prospective cohort study among middle age and elderly persons living in the Ommoord district in the city of Rotterdam, the Netherlands.[8] The RS consists of 3 cohorts where participants undergo follow-up every 3 to 4 years. In 1990 to 1993, 7983 persons participated and were re-examined every 3 to 4 years (RS-I). In 2000 to 2001, the cohort was expanded by 3011 persons who had not yet been part of the RS-II. MRI was incorporated into the core study protocol from 2005 onwards. All the eligible participants (without MRI contraindications and claustrophobia) are invited for MRI scans. For the current analyses, we retrospectively graded the MRI scans that were acquired between 2009 and 2012 in RS-I and RS-II which included T1-weighted, proton-density weighted (PD), Fluid Attenuated Inversion Recovery (FLAIR), and T2*weighted gradient recalled echo. Of the 3230 participants who took part in the RS, 2811 were considered eligible for MRI scans. Out of 2811 eligible participants, 2249 (80%) underwent MR imaging whereas the rest of the 562 (20%) subjects did not undergo MRI due to inability to tolerate MRI scan (claustrophobia, physical complaints) or refusal to participate (Figure I in the Data Supplement). Individuals with incomplete or ungradable scans (n=47) or prevalent dementia (n=46) were excluded giving a sample size of 2156 for final analysis. All the individuals underwent interviews and extensive set of examinations (clinical information and brain MRI) on the same day in the specially built research facility. All individuals who have provided consent for imaging underwent one MRI acquisition in each visit.

The RS has been approved by the Medical Ethics Committee of the Erasmus MC and by the Dutch Ministry of Health, Welfare and Sport. All participants provided written informed consent to have their information being obtained from their treating physicians.

Associated Factors

For this study, data on demographics and medical history were used that were collected at baseline (2009–2012). Education was categorized as primary or lower/intermediate vocational education and higher vocational education. Hypertension was defined as the systolic blood pressure ≥140 mm Hg and diastolic blood pressure ≥90 mm Hg, or prescription of antihypertensive medication. Hyperlipidemia was defined as the total cholesterol levels ≥5.2 mmol/L or prescription of lipid-lowering medication. Diabetes was defined as fasting blood glucose ≥7 mmol/L or receiving treatment for diabetes. Smoking was categorized into former, current, and never smokers. APOE (apolipoprotein E) genotype was determined using polymerase chain reaction on coded DNA samples. Distribution of APOE genotype and allele frequencies was in the Hardy-Weinberg equilibrium. APOE-ε4 carrier status was defined by the presence of at least one ε4 allele. Body mass index was defined as the weight in kilograms per height in m^2. Heart disease was defined as the history of myocardial infarction, atrial fibrillation, and cardiac intervention such as angioplasty, coronary artery bypass grafting, and other coronary revascularization procedures. History of stroke was assessed by using home interviews and by reviewing medical records. Incident cardiovascular factors were defined as the development of hypertension, hyperlipidemia, diabetes, and heart diseases on follow-up.

Brain Imaging

Brain MRI scans were performed on a 1.5T scanner (GE Signa Excite; GE Healthcare, Waukesha, WI) with multi-sequence

protocol.[8] The sequences included 3-dimensional T1-weighted, 2-dimensional PD, 2-dimensional FLAIR, and 3-dimensional T2*weighted gradient recalled echo images, the details of which are provided in the Data Supplement. The following MRI markers were determined:

Cortical CMIs

Cortical CMIs were graded on T1, PD-weighted, and FLAIR sequences and were defined as hypointense lesions on T1-weighted images (sagittal and axial planes), <5 mm in diameter, restricted to cortex and perpendicular to the cortical surface.[4,6] The location of a hypointense cortical lesion found on T1 was confirmed on FLAIR and PD-weighted images. The lesion was rated as a definite cortical CMI if hyperintense or isointense (with the surrounding tissue) on FLAIR and PD and isointense on T2*weighted gradient recalled echo images (Figure 1). Moreover, a T1-weighted hypointensity with cavitation on FLAIR images and those that are located >1 cm distance from the large cortical infarcts were still considered as cortical CMIs. The lesion was discarded as a cortical CMI if at the same location a hypointense signal was found on FLAIR or PD, indicating the T1 hypointense lesion was either due to a hemorrhagic lesion, a vessel, or an artifact.[6] The grading of cortical CMIs followed the established criteria developed on 3T and 7T MRI scans previously.[6] MRI rating for cortical CMIs was independently performed by 2 trained graders (Dr Hilal and A. Doolabi) with 7 and one year of experience in neuroimaging gradings. All cortical CMIs were graded blinded to subject's characteristics. A subset of 60 scans enriched with cortical CMIs, was randomly selected among the MRI scans, which were graded by both raters. All the identified cortical CMIs were discussed in the weekly consensus meetings. The inter-rater reliability performed on 60 scans showed good to excellent agreement (κ for presence and absence of CMIs=0.83, Dice similarity coefficient for numbers of CMIs=0.80).

Markers of CeVD

Large cortical infarcts were identified as focal lesions of >5 mm involving cortical gray matter with hyperintense rim on FLAIR images and center following CSF intensity. Lacunes were defined as lesions involving the subcortical regions, 3 to 15 mm in diameter, with low signal on T1-weighted image, a high signal on PD image and a hyperintense rim with a center following CSF intensity on FLAIR sequence.[9] Cerebral microbleeds were graded as small, round to ovoid focal areas of hypointense signal intensity with blooming on 3-dimensional T2*-weighted gradient recalled echo images.[10] White matter hyperintensities (WMH) were automatically segmented using a custom-developed method as described below.

Brain Tissue Segmentation

Image preprocessing and the tissue classification algorithm have been described elsewhere.[11] Briefly, a k-nearest-neighbor brain tissue technique was used to classify voxels into CSF, gray matter, and normal appearing white matter, and volume (mL) was calculated from these measurements. WMH volumes were detected using an adapted threshold technique making use of the tissue segmentation method.[12] Intracranial volume was the sum of the CSF, gray matter, normal white matter, and WMH. Hippocampus volume was derived using a model-based automated procedure (Free Surfer,v.5.1.0) on T1-weighted images.[13]

Cognitive Assessment

A detailed neuropsychological test battery was used to assess cognitive function which included Letter Digit Substitution Task, Word Fluency Test, Stroop test (consisting of reading, color-naming, and interference subtasks), 15-word Verbal Learning Test (consisting of immediate, delayed and recognition), and Perdue Peg Board Test at baseline (2009–2012) and on follow-up (2013–2016).[14,15] To be included in the cognitive decline analysis, the participants must have a minimum of 2 cognitive scores, that is, baseline and that of follow-up.

Assessment of Dementia

Participants were screened for dementia at baseline and at follow-up examinations using Mini-Mental State Examination and the Geriatric Mental Schedule organic level.[16] Screen-positives (Mini-Mental State Examination <26 or Geriatric Mental Schedule organic level >0) subsequently underwent an examination and informant interview with the Cambridge Examination for Mental Disorders in the Elderly. A consensus panel led by a consultant neurologist established the final dementia diagnosis (*Diagnostic and Statistical Manual of Mental Disorders*, Third Edition - Revised). Additionally, the whole cohort was continuously monitored for dementia through computerized linkage of the study database and digitized medical records from general practitioners and the Regional Institute for Outpatient Mental Health Care. When required and available, neuroimaging was used to facilitate dementia diagnosis.

Assessment of Stroke

Stroke was defined according to the World Health Organization criteria. On study entry, history of stroke was assessed during baseline interview and verified by reviewing medical records. After enrolment, participants were continuously monitored for incident stroke through automated linkage of general practitioners' medical records with the study database, as done for dementia. Nursing home physicians' and general practitioner files of participants who moved out of the district were checked on a regular basis. Research physicians reviewed all potential strokes using hospital discharge letters and information from general practitioners and nursing home physicians.[14] Strokes were further classified as ischemic or hemorrhagic stroke based on neuroimaging reports and hospital discharge letters, and unspecified if neuroimaging was absent.

Mortality

Vital status was obtained on a weekly basis via municipal population registries and through general practitioners' and hospitals' databases. All-cause mortality was defined as participants who died from any cause during the total follow-up period.

Participants were followed from baseline (2009–2012) until the date of stroke, dementia, or death or date of the last contact in case of lost to follow-up or January 1, 2016 (end of the current study period for stroke and dementia) and June 1, 2018 (end of the study period for mortality), whichever came

 Stroke. 2021;52:922–930. DOI: 10.1161/STROKEAHA.120.032085

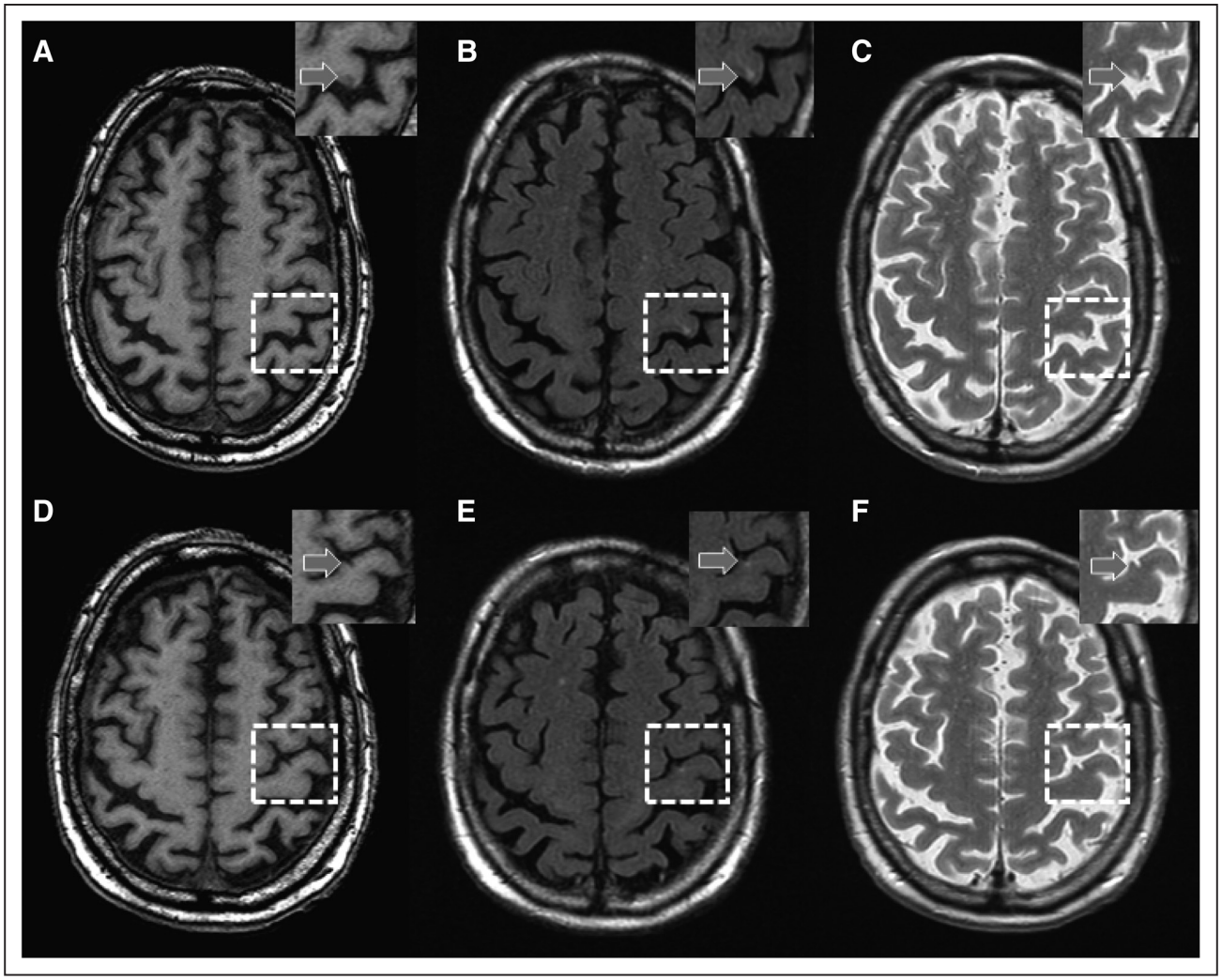

Figure 1. Example of cortical cerebral microinfarct (CMI) on 1.5T magnetic resonance imaging (MRI).
A representative cortical CMIs on 1.5T MRI which was first explored on a 3-dimensional T1-weighted image (**A** and **D**) and further confirmed on fluid-attenuated inversion recovery (FLAIR; **B** and **E**) and T2-weighted (**D** and **F**) images. White boxes indicate area with cortical CMI as shown in the magnified view in inlets (red arrow).

first. Follow-up data for stroke, dementia, and death was complete for 83%, 90%, and 99.8% of potential person-years.

Statistical Analysis

To determine the differences in demographic, vascular risk factors, cardiovascular disease, and MRI markers among participants with presence and absence of CMIs, χ^2 test, or Mann-Whitney *U* test was used for categorical and continuous variables. Log transformation was performed for skewed distributed variable (WMH volume). To determine the association between associated factors (demographic, vascular risk factors, cardiovascular disease, and MRI markers) and CMIs (presence versus absence), we performed logistic regression with odds ratios and 95% CI. We additionally constructed Poisson regression with CMI counts as outcome and computed rate ratios and 95% CIs. Initially, we adjusted the model for age, sex, vascular risk factors, and cardiovascular disease (APOE-ε4 carrier, body mass index, smoking, hypertension, diabetes, hyperlipidemia, heart disease, history of stroke) and MRI markers (cerebral microbleeds, lacunes, WMH, large cortical infarcts, total brain volume, hippocampal volume). The models with WMH, total brain volume, and hippocampal volume were further adjusted for intracranial volume.

To determine the association between cortical CMIs and cognitive decline, we fitted a linear regression model with generalized estimating equations. For this analysis, we included individuals with no history of stroke and cognitive data available at 2 time points, given that the missingness was not at random (most individuals with missing data had died before the second visit; n=230). We specified the correlation structure to be first order autoregressive and robust variance estimators were used. To investigate whether the effect of baseline CMIs on cognitive score is different at follow-up, we included an interaction term CMIs×time. We built models in the following fashion: Model I, adjusted for age, sex, education, vascular risk factors (smoking, hypertension, diabetes, hyperlipidemia), heart disease, and model II was additionally adjusted for all MRI markers. Because of the multiple testing performed within 9 cognitive tests, Bonferroni correction was applied to obtain a revised significance level of 0.05/9 to 0.0055. This was

applied to overall *P* value from the interaction term and for different cognitive test. The estimated marginal mean of each cognition score at baseline and follow-up were used to plot the trajectory of the expected cognition score among presence and absence of CMIs.

Cox-proportional hazard models were fitted to obtain estimated hazard ratios and 95% CI for the association between cortical CMIs (both presence and absence and counts) and incident stroke (including ischemic stroke), dementia, and mortality. We also removed persons with history of stroke for these analyses to determine the independent effects of cortical CMIs on incident outcomes and in particular first-ever stroke. We constructed 2 models to adjust for potential confounding in the similar fashion as described above. The proportionality assumption was tested by adding the interaction term CMIs×time in different models. All longitudinal analyses were further adjusted for incident cardiovascular factors as a sensitivity analysis.

RESULTS

Of the total 2156 participants, the average age at baseline was 75.7 years (interquartile range, 70.7–79.3) with 55.6% women. Prevalence of cortical CMIs was 10.5% (95% CI, 9.3%–11.9%), (227/2156). Among individuals with cortical CMIs, 140 (61.6%) had a single CMI, 69 (30.3%) had 2 to 4 CMIs, and 18 (7.9%) had ≥5 CMIs (range: 1–27). The cortical CMIs were frequently observed in parietal lobe (59%), followed by frontal (48%), occipital (11.4%), and temporal (5.2%) lobes. Characteristics of the study population based on presence and absence of CMIs are shown in Table 1.

Older age, male sex, current smoking, hyperlipidemia, previous heart disease, history of stroke, and MRI markers, that is, lacunes, WMH, large cortical infarcts, and total brain volume were associated with cortical CMIs in age-and-sex adjusted models (Table I in the Data Supplement). After including all vascular risk factors, cardiovascular disease and MRI markers in the final model, only male sex, current smoking, previous heart disease, history of stroke, lacunes, large cortical infarcts, and total brain volume were related to cortical CMIs on the scans (Table 2).

For all longitudinal analyses, a total of 75 individuals were excluded based on the history of stroke. During the mean follow-up of 5.2 years, 375 (17.3%) individuals were lost to follow-up and 150 had no cognitive assessment due to physical disability, refusal, and cognitive limitation giving a sample of 1556 for cognitive decline analysis. In the same duration, 375 individuals were lost to follow-up for incident stroke and 204 for incident dementia giving rise to a sample of 1706 for incident stroke analysis and 1877 for incident dementia analysis. Mortality was virtually complete for all 2081 participants.

Table 3 shows the association of cortical CMI counts with each cognitive score at follow-up. Individuals with increasing CMIs counts showed decline in Stroop tests 2 and 3 in all models. The interaction term between cortical CMIs and time was significant for Stroop tests. In terms of the rest of the cognitive tests, though a negative association was observed between cortical CMIs and cognitive performance, the effects were homogenous across all cognitive tests (nonsignificant interaction between cortical CMIs and time). After applying Bonferroni correction, these associations remained significant. Upon further adjustment for incident cardiovascular factors, the effect estimates remained unchanged (Table II in the Data Supplement).

The estimated marginal means of each cognitive test at baseline and follow-up visit stratified by presence and absence of CMIs are shown in Figure 2. Although we observed a difference in the means at each time point for presence and absence of CMIs, no association was observed between cortical CMIs and cognitive tests after adjusting for age, sex, education, vascular risk factors, heart disease, history of stroke, and MRI markers.

During a mean follow-up of 5.2 years, 73 (4.3%) individuals developed incident stroke (of whom 61 [3%] had incident ischemic stroke), 95 (5.1%) developed incident dementia (of whom 70 [3.4%] individuals had Alzheimer's dementia) and 399 (19.2%) individuals died. A total of 15/73 (20.5%) individuals with incident strokes and 10/95 (10.5%) with incident dementia had cortical CMIs on their baseline scans whereas the respective numbers for presence of cortical CMIs among persons who died were 61/399 (15.2%). People with increasing cortical CMIs counts at baseline were at increased risk of stroke and mortality as compared to without cortical CMIs (Table 4). Hazard ratio for stroke and mortality became slightly attenuated but remained significant after including MRI markers in the model. Moreover, cortical CMIs were independently associated with incident ischemic stroke in final adjusted model (hazard ratio for presence of CMIs, 2.18 [95% CI, 1.03–4.63] and hazard ratio for CMI counts, 1.87 [95% CI, 1.09–1.29]). No association was observed with incident dementia and its subtypes in all models. These results were also independent of incident cardiovascular factors (Table III in the Data Supplement).

DISCUSSION

In this study, we found that cortical CMIs are a common finding in a population-based setting with a prevalence of 10.5% graded on 1.5T MRI. The major risk factors for cortical CMIs are male sex, current smoking, previous heart disease, history of stroke, lacunes, large cortical infarcts, and total brain volume. Persons with cortical CMIs performed worse on Stroop testing on follow-up visit. More importantly, persons with cortical CMIs were at increased risk of clinical stroke and mortality.

Recently, cortical CMIs have gained increasing attention because of the feasibility of detecting these lesions on 3T MRI.[6] Using this imaging modality, the prevalence

Table 1. Characteristics of the Study Population (n=2156)

	CMI absent (n=1929)	CMI present (n=227)	*P* value
Age, y, median (IQR)	74.7 (70.9–79.3)	76.7 (71.7–82.0)	0.001*
Men, n (%)	819 (42.5)	138 (60.8)	<0.001*
Lower education, n (%)†	956 (50.4)	103 (46.2)	0.232
APOE-ε4 carrier, n (%)	454 (24.9)	64 (29.1)	0.182
BMI, kg/m^2, median (IQR)	27.0 (24.7–29.6)	26.9 (24.4–29.7)	0.694
Systolic blood pressure, mm Hg, median (IQR)	150 (137–165)	148.5 (137–165)	0.475
Diastolic blood pressure, mm Hg, median (IQR)	85 (78–92)	83.5 (77–91)	0.158
Total cholesterol, mmol/L, median (IQR)	5.4 (4.7–6.1)	5.1 (4.3–6)	0.002*
Glucose, mmol/L, median (IQR)	5.5 (5.2–6.1)	5.5 (5.1–6.2)	0.504
Smoking, n (%)*			0.002*
Former smoker	1052 (54.8)	141 (62.1)	
Current smoker	214 (11.2)	34 (15)	
Hypertension, n (%)	1652 (85.8)	205 (90.3)	0.063
Diabetes, n (%)	262 (13.6)	38 (16.7)	0.196
Hyperlipidemia, n (%)	1387 (72.0)	175 (77.1)	0.102
History of heart disease, n (%)	242 (12.8)	63 (28.1)	<0.001*
History of stroke, n (%)	52 (2.7)	23 (10.1)	<0.001*
Cerebral microbleeds, n (%)	574 (29.8)	85 (37.4)	0.017*
Lacunes, n (%)	227 (11.8)	57 (25.1)	<0.001*
WMH, mL, median (IQR)	5.6 (3.0–12.4)	8.8 (4.2–19.4)	<0.001*
Large cortical infarcts, n (%)	57 (3)	53 (23.3)	<0.001*
Total brain volume, mL, median (IQR)	894.7 (833.9–958.4)	893.1 (846.3–966.7)	0.412
Hippocampus volume, mL, median (IQR)	7.35 (6.64–8.01)	7.36 (6.54–7.89)	0.498

APOE indicates apolipoprotein E; BMI, body mass index; CMI, cortical cerebral microinfarct; IQR, interquartile range; and WMH, white matter hyperintensities.

*P<0.05.

†Missing in 1.7% participants.

Table 2. Association of Possible Risk Factors With Cortical CMIs

Risk factors	CMI (presence vs absence), OR (95% CI)	CMI (counts), RR (95% CI)
Demographic factors		
Age, y	0.98 (0.95–1.02)	0.95 (0.90–1.00)
Men	1.67 (1.07–2.62)	0.92 (0.68–1.25)
Cardiovascular factors		
APOE-ε4 carrier	1.43 (0.99–1.89)	1.11 (0.84–1.47)
BMI, (per unit increase)	0.97 (0.93–1.02)	0.95 (0.92–1.00)
Smoking		
Former	0.99 (0.67–1.48)	1.01 (0.76–1.34)
Current	1.25 (0.67–2.33)	1.54 (1.03–2.30)
Hypertension	1.06 (0.62–1.82)	1.35 (0.89–2.06)
Diabetes	1.06 (0.64–1.73)	0.87 (0.61–1.24)
Hyperlipidemia	1.20 (0.79–1.82)	1.17 (0.87–1.58)
History of heart disease	1.72 (1.11–2.65)	1.84 (1.36–2.48)
History of stroke	2.45 (1.11–5.41)	3.20 (2.70–4.05)
MRI markers		
Cerebral microbleeds	0.98 (0.67–1.43)	0.95 (0.72–1.25)
Lacunes	1.34 (0.83–2.16)	2.15 (1.60–2.87)
Large cortical infarcts	5.27 (1.50–18.50)	3.69 (1.77–7.72)
WMH (log)*	1.15 (0.91–1.46)	1.07 (0.90–1.26)
Total brain volume, per SD increase*	0.99 (0.98–0.99)	0.98 (0.97–0.98)
Hippocampal volume, per SD increase*	1.05 (0.84–1.31)	1.00 (0.85–1.18)

All possible risk factors are added together in the model. APOE indicates apolipoprotein E; BMI, body mass index; CMI, cortical cerebral microinfarct; MRI, magnetic resonance imaging; OR, odds ratio; RR, rate ratio; and WMH, white matter hyperintensities.

*Adjusted for intracranial volume.

of cortical CMIs were reported at 57% for cerebral amyloid angiopathy patients,[17] 32% in memory-clinic patients[6] and at 14% in patients with stroke.[7] In a subsample of population-based study (EDIS [Epidemiology of Dementia in Singapore]), cortical CMIs were present at a frequency of 6.2%.[4] In the present study, we found a prevalence of 10.5% on 1.5T MRI, higher than the previous observation in a population-based setting using 3T. This higher prevalence might be due to differences in image resolution with T1-weighted images having a voxel size of 0.8 mm in RS[8] compared with 1 mm in EDIS study.[4] Given that in our study a lower magnetic field strength MRI was used to identify these lesions, a prevalence of 10.5% indicates that these lesions occur at a much higher rate even in a population-based setting.

The most important associated risk factors for cortical CMIs were smoking, heart disease, history of stroke, infarcts, and smaller brain volume. These findings suggest an association of cortical CMIs with vascular disease independent of traditional vascular risk factors and other MRI markers of CeVD and neurodegeneration. Recent data has reported that subclinical cardiac dysfunction and clinically manifest cardiac diseases such as myocardial infarction, atrial fibrillation, and congestive cardiac failure may contribute to the development of cortical CMIs in the brain suggesting an origin from large vessel disease.[18] Of note, it has been previously suggested that cortical CMIs represent both ischemic and hemorrhagic mechanisms due to its link with cerebral microbleeds and infarcts.[4] Our findings, on the contrary, suggest that cortical CMIs represent more of an ischemic damage rather than hemorrhagic but this may require further confirmation.

Interestingly, cortical CMIs are suggested to have a heterogeneous cause due to its link with both small (lacunes, cerebral microbleeds, and WMH) and large vessel disease (large infarcts and intracranial stenosis).[4] Though we observed a possible association between

Table 3. Association of Cortical CMIs With Cognitive Decline

CMIs counts	LDST β (95% CI), *P* value	Stroop test 1 β (95% CI), *P* value	Stroop test 2 β (95% CI), *P* value	Stroop test 3 β (95% CI), *P* value	Word fluency test β (95% CI), *P* value	Immediate recall β (95% CI), *P* value	Delayed recall β (95% CI), *P* value	Recognition β (95% CI), *P* value	Perdue peg board test β (95% CI), *P* value
Model I	−0.29 (−0.48 to −0.09), *P*=0.004	0.14 (0.03 to 0.25), *P*=0.013	0.24 (0.09 to 0.38), *P*=0.001*	1.64 (0.51 to 2.77), *P*=0.004*	−0.33 (−0.50 to −0.15), *P*≤0.001	−0.05 (−0.13 to 0.03), *P*=0.230	−0.01 (−0.09 to 0.07), *P*=0.750	0.01 (−0.05 to 0.09), *P*=0.585	−0.23 (−0.38 to −0.08), *P*=0.002
P value†	*P*=0.663	*P*=0.973	*P*=0.003*	*P*=0.022	*P*=0.511	*P*=0.503	*P*=0.284	*P*=0.520	*P*=0.143
Model II	−0.24 (−0.42 to −0.06), *P*=0.011	0.082 (−0.04 to 0.21), *P*=0.189	0.18 (0.04 to 0.33), *P*=0.002*	1.74 (0.66 to 2.82), *P*=0.002*	−0.21 (−0.35 to −0.07), *P*=0.004	−0.02 (−0.10 to 0.07), *P*=0.713	0.02 (−0.08 to 0.11), *P*=0.693	0.02 (−0.08 to 0.11), *P*=0.700	−0.21 (−0.40 to −0.02), *P*=0.032
P value†	*P*=0.192	*P*=0.246	*P*≤0.001*	*P*≤0.001*	*P*=0.668	*P*=0.557	*P*=0.044	*P*=0.897	*P*=0.192

Model I: age, sex, education, history of heart disease, smoking, hypertension, hyperlipidemia, and diabetes. Model II: model I+MRI markers. CMI indicates cortical cerebral microinfarct; LDST, Letter Digit Symbol Test; and MRI, magnetic resonance imaging.

*Significant after Bonferroni coarrection *P*<0.005.

†P value for the interaction between CMI and time.

MRI markers of CeVD and brain tissue volumes in age- and-sex adjusted models, these associations were attenuated in the final adjusted model except for lacunes, large cortical infarcts, and total brain volume. Previous research in mice has shown microscopic cortical injury beyond the CMI core,[19,20] with one study estimating that

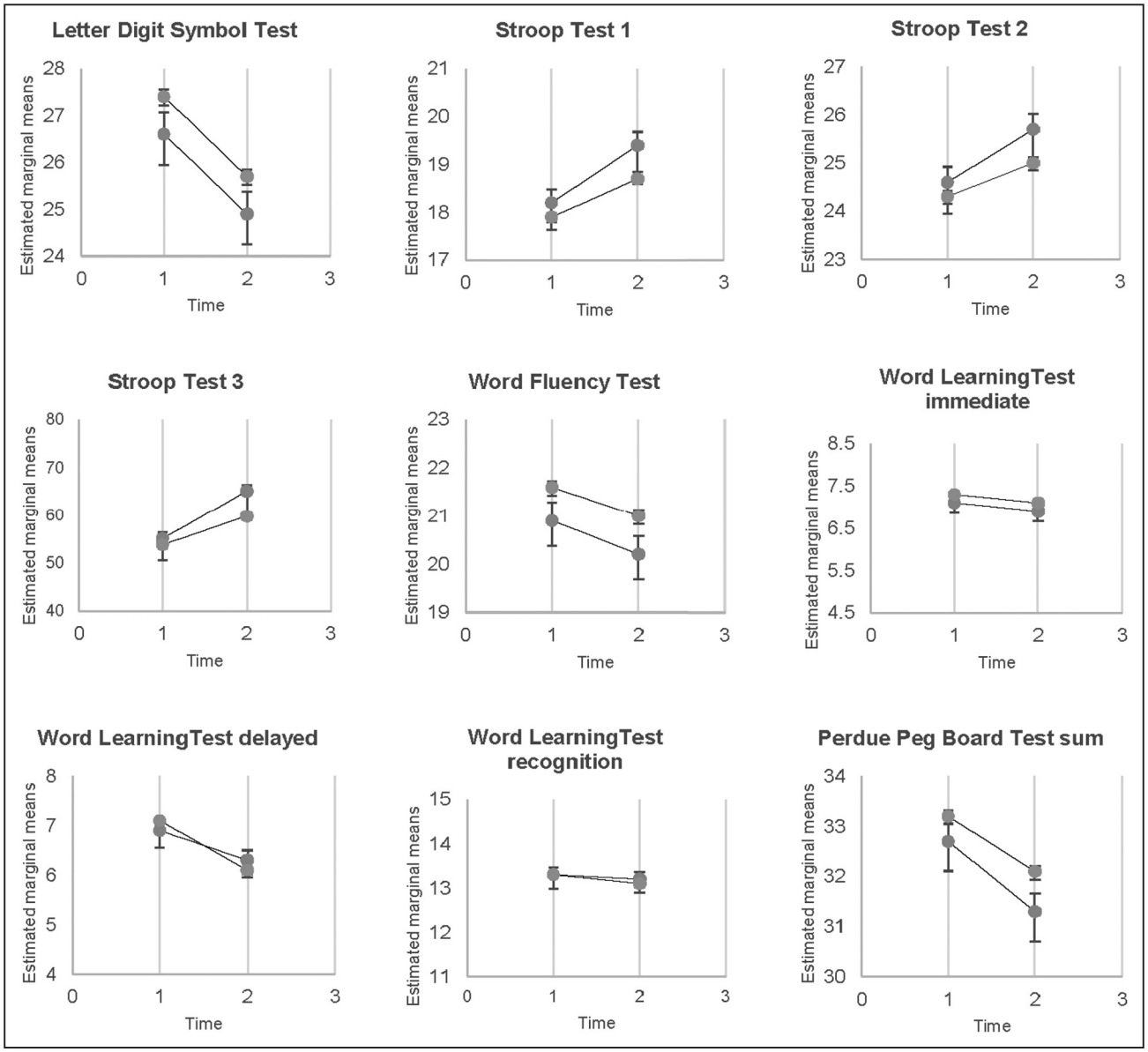

Figure 2. Mean cognitive functioning at baseline and follow-up visits, stratified by presence and absence of cortical cerebral microinfarcts (CMIs).

Table 4. Association Between Cortical CMIs and Clinical Outcomes

	Hazard ratios (95% CI)		
	Stroke (n=73)	Dementia (n=95)	Mortality (n=399)
Presence of CMIs			
Model I	2.19 (1.22–3.95)	0.83 (0.42–1.62)	1.37 (1.14–1.66)
Model II	1.99 (0.98–4.05)	1.59 (0.81–3.14)	1.21 (1.02–1.52)
CMI counts			
Model I	1.23 (1.15–1.31)	0.81 (0.53–1.24)	1.35 (1.12–1.64)
Model II	1.18 (1.0–1.28)	1.0 (0.74–1.35)	1.11 (1.01–1.22)

Model I: age, sex, education, history of heart disease, smoking, hypertension, hyperlipidemia, and diabetes. Model II: model I+MRI markers. CMI indicates cortical cerebral microinfarct.

this perilesional zone was 12-fold greater than the CMI core itself. The perilesional zone displayed several histopathologic alterations, including neuronal death, dendritic spine loss, astrogliosis, and blood-brain-barrier leakage.[19–21] It is suggested that CMIs directly incur this damage through ischemic cortical spreading depression and disrupted cortico-cortical and cortico-subcortical circuits.[20] It is possible that CMIs represent a marker of generalized vascular pathology in the brain rather than specific small vessel pathology.

In our study, cortical CMIs were also associated with cognitive decline particularly on Stroop test representing executive function. Limited data have shown that cortical CMIs were associated with worse performance on memory and language over a 2-year follow-up in a memory-clinic setting.[5] EDIS has further shown that cortical CMIs were associated with poorer memory and executive function in a cross-sectional analysis. The mechanisms by which cortical CMIs affect cognition remain unexplored and may be causal or noncausal. Cortical CMIs located strategically in brain regions may cause focal damage as well as concomitant microstructural damage of the surrounding tissue.[20] It must be acknowledged that the presence of a single cortical CMI on imaging or microscopy likely reflects hundreds and thousands of CMIs in the rest of the brain leading to disruption of neuronal tracts and thus interference with specific cognitive domains.[2] On the contrary, cortical CMIs are also suggested to be a proxy for underlying CeVD, and their higher numbers may indicate more extensive and severe microvascular damage.[3]

Finally, cortical CMIs at baseline were associated with an increased risk of stroke and mortality independent of vascular risk factors and MRI markers. No previous studies have examined the risk of stroke, dementia, and mortality in persons with cortical CMIs. Previous longitudinal studies have shown that chronic silent infarcts (without symptoms) increase the risk of further strokes and mortality by 2- to 4-fold independent of cardiovascular risk factors.[22,23] Our study adds further to the clinical relevance of cortical CMIs by demonstrating that the group with cortical CMIs at baseline (representing chronic ischemia) had an increased risk of subsequent overt strokes and death, although the number of events was small overall. In this study, we did not report an association between cortical CMIs and incident dementia. This might be explained by the fact that many individuals had died in this study and did not survive long enough to develop dementia. Hence, an opposite effect was observed (although nonsignificant) in the survival analysis.

Limitations of the study include; first, due to lower field strength and resolution of 1.5T MRI compared with 3T and 7T, small cortical CMIs <2 to 3 mm might have gone undetected. Hence, it is likely that the true prevalence of these lesions is higher, and the effect sizes between the associations may have been larger. Second, the cortical CMIs may reflect other vascular pathology beyond systemic vascular risk factors, and hence an association was still observed with incident stroke after adjusting for standard vascular risk factors. Third, a considerable proportion of participants (24%) did not had cognitive assessment at follow-up and hence were not included in cognitive decline analysis which may lead to selection bias and underestimation of effect sizes. Fourth, as this study had relatively short follow-up, the incidence of dementia might be underestimated. Finally, though we corrected for all known confounders, some residual confounding still remains from other factors such as inflammation and renal impairment. Strengths of the study include prospective population-based study design, virtually complete information on dates of clinical outcomes and use of medical records to ascertain their diagnosis, and to establish whether the CMI lesions developed before stroke.

CONCLUSIONS

Our study provides evidence that the cortical CMIs reflect measurable vascular brain damage and are related to cognitive decline, incident stroke, and mortality. Identification of cortical CMIs on 1.5T MRI have made it possible to study the clinical relevance and consequences of these lesions in a large population-based setting where 1.5T remains the main imaging modality. Though the possible effect of cortical CMIs on dementia risk remains under debate, our findings nonetheless do suggest that cortical CMIs may be a useful biomarker to intervene upon to improve cognitive functioning and reduce the burden of stroke.

ARTICLE INFORMATION

Received August 2, 2020; final revision received October 8, 2020; accepted November 18, 2020.

Affiliations

Department of Radiology and Nuclear Medicine (S.H., M.A.I., M.W.V.), Department of Epidemiology (S.H., A.D., M.K.I., M.A.I., M.W.V.), Departments of Radiology and Medical Informatics (H.V.), and Department of Neurology (M.K.I.), Erasmus Medical Center, Rotterdam, the Netherlands. Saw Swee Hock School of Public Health, National University of Singapore and National University Health System (S.H.).

Acknowledgments

Made to the donors of the Alzheimer's disease research, a program of the BrightFocus Foundation, for support of this research.

Sources of Funding

This work is supported by BrightFocus foundation (reference No. A2018165F) awarded to Dr Hilal. The RS (Rotterdam Study) is supported by the Erasmus Medical Center and Erasmus University Rotterdam; the Netherlands Organization for Scientific Research; the Netherlands Organization for Health Research and Development (ZonMw); Research Institute for Diseases in the Elderly; Ministry of Education, Culture and Science; Ministry of Health, Welfare and Sports; European Commission; and Municipality of Rotterdam.

Disclosures

Dr Hilal received research support from BrightFocus Foundation. The other authors report no conflicts.

Supplemental Materials

Expanded Methods
Figure I
Tables I–III

REFERENCES

1. van Veluw SJ, Shih AY, Smith EE, Chen C, Schneider JA, Wardlaw JM, Greenberg SM, Biessels GJ. Detection, risk factors, and functional consequences of cerebral microinfarcts. *Lancet Neurol*. 2017;16:730–740. doi: 10.1016/S1474-4422(17)30196-5
2. Westover MB, Bianchi MT, Yang C, Schneider JA, Greenberg SM. Estimating cerebral microinfarct burden from autopsy samples. *Neurology*. 2013;80:1365–1369. doi: 10.1212/WNL.0b013e31828c2f52
3. Brundel M, de Bresser J, van Dillen JJ, Kappelle LJ, Biessels GJ. Cerebral microinfarcts: a systematic review of neuropathological studies. *J Cereb Blood Flow Metab*. 2012;32:425–436. doi: 10.1038/jcbfm.2011.200
4. Hilal S, Sikking E, Shaik MA, Chan QL, van Veluw SJ, Vrooman H, Cheng CY, Sabanayagam C, Cheung CY, Wong TY, et al. Cortical cerebral microinfarcts on 3T MRI: A novel marker of cerebrovascular disease. *Neurology*. 2016;87:1583–1590. doi: 10.1212/WNL.0000000000003110
5. Hilal S, Tan CS, van Veluw SJ, Xu X, Vrooman H, Tan BY, Venketasubramanian N, Biessels GJ, Chen C. Cortical cerebral microinfarcts predict cognitive decline in memory clinic patients. *J Cereb Blood Flow Metab*. 2020;40:44–53. doi: 10.1177/0271678X19835565
6. van Veluw SJ, Hilal S, Kuijf HJ, Ikram MK, Xin X, Yeow TB, Venketasubramanian N, Biessels GJ, Chen C. Cortical microinfarcts on 3T MRI: Clinical correlates in memory-clinic patients. *Alzheimers Dement*. 2015;11:1500–1509. doi: 10.1016/j.jalz.2014.12.010
7. Wang Z, van Veluw SJ, Wong A, Liu W, Shi L, Yang J, Xiong Y, Lau A, Biessels GJ, Mok VC. Risk factors and cognitive relevance of cortical cerebral microinfarcts in patients with ischemic stroke or transient ischemic attack. *Stroke*. 2016;47:2450–2455. doi: 10.1161/STROKEAHA.115.012278
8. Ikram MA, van der Lugt A, Niessen WJ, Koudstaal PJ, Krestin GP, Hofman A, Bos D, Vernooij MW. The Rotterdam Scan Study: design update 2016 and main findings. *Eur J Epidemiol*. 2015;30:1299–1315. doi: 10.1007/s10654-015-0105-7
9. Wardlaw JM, Smith EE, Biessels GJ, Cordonnier C, Fazekas F, Frayne R, Lindley RI, O'Brien JT, Barkhof F, Benavente OR, et al; STandards for ReportIng Vascular changes on nEuroimaging (STRIVE v1). Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. *Lancet Neurol*. 2013;12:822–838. doi: 10.1016/S1474-4422(13)70124-8
10. Akoudad S, Wolters FJ, Viswanathan A, de Bruijn RF, van der Lugt A, Hofman A, Koudstaal PJ, Ikram MA, Vernooij MW. Association of cerebral microbleeds with cognitive decline and dementia. *JAMA Neurol*. 2016;73:934–943. doi: 10.1001/jamaneurol.2016.1017
11. Vrooman HA, Cocosco CA, van der Lijn F, Stokking R, Ikram MA, Vernooij MW, Breteler MM, Niessen WJ. Multi-spectral brain tissue segmentation using automatically trained k-Nearest-Neighbor classification. *Neuroimage*. 2007;37:71–81. doi: 10.1016/j.neuroimage.2007.05.018
12. de Boer R, Vrooman HA, Ikram MA, Vernooij MW, Breteler MM, van der Lugt A, Niessen WJ. Accuracy and reproducibility study of automatic MRI brain tissue segmentation methods. *Neuroimage*. 2010;51:1047–1056. doi: 10.1016/j.neuroimage.2010.03.012
13. Fischl B, Salat DH, Busa E, Albert M, Dieterich M, Haselgrove C, van der Kouwe A, Killiany R, Kennedy D, Klaveness S, et al. Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain. *Neuron*. 2002;33:341–355. doi: 10.1016/s0896-6273(02)00569-x
14. Hilal S, Baaij LGA, de Groot M, Niessen WJ, Ikram MK, Ikram MA, Vernooij MW. Prevalence and clinical relevance of diffusion-weighted imaging lesions: The Rotterdam study. *Neurology*. 2019;93:e1058–e1067. doi: 10.1212/WNL.0000000000008090
15. Prins ND, van Dijk EJ, den Heijer T, Vermeer SE, Jolles J, Koudstaal PJ, Hofman A, Breteler MM. Cerebral small-vessel disease and decline in information processing speed, executive function and memory. *Brain*. 2005;128(pt 9):2034–2041. doi: 10.1093/brain/awh553
16. de Bruijn RF, Bos MJ, Portegies ML, Hofman A, Franco OH, Koudstaal PJ, Ikram MA. The potential for prevention of dementia across two decades: the prospective, population-based Rotterdam Study. *BMC Med*. 2015;13:132. doi: 10.1186/s12916-015-0377-5
17. van den Brink H, Zwiers A, Switzer AR, Charlton A, McCreary CR, Goodyear BG, Frayne R, Biessels GJ, Smith EE. Cortical microinfarcts on 3T magnetic resonance imaging in cerebral amyloid angiopathy. *Stroke*. 2018;49:1899–1905. doi: 10.1161/STROKEAHA.118.020810
18. Hilal S, Chai YL, van Veluw S, Shaik MA, Ikram MK, Venketasubramanian N, Richards AM, Biessels GJ, Chen C. Association between subclinical cardiac biomarkers and clinically manifest cardiac diseases with cortical cerebral microinfarcts. *JAMA Neurol*. 2017;74:403–410. doi: 10.1001/jamaneurol.2016.5335
19. Coban H, Tung S, Yoo B, Vinters HV, Hinman JD. Molecular disorganization of axons adjacent to human cortical microinfarcts. *Front Neurol*. 2017;8:405. doi: 10.3389/fneur.2017.00405
20. Summers PM, Hartmann DA, Hui ES, Nie X, Deardorff RL, McKinnon ET, Helpern JA, Jensen JH, Shih AY. Functional deficits induced by cortical microinfarcts. *J Cereb Blood Flow Metab*. 2017;37:3599–3614. doi: 10.1177/0271678X16685573
21. Silasi G, She J, Boyd JD, Xue S, Murphy TH. A mouse model of small-vessel disease that produces brain-wide-identified microocclusions and regionally selective neuronal injury. *J Cereb Blood Flow Metab*. 2015;35:734–738. doi: 10.1038/jcbfm.2015.8
22. Bernick C, Kuller L, Dulberg C, Longstreth WT Jr, Manolio T, Beauchamp N, Price T; Cardiovascular Health Study Collaborative Reseach Group. Silent MRI infarcts and the risk of future stroke: the cardiovascular health study. *Neurology*. 2001;57:1222–1229. doi: 10.1212/wnl.57.7.1222
23. Vermeer SE, Hollander M, van Dijk EJ, Hofman A, Koudstaal PJ, Breteler MM; Rotterdam Scan Study. Silent brain infarcts and white matter lesions increase stroke risk in the general population: the Rotterdam Scan Study. *Stroke*. 2003;34:1126–1129. doi: 10.1161/01.STR.0000068408.82115.D2