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Retinal microvascular abnormalities and subclinical magnetic resonance imaging brain infarct: a prospective study

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Silent brain infarct and white matter lesions are common radiological findings associated with the risk of clinical stroke and dementia; however, our understanding of their underlying pathophysiology and risk factors remains limited. This study aimed to determine whether assessment of retinal microvascular abnormalities could provide prognostic information regarding the risk of brain infarct and white matter lesions on magnetic resonance imaging. This study is based on a subset of 810 middle-aged persons without clinical stroke or baseline magnetic resonance imaging infarct enrolled in the Atherosclerosis Risk in Communities Brain Magnetic Resonance Imaging Study, a prospective, population-based study. Participants had a baseline magnetic resonance imaging brain examination and retinal photography in 1993–1995, and returned for a repeat magnetic resonance imaging examination in 2004–2006. Magnetic resonance images were graded for presence of any cerebral infarct, infarct with lacunar characteristics and white matter lesions according to standardized protocols. Retinal photographs were graded for presence of retinopathy lesions and retinal arteriolar abnormalities following a standardized protocol. Over a median follow-up of 10.5 years, 164 (20.2%) participants developed cerebral infarct, 131 (16.2%) developed lacunar infarct, 182 (24.2%) developed new white matter lesions and 49 (6.1%) had evidence of white matter lesion progression. After adjusting for age, gender, race, cardiovascular risk factors and carotid intima-media thickness, retinopathy was associated with incident cerebral infarct (odds ratio 2.82; 95% confidence interval 1.42–5.60) and lacunar infarct (odds ratio 3.19; 95% confidence interval: 1.56–6.50). Retinal arteriovenous nicking was associated with incident cerebral infarct (odds ratio 2.82; 95% confidence interval: 1.66–4.76), lacunar infarct (odds ratio 2.48; 95% confidence interval: 1.39–4.40) and white matter lesion

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incidence (odds ratio 2.12; 95% confidence interval: 1.18–3.81) and progression (odds ratio 2.22; 95% confidence interval: 1.00–5.88). In conclusion, retinal microvascular abnormalities are associated with emergence of subclinical magnetic resonance imaging brain infarcts and white matter lesions, independent of shared risk factors. Retinal vascular imaging may offer a non-invasive tool to investigate the pathogenesis and natural history of cerebral small-vessel disease.

Keywords: cerebral infarction; cerebral ischaemia; epidemiology; retina; stroke

Abbreviations: ARIC = Atherosclerosis Risk in Communities study; OR = odds ratio; WML = white matter lesion

Introduction

Silent brain infarct is a common incidental finding on MRI (Vermeer *et al.*, 2007; Vernooij *et al.*, 2007). It is associated with subtle neurological deficits in physical and cognitive function, and its presence doubles the risk of future clinical stroke and dementia (Vermeer *et al.*, 2007).

However, our knowledge about the pathophysiology of and risk predictors for silent brain infarct remains limited. Although cerebral small-vessel disease may play a key pathogenic role (Greenberg, 2006; Vermeer *et al.*, 2007), testing and gaining more insights into such a hypothesis is challenging, due to the paucity of non-invasive tools that assess the cerebral microvasculature objectively and reliably.

The retina offers a unique, non-invasive and easily accessible window to study the microvascular aetiology of cerebrovascular disease (Wong, 2004; Ikram *et al.*, 2006). Retinal and cerebral small vessels share similar embryological origin, anatomical features and physiological properties (Baker *et al.*, 2007). Pathological changes in the retinal vasculature may therefore reflect similar microangiopathic processes occurring in the brain that predispose people to develop brain infarct. In support of this concept, cross-sectional MRI studies have demonstrated independent associations between retinal microvascular abnormalities and ischaemic cerebrovascular lesions, including cerebral infarcts and white matter lesions (WMLs) (Kwa *et al.*, 2002; Wong *et al.*, 2002; Cooper *et al.*, 2006; Longstreth *et al.*, 2007; Qiu *et al.*, 2008; Lindley *et al.*, 2009). To date, however, there are a lack of prospective data to verify the temporality of these associations. Thus, it remains uncertain whether retinal vascular imaging and assessment could in fact provide predictive information on the risk of silent brain infarct above and beyond traditional cerebrovascular risk factors.

In this study, we examined the relationship of retinal microvascular abnormalities to 10-year incidence of ischaemic cerebrovascular changes on MRI (brain infarcts and WMLs) in middle-aged individuals without clinical stroke.

Materials and methods

Study population

The Atherosclerosis Risk in Communities Study (ARIC) is a population-based study of cardiovascular disease among 15 792 middle-aged persons, selected from four US communities, who were first examined in 1987–1989 (The ARIC Investigators, 1989, Wong *et al.*, 2001, 2002). Participants underwent a second examination 3 years later in 1990–1992 (93% return rate) and a third examination in 1993–1995 (86% return rate).

At the third ARIC examination, participants 55 years and older at two study sites (Forsyth County, North Carolina and Jackson, Mississippi) were invited to participate in a brain MRI study. A total of 2891 participants were screened for willingness to participate and exclusions were applied for safety reasons (prior surgery for an aneurysm in the brain; metal fragments in the eyes, brain or spinal cord; valvular prosthesis, cardiac pacemaker, cochlear implant, spinal-cord stimulator or other internal electrical device; pregnancy; and occupations associated with exposure to metal fragments) (Knopman *et al.*, 2009). Of these, 1920 participants underwent brain MRI and had scans of sufficient quality for analysis. Retinal photography was also performed at the third examination. Of the 1684 participants who had complete data from brain MRI and retinal examinations at the third visit, 1031 (61%) had a repeat MRI examination at the fifth visit in 2004–2006 (median follow-up of 10.5 years). Of these 1031 participants, we excluded those with a history of stroke ($n = 25$) or transient ischemic attack ($n = 18$), those with cerebral infarct on MRI at the third examination ($n = 64$), and those with incomplete retinal or MRI data ($n = 84$). We further excluded those who developed clinical stroke between the first and the follow-up MRI examinations ($n = 30$), leaving 810 participants for the current analyses. Compared with the excluded participants, the included participants generally had a better cardiovascular risk profile (Supplementary Table 1).

Institutional Review Boards at each study site, the MRI reading centre and the University of Wisconsin Fundus Photograph Reading Centre approved the study. Informed consent was obtained from all participants.

Cerebral MRI examination and definitions

Cerebral MRI scanning and image interpretation have been described (Wong *et al.*, 2002, 2003; Cooper *et al.*, 2006). In brief, T_1- and T_2-weighted images were obtained. Axial images were angled to be parallel to the anterior commissure–posterior commissure line. Most of the scans were done on 1.5 T GE scanners. The follow-up scanners and scan parameters were chosen to match best those of the initial scans. Trained and certified MRI readers, who were masked to participants' clinical condition and retinal findings, evaluated the digitized scan data on a personal display workstation at the MRI reading centre. Quality control procedures and reliability data for all MRI grading have been described (Wong *et al.*, 2002, 2003; Cooper *et al.*, 2006).

Infarct-like lesions were defined based on signal characteristics on the T_1, T_2 and proton-density images. In general, these lesions were defined as bright on T_2 and proton-density but dark on T_1 images.

These features distinguished them from WMLs (not dark on T_1) and Virchow–Robin spaces (not bright on proton-density). In the basal ganglia and brainstem, lesions not dark on T_1 were also defined as infarcts.

Cerebral infarct was defined as any lesion ≥3 mm in maximum diameter in a vascular distribution with typical imaging characteristics of infarction and was distinguished from the smaller, more common cerebral WMLs. Lacunar infarct was defined as a subset of those infarcts which were <20 mm in maximum diameter and located in the basal ganglia, internal capsule, thalamus or deep cerebral white matter. A total of 104 randomly selected cases were re-read to assess intergrader agreement, which was 89%.

The spin-density images (repetition time, 3000 ms; echo time, 30 s) were used to estimate the overall volume of periventricular and subcortical WMLs. These were coded on a scale from 0 to 9, based on 'pattern matching' of a scan to a set of reference standards, which are described elsewhere (Wong *et al.*, 2002). We defined 'significant WML' as grade 3 or higher and 'little or no WML' as grade 2 or lower (cut-off corresponds to the 90th percentile of WML scores in the sample) (Wong *et al.*, 2002). 'Incident WML' was defined as presence of significant WML at the follow-up MRI examination with little or no WML at the first MRI examination. 'WML progression' was defined as at least two-step increase in WML grades at the follow-up MRI examination in participants with significant WML at the first MRI examination. A total of 97 randomly selected cases were re-read to assess the reproducibility in the detection of WML progression, and the inter-reader kappa was 0.48.

Retinal photography and assessment

Retinal photography procedures have been described in detail elsewhere (Hubbard *et al.*, 1999, Wong *et al.*, 2001, 2002, 2003). In brief, a retinal photograph of one randomly selected eye was taken after 5 min of dark adaptation through a non-pharmacologically dilated pupil. Trained graders at the Fundus Photograph Reading Centre who were masked to participant characteristics evaluated the photographic slides for presence of microvascular abnormalities using a standardized protocol (Hubbard *et al.*, 1999). Retinopathy was defined as present if any of the following lesions were detected: retinal microaneurysms, haemorrhages, soft exudates and other less common lesions (e.g. hard exudates, macular oedema, optic disc swelling). Retinal arteriovenous nicking and focal arteriolar narrowing were separately defined as present if graded definite or probable. Quality control procedures and reliability data for all retinal grading have been reported (Hubbard *et al.*, 1999).

Assessment of cardiovascular risk factors

Participants underwent standardized interviews, clinical examinations and laboratory investigation for the assessment of cardiovascular risk factors at all visits (Wong *et al.*, 2001, 2002, 2003; Cooper *et al.*, 2006). Mean arterial blood pressure was computed as two thirds of the diastolic plus one third of the systolic value, and the average of this over the first three examinations (6-year mean arterial blood pressure) was used as a covariate for adjustment of blood pressure. Hypertension was defined as systolic blood pressure of 140 mmHg or greater, diastolic blood pressure of 90 mmHg or greater or use of antihypertensive medication during the previous 2 weeks. Diabetes mellitus was defined as fasting glucose level of at least 126 mg/dl (7·0 mmol/l), a non-fasting glucose level of at least 200 mg/dl (11·1 mmol/l) or a self-reported history of physician-diagnosed diabetes or treatment for diabetes. Average internal carotid intima-media wall thickness was measured using B-mode ultrasonography. Collection of fasting blood samples and assays of total cholesterol and glucose levels were also performed. All risk factor measurements were from the third examination (considered baseline in this study), except for 6-year mean arterial blood pressure.

Statistical analysis

Retinal vascular signs were analysed as categorical variables. We compared baseline characteristics between included and excluded participants, and also between participants with and without retinopathy or retinal arteriovenous nicking using chi-square test or independent sample *t*-tests. Follow-up time was defined as the time from the third examination to the date of repeat brain MRI at the fifth examination. We estimated the 10-year cumulative incidence of MRI brain abnormalities and constructed logistic regression models to determine the odds ratio (OR) for incident MRI brain abnormalities in relation to retinal vascular signs, initially adjusted for age, gender, race and study centre, and further adjusted for other vascular risk factors. Finally, we performed stratified analyses by hypertension and diabetes status. All analyses were performed using Statistical Package for the Social Sciences version 16.0.1 (SPSS Inc, Chicago, Ill).

Results

Table 1 shows that, compared to participants without retinopathy, those with retinopathy were more often African-American and hypertensive or diabetic, and had higher systolic blood pressure and fasting glucose and greater carotid thickness. Participants with arteriovenous nicking were more likely to be smokers, less likely to be high school graduates and had higher blood pressure levels than those without arteriovenous nicking.

Over a median follow-up of 10.5 years (inter-quartile range 9 months), 164 (20.2%) participants developed MRI evidence of cerebral infarct, 131 (16.2%) developed lacunar infarct, 182 (24.2) developed WMLs and 49 (6.1%) demonstrated progression of WMLs.

Table 2 shows that any retinopathy (and some of its component lesions) and retinal arteriovenous nicking were associated with increased odds of incident cerebral infarct (OR 2.82 for retinopathy and for arteriovenous nicking) and lacunar infarct (OR 3.19 for retinopathy and OR 2.48 for arteriovenous nicking) after adjusting for age, gender, race, carotid intima-media wall thickness, coronary heart disease and other cardiovascular risk factors, including cigarette smoking, 6-year mean arterial blood pressure, anti-hypertensive medication, fasting glucose and total cholesterol. Table 3 shows that in multivariate analysis, only retinal arteriovenous nicking was associated with incident WMLs (OR 2.12) and progression of WMLs (OR 2.22) in multivariate analysis.

In supplementary analysis (Supplementary Table 2), the associations of retinopathy with incident cerebral and lacunar infarct were stronger in participants with diabetes and in those without hypertension. Additionally, retinopathy was significantly associated with incidence and progression of WML in participants without diabetes. Conversely, the associations of retinal arteriovenous nicking with incident cerebral infarct and lacunar infarct were stronger

Table 1 Baseline (1993–95) characteristics of participants in the ARIC brain MRI study

	All	No retinopathy $n=760$	Retinopathy $n=50$	P^a	No arteriovenous nicking $n=697$	Arteriovenous nicking $n=100$	P^a
Race, white	427 (52.5)	407 (53.7)	19 (35.8)	0.01	373 (53.7)	43 (43.0)	0.05
Gender, male	326 (40.1)	301 (39.7)	25 (47.2)	0.28	270 (38.8)	49 (49.0)	0.05
Hypertension	337 (41.6)	303 (40.1)	33 (62.3)	<0.01	284 (41.0)	47 (47.0)	0.25
Diabetes	111 (13.7)	89 (11.8)	22 (41.5)	<0.01	93 (13.4)	16 (16.5)	0.41
Cigarette smoker, current	115 (14.2)	106 (14.0)	9 (17.0)	0.55	90 (13.0)	22 (22.2)	0.01
High school graduate	641 (78.8)	604 (79.7)	35 (66.0)	0.02	555 (79.9)	71 (71.0)	0.04
Age, years	61.6 (4.2)	61.7 (4.2)	60.9 (4.0)	0.20	61.5 (4.2)	61.9 (4.1)	0.38
Systolic blood pressure, mmHg	125.5 (18.8)	125.0 (18.5)	132.4 (21.6)	0.01	124.6 (18.5)	131.0 (20.7)	<0.01
Diastolic blood pressure, mmHg	72.4 (10.4)	72.4 (10.4)	72.9 (11.6)	0.71	72.2 (10·,)	74.9 (9.8)	0.01
Fasting glucose, mg/dl	109.6 (38.9)	106.7 (32.1)	151.7 (81.6)	<0.01	110.1 (41.1)	107.7 (22.4)	0.58
Total cholesterol, mg/dl	209.7 (38.2)	209.5 (38.2)	212.9 (37.1)	0.52	210.3 (38.3)	204.9 (37.1)	0.19
HDL cholesterol, mg/dl	55.5 (18.9)	55.9 (19.0)	50.1 (17.1)	0.03	55.6 (19.1)	54.6 (17.0)	0.63
Triglyceride, mg/dl	132.2 (75.1)	131.7 (75.7)	139.6 (67.7)	0.46	132.5 (76.3)	132.5 (69.5)	0.99
Carotid intima-media thickness, mm	0.75 (0.18)	0.75 (0.18)	0.83 (0.21)	<0.01	0.75 (0.18)	0.78 (0.17)	0.18

Data are expressed as numbers (percentages) for categorical variables or means (standard deviations) for continuous variables. HDL = high-density lipoprotein.
a Based on chi-square (categorical) or independent sample *t*-tests, comparing characteristics between participants with and without retinopathy or arteriovenous nicking.

in participants with hypertension and in those without diabetes. However, we found no significant interaction ($P<0.10$) by adding cross-product terms of race, gender, diabetes or hypertension into the models of the studied population.

Discussion

In this population-based cohort of middle-aged persons without clinical stroke, retinal microvascular abnormalities measured at baseline were prospectively associated with long-term risk of subclinical cerebrovascular disease on MRI, independent of conventional risk factors. The presence of retinopathy signs was associated with more than 2-fold higher odds of cerebral infarct and 3-fold higher odds of lacunar infarct. The presence of retinal arteriovenous nicking was associated with more than 2-fold higher odds of not only brain infarcts but also with incidence and progression of WMLs. Associations were similar in people with and without diabetes and hypertension.

While several studies have reported associations of retinal microvascular changes with increased risk of clinical stroke events and stroke mortality (Wong *et al.*, 2001, 2002; Mitchell *et al.*, 2005; Cheung *et al.*, 2007; Cheung and Wong, 2008), we are unaware of any prospectively collected data for the associations of retinal microvascular abnormalities with incidence and progression of subclinical cerebrovascular disease. Cross-sectional studies suggest that people with retinopathy are more likely to have concomitant cerebral infarct, WMLs and microbleeds on MRI than those without retinopathy (Kwa *et al.*, 2002; Wong *et al.*, 2002; Cooper *et al.*, 2006; Longstreth *et al.*, 2007; Qiu *et al.*, 2008). Our current study extends these observations and offers new evidence that the presence of retinal microvascular abnormalities may predict the risk of ischaemic brain changes (cerebral infarct and WMLs) over a 10-year period. Furthermore, the stronger association of retinopathy with lacunar, compared to cerebral, infarct supports a key hypothesis that lacunar stroke is more specifically related to small-vessel non-atherothrombotic, rather than large-vessel atherothrombotic, disease, as reported recently in a cross-sectional study (Lindley *et al.*, 2009).

The mechanistic pathways that underlie our findings remain a focal point for future research. Here, we propose a possible explanation. Pathophysiological processes underlying diabetic and hypertensive retinopathy may mirror similar disease processes occurring in the brain that lead to the development of ischaemic cerebral small-vessel disease. It is generally accepted that a disruption of the blood–retinal barrier from retinal hypoxia caused by diabetes and hypertension plays a central role in the pathogenesis of retinal microangiopathy (Kaur *et al.*, 2008). The blood–retinal barrier could be considered analogous to the blood–brain barrier (Baker *et al.*, 2007). A breakdown of the blood–brain barrier might be a key pathogenic component leading to cerebral microangiopathy (Wardlaw *et al.*, 2001, 2003). Unlike the retinal circulation, however, the cerebral microcirculation cannot be easily, directly and non-invasively visualized *in vivo*. Early structural alterations in the cerebral microvasculature caused by blood–brain barrier dysfunction may remain masked until significant vasogenic oedema (which accompanies WMLs) and brain tissue damage (small infarcts) occur. This might explain why retinal microvascular abnormalities preceded the emergence of visible ischaemic cerebrovascular abnormalities on MRI. However, it is important to note that other pathophysiological mechanisms, such as those related to inflammation and endothelial dysfunction (Baker *et al.*, 2007), may also contribute to our findings. Additional studies are clearly needed to delineate these mechanisms further.

Another noteworthy observation is that the retinal signs were associated similarly with two different MRI signs of cerebral small-vessel disease. While the similarity is expected because of

Table 2 Retinal microvascular changes and incidence of MRI silent brain infarct

Retinal microvascular abnormalities		Incident silent cerebral infarct, $n = 164$ (20.2%)			Incident silent lacunar infarct, $n = 131$ (16.2%)		
		No. at risk (% with infarct)	Age–gender–race[a] OR (95% CI)[a]	Multivariable[b] OR (95% CI)[b]	No. at risk (% with infarct)	Age–gender–race[a] OR (95% CI)[a]	Multivariable[b] OR (95% CI)[b]
Any retinopathy	Absent	757 (19.0)	1.00	1.00	758 (14.8)	1.00	1.00
	Present	53 (37.7)	2·77 (1.52–5.06)	2.82 (1.42–5.60)	53 (35.8)	3.44 (1.86–6.35)	3.19 (1.56–6.50)
Microaneurysm	Absent	697 (19.1)	1.00	1.00	698 (14.6)	1.00	1.00
	Present	28 (39.3)	3.16 (1.41–7.09)	2.78 (1.10–8.89)	28 (35.7)	3.72 (1.62–8.53)	2.69 (1.01–7.18)
Retinal haemorrhage	Absent	749 (19.5)	1.00	1.00	750 (15.3)	1.00	1.00
	Present	22 (40.9)	2.99 (1.22–7.29)	3.46 (1.31–9.12)	22 (36.4)	3.20 (1.28–8.00)	3.25 (1.18–8.92)
Cotton wool spot	Absent	777 (19.7)	1.00	1.00	778 (15.6)	1.00	1.00
	Present	13 (46.2)	3.45 (1.11–10.69)	3.84 (1.07–13.81)	13 (38.5)	3.32 (1.04–10.60)	3.08 (0.79–12.01)
Arteriovenous nicking	Absent	694 (17.7)	1.00	1.00	695 (14.1)	1.00	1.00
	Present	100 (35.0)	2.60 (1.63–4.15)	2.82 (1.66–4.76)	100 (27.0)	2.34 (1.42–3.86)	2.48 (1.39–4.40)
Focal narrowing	Absent	664 (19.3)	1.00	1.00	664 (14.9)	1.00	1.00
	Present	111 (24.3)	1.29 (0.79–2.08)	1.02 (0.58–1.78)	112 (21.4)	1.52 (0.92–2.53)	1.13 (0.62–2.07)

a OR and 95% confidence interval (CI) adjusted for age, gender, race and study centre.
b Further adjusted for cigarette smoking, 6-year mean arteriolar blood pressure, anti-hypertensive medication, education, fasting glucose, total cholesterol, triglycerides and carotid intima-media thickness, and prevalent coronary heart disease.

Table 3 Retinal microvascular changes and incidence and progression of MRI WML

Retinal microvascular abnormalities		Incident WML, $n = 182$ (24.2%)			WML progression, $n = 49$ (6.1%)		
		No. at risk (% with infarct)	Age–gender–race[a] OR (95% CI)[a]	Multivariable[b] OR (95% CI)[b]	No. at risk (% with infarct)	Age–gender–race[a] OR (95% CI)[a]	Multivariable[b] OR (95% CI)[b]
Any retinopathy	Absent	706 (23.7)	1.00	1.00	748 (5.7)	1.00	1.00
	Present	53 (33.3)	1.68 (0.86–3.29)	1.54 (0.65–3.61)	53 (11.8)	1.78 (0.71–4.49)	1.18 (0.38–3.60)
Microaneurysm	Absent	649 (23.0)	1.00	1.00	689 (5.5)	1.00	1.00
	Present	28 (26.1)	1.23 (0.46–3.29)	0.71 (0.23–2.25)	26 (11.5)	1.67 (0.47–6.01)	1.02 (0.23–4.21)
Retinal haemorrhage	Absent	693 (22.8)	1.00	1.00	739 (5.5)	1.00	1.00
	Present	21 (33.3)	1.98 (0.76–5.16)	2.18 (0.69–6.87)	22 (13.6)	2.61 (0.71–9.54)	2.65 (0.63–11.12)
Cotton wool spot	Absent	722 (23.3)	1.00	1.00	766 (5.6)	1.00	1.00
	Present	13 (27.3)	1.03 (0.26–4.12)	0.58 (0.08–3.74)	13 (7.7)	0.97 (0.12–7.74)	0.72 (0.08–6.63)
Arteriovenous nicking	Absent	648 (21.8)	1.00	1.00	685 (5.3)	1.00	1.00
	Present	88 (38.6)	2.14 (1.32–3.47)	2.12 (1.18–3.81)	98 (11.2)	1.92 (0.92–3.99)	2.22 (1.00–5.88)
Focal narrowing	Absent	622 (22.8)	1.00	1.00	654 (6.3)	1.00	1.00
	Present	96 (28.1)	1.24 (0.75–2.04)	0.89 (0.46–1.67)	110 (4.5)	0.77 (0.29–2.02)	0.61 (0.19–1.93)

a OR and 95% confidence interval (CI) adjusted for age, gender, race and study centre.
b Further adjusted for cigarette smoking, 6-year mean arteriolar blood pressure, anti-hypertensive medication, education, fasting glucose, total cholesterol, triglycerides and carotid intima-media thickness, prevalent coronary heart disease and baseline WML grades (for WML progression).

shared pathogenesis, it may also be related to overlapping imaging features between lacunar infarcts and WMLs on MRI (e.g. a lacunar infarct lesions are not always distinguishable from WMLs in basal ganglia).

The strengths of this study include the prospective design, biracial community-based cohort and standardized and masked assessment of MRI scans, retinal photographs and cardiovascular risk factors. Limitations should also be noted. First, selection bias might have influenced our results. A significant proportion of participants were excluded from our analysis because participants did not return for repeat MRI examination or have gradable retinal photographs. The excluded participants in general were more likely to have a poorer cardiovascular profile (Supplementary Table 1). Thus, the associations observed here might have been attenuated by selective inclusion. Moreover, selection bias might also explain the relatively low incidence of clinical stroke ($n = 30$) in our cohort. Second, the infrequency of the retinal signs limited the precision of our risk estimates, and larger prospective studies may be required to provide more precise estimates. Third, the less robust inter-reader agreement for WML assessment (inter-reader kappa of 0.48) might explain the generally weaker associations between retinal signs and WMLs. Fourth, the MRI examinations were done almost 10 years apart. There had been interval upgrades in scanners that might allow more sensitive detection of cerebral abnormalities at the follow-up than the baseline MRI examination. However, this should affect all the participants equally and thus should not bias the reported associations with retinal signs. Finally, hypertension is a major risk factor shared by the MRI brain and retinal microvascular abnormalities. The confounding effect of blood pressure might be considerable. However, the associations remained significant despite adjusting for the average of blood pressure measurements on three separate occasions over a 6-year period and for the use of antihypertensive medications, and they were seen in persons without hypertension.

In summary, our data show that independent of cerebrovascular risk factors, the presence of retinal microvascular abnormalities may double or triple the odds of developing subclinical cerebral small-vessel disease on MRI over the next decade. These findings suggest that retinal microvascular abnormalities are early, and possibly more sensitive, markers of subclinical cerebral small-vessel disease before its radiological and clinical manifestations. Retinal vascular imaging may offer a surrogate platform for further investigation on the pathogenesis of and therapeutic strategies for cerebral small-vessel disease.

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Supplementary material

Supplementary material is available at *Brain* online.

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