

Rationale and design of a longitudinal study of cerebral small vessel diseases, clinical and imaging outcomes in patients presenting with mild ischaemic stroke: Mild Stroke Study 3

European Stroke Journal
2021, Vol. 6(1) 81–88
© European Stroke Organisation 2020



Article reuse guidelines:
sagepub.com/journals-permissions
DOI: 10.1177/2396987320929617
journals.sagepub.com/home/eso



Una Clancy , Daniela Jaime Garcia, Michael S Stringer, Michael J Thrippleton, Maria C Valdés-Hernández, Stewart Wiseman, Olivia KL Hamilton , Francesca M Chappell, Rosalind Brown, Gordon W Blair, Will Hewins, Emilie Sleight, Lucia Ballerini , Mark E Bastin, Susana Munoz Maniega, Tom MacGillivray, Kirstie Hetherington, Charlene Hamid, Carmen Arteaga, Alasdair G Morgan, Cameron Manning, Ellen Backhouse, Iona Hamilton, Dominic Job, Ian Marshall, Fergus N Doubal and Joanna M Wardlaw 

Abstract

Background: Cerebral small vessel disease is a major cause of dementia and stroke, visible on brain magnetic resonance imaging. Recent data suggest that small vessel disease lesions may be dynamic, damage extends into normal-appearing brain and microvascular dysfunctions include abnormal blood–brain barrier leakage, vasoreactivity and pulsatility, but much remains unknown regarding underlying pathophysiology, symptoms, clinical features and risk factors of small vessel disease.

Patients and Methods: The Mild Stroke Study 3 is a prospective observational cohort study to identify risk factors for and clinical implications of small vessel disease progression and regression among up to 300 adults with non-disabling stroke. We perform detailed serial clinical, cognitive, lifestyle, physiological, retinal and brain magnetic resonance imaging assessments over one year; we assess cerebrovascular reactivity, blood flow, pulsatility and blood–brain barrier leakage on magnetic resonance imaging at baseline; we follow up to four years by post and phone. The study is registered ISRCTN 12113543.

Summary: Factors which influence direction and rate of change of small vessel disease lesions are poorly understood. We investigate the role of small vessel dysfunction using advanced serial neuroimaging in a deeply phenotyped cohort to increase understanding of the natural history of small vessel disease, identify those at highest risk of early disease progression or regression and uncover novel targets for small vessel disease prevention and therapy.

Keywords

Cerebral small vessel diseases, lacunar stroke, white matter hyperintensities, magnetic resonance imaging, longitudinal studies, dementia, cognitive dysfunction, symptom assessment, blood–brain barrier, cerebrovascular circulation

Date received: 25 February 2020; accepted: 14 April 2020

Corresponding author:

Joanna Wardlaw, University of Edinburgh, Chancellor's Building, 49 Little
France Crescent, Edinburgh EH16 4SB, UK.
Email: joanna.wardlaw@ed.ac.uk

Background

Cerebral small vessel disease (SVD) describes diffuse disease processes affecting the perforating cerebral arterioles, capillaries, venules and consequent damage to the white and deep grey matter.¹ This damage is visible on brain MRI as white matter hyperintensities (WMH), recent small subcortical infarcts, perivascular spaces, brain atrophy and cerebral microbleeds.²

SVD causes 20% of ischaemic strokes and almost half of all dementias,^{3,4} contributing to both vascular and Alzheimer's dementia subtypes,⁵ its presence more than doubling future risk of stroke, dementia and functional impairment.⁶

Recent advances in neuroimaging have uncovered candidate mechanisms for underlying pathophysiological processes. Furthermore, SVD appears to be more dynamic and global than previously thought, since recent studies show: (a) WMH can regress as well as progress;^{7–10} (b) SVD is associated with cerebrovascular dysfunction including diffuse blood–brain barrier (BBB) failure;¹¹ (c) with some evidence for other vascular dysfunctions including reduced cerebrovascular reactivity (CVR) and increased intracranial pulsatility^{12–14} and (d) acute, apparently 'silent', lesions on diffusion weighted imaging (DWI) may be more frequent than previously thought.^{15–17}

Most SVD lesions are thought to develop 'silently'. However, some studies suggest that SVD lesions are associated with subjective cognitive complaints,¹⁸ gait disturbance,¹⁹ mood disorders and apathy.²⁰ Moreover, subtle symptoms have been associated with acute DWI lesions in a few small cross-sectional studies in non-stroke populations ($n = 6/649$; $n = 10/30$),^{21,22} while apparently 'silent' acute DWI lesions have been noted in up to 25% following recent stroke, mostly in small studies (e.g. $n < 105$) that sought typical stroke symptoms.^{16,23} Thus, knowledge of the extent of clinical correlates of SVD lesions, in particular, any 'red flag' symptoms or signs that might highlight lesion worsening, remains limited and may be being clinically overlooked.

SVD is commonly attributed to traditional vascular risk factors, particularly hypertension, but also smoking and diabetes, yet these factors only account for 2% of WMH variance.²⁴ Less is known about potential contributors such as diet, lifestyle and premorbid factors.²⁵ Extending the search beyond an individual's current clinical status to early and mid-life stages is an important target for SVD research.²⁶ Understanding whether combined risk factors have a synergistic effect on an individual's risk of developing SVD, as well as improved recognition of symptoms, would provide better recognition of persons at risk of SVD development or progression, providing insight on whether

multimodal approaches to prevention and treatment should be taken.

Few studies have comprehensively assessed SVD lesion progression, symptomatology and wide-ranging risk factors. Hence we describe the protocol for a detailed study to assess the role of cerebrovascular dysfunctions in combination, symptoms, and risk factors including diet, sleep and early life factors, on longitudinal SVD lesion change in patients presenting with stroke-related SVD.

Methods

Study design

The Mild Stroke Study 3 (MSS-3: ISRCTN 12113543) is a detailed prospective observational cohort study with clinical and imaging follow-up which aims to recruit up to 300 participants. In addition to a scan at initial stroke diagnosis, the patients undergo a minimum of three study scans over a year; in addition, those with lacunar stroke or moderate-to-severe WMH are invited for a further one or two visits between baseline and six months. The baseline assessment occurs within a maximum of three months of index stroke (Table 1). All participants return 6 and 12 months, respectively, after the baseline assessment. Annually, thereafter up to four years, we will invite participants to continue annual postal or phone follow-up with another MRI at three years. The follow-up questionnaire includes recurrent vascular events, cognition and functional status. MSS-3 benefits from systems established during the MSS-1 and MSS-2 studies^{27,28} and commenced in August 2018.

Study population

Adults >18 years old with mild ischaemic stroke with a modified Rankin scale (mRS) ≤ 2 at recruitment presenting to Edinburgh/Lothian stroke services.

Eligibility criteria

We define stroke as described previously:^{27,28} clinical lacunar stroke syndrome (50%) and control participants with non-lacunar ischaemic stroke syndromes (50%) i.e. partial anterior circulation syndrome or posterior circulation syndrome, with recent infarct visible on diagnostic MRI or CT scan compatible with the clinical syndrome, or if no visible infarct, no other lesion explaining the stroke symptoms. Participants with non-lacunar stroke form controls, since they have similar vascular risk factors and follow similar secondary prevention, accounting for medication effects on vessel function.

Table 1. Study assessments.

Activity/assessment	Pre-visit	V1 ^a : baseline	V2 ^b : 2–3 months	V3 ^c : 6 months	V4 ^d : 12 months
Eligibility	X				
Diagnostic MRI/CT	X				
Consent		X			
Routine blood tests	X				
Electrocardiogram	X				
Carotid Doppler ultrasound	X				
Symptom assessment		X	X	X	X
Cognitive tests		X	X	X	X
MRI		X	X	X	X
Retinal imaging		X	X	X	X
Blood pressure		X	X	X	X
Recurrent vascular events		X	X	X	X
NIHSS		X	X	X	X
mRS		X		X	X
Medical history/vascular risk factors		X		X	X
Demographic/socioeconomic factors		X			
Diet questionnaire		X			
Sleep questionnaire		X			
Mood/fatigue questionnaires		X			
Blood/urine collection		X			
24-h blood pressure monitoring		X			
Premorbid IQ (education, National Adult Reading Test)		X			
Informant questionnaire: IQCODE, NPI-Q, AES-I		X			X
Pulse wave measures		X			
Nine-hole peg test		X			X
Timed up + go		X			X
Stroke Impact Scale				X	X

^aVisit 1: baseline visit within three months of index stroke \pm 1 week.

^bVisit 2: \pm 1 week.

^cVisit 3: \pm 2 weeks.

^dVisit 4: \pm 3 weeks.

MRI: magnetic resonance imaging; NIHSS: National Institute of Health Stroke Scale; IQCODE: Informant Questionnaire for Cognitive Decline in the Elderly; NPI-Q: Neuropsychiatric Inventory Questionnaire; AES-I: Apathy Evaluation Scale-Informant.

We exclude participants with MRI contraindications, major neurological conditions, severe cardiac and respiratory disease. All participants give written informed consent. The study was granted ethical approval by Southeast Scotland Regional Ethics Committee (reference 18/SS/0044).

Diagnosis

An expert panel of stroke physicians and neuroradiologists reach final stroke diagnosis by consensus following review of presenting symptoms and signs including motor or sensory deficit, hemianopia, visuospatial disorder, ataxia, dysphasia, dysarthria, cerebellar or brainstem symptoms, supplemented by diagnostic brain MRI or CT and other relevant investigations, as previously described.^{27,28} An experienced neuroradiologist (JMW) assesses all scans for acute ischaemic lesions including recent small subcortical infarcts,

prior infarcts or haemorrhages, WMH, lacunes, PVS, microbleeds, siderosis and atrophy, using standardised validated scales.^{2,28}

Vascular risk factors, past medical history, medications and incident vascular events

Each participant provides a medical history of diagnoses confirmed by a physician, supplemented by hospital medical records and general practitioner correspondence, following standard definitions, including diabetes mellitus, hypertension, hypercholesterolaemia, previous stroke or TIA, peripheral vascular disease, atrial fibrillation, ischaemic heart disease, valvular defects, heart failure and physician-diagnosed anxiety, depression or delirium. We record current medications, cross-checking with electronic medical records. At follow-up, we record recurrent stroke, TIA and cardiac events.

Subjective symptoms

We use a structured questionnaire to ask open-ended questions about subjective symptoms experienced prior to, at the time of, and since index stroke diagnosis (see online Supplementary Appendix 1). Participants also answer questions based on previous clinico-radiological studies regarding symptoms within the past month including subjective memory concerns, confusional episodes, unsteadiness, falls, dizziness and headaches.²⁹

Participants self-administer the Fatigue Severity Scale,³⁰ Generalized Anxiety Disorder-7,³¹ the Center for Epidemiologic Studies-Depression Scale³² and an adapted Pittsburgh Sleep Quality Index.³³

Informant-reported symptoms

A nominated close friend or relative completes the following prior to the baseline visit: Neuropsychiatric Inventory Questionnaire,³⁴ behavioural changes since stroke,³⁵ Apathy Evaluation Scale, Informant version³⁶ and the Informant Questionnaire for Cognitive Decline in the Elderly³⁷ repeated at 12 months.

Family, lifestyle, social and early life factors

We record stroke or dementia family history including age at diagnosis, alcohol consumption and smoking status including quantity and duration. Participants self-administer the EPIC-Norfolk Food Frequency Questionnaire, a comprehensive dietary overview including salt intake.^{38,39}

To assess early life socioeconomic status, we record childhood postal address, number of individuals, rooms and toilets in the property and parental occupations. We note ethnicity, educational duration and attainment,⁴⁰ occupation, current postcode and retirement age.

Physical examination

We record presenting and current neurological deficits and stroke severity (NIHSS), blood pressure three times, gait (Timed Up and Go), manual dexterity (9-Hole Peg Test), height and weight.

Cognitive assessment

Participants complete the comprehensive 30-min neuropsychological test protocol based on the National Institute of Neurological Disorders and Stroke-Canadian Stroke Network (NINDS-CSN) Vascular Cognitive Impairment Harmonization Standards. This battery spans multiple cognitive domains and includes the Montreal Cognitive Assessment (MoCA), Hopkins Verbal Learning Test-revised, Controlled

Oral Word Association Test, Animal Naming, Letter Digit Coding and Trailmaking Tests A + B.

We estimate peak adult intelligence using the National Adult Reading Test.⁴⁰

Participants repeat MoCA and Trailmaking Tests A + B at each visit. We use three different MoCA versions, randomly assigning a test sequence to each participant to minimise learning effects on serial test performance.

Functional recovery

We administer the mRS⁴¹ at baseline, 6 and 12 months and the Stroke Impact Scale⁴² at 6 and 12 months.

Magnetic resonance imaging

We scan all participants at diagnosis at 1.5 T (General Electric Signa HDxt) or 3 T (Siemens Prisma) MRI or CT with core structural brain MRI sequences at each visit: 3D T1w, T2w, fluid attenuated inversion recovery, susceptibility-weighted (SWI/SWAN/GRE) and single- or multi-shell diffusion imaging (dMRI). Subsequent full cerebrovascular assessment and all follow-up imaging are at 3 T.

At one to three months post-stroke, participants undergo 3 T MRI to measure BBB integrity, CVR, cerebral blood flow (CBF) and intracranial vascular and CSF pulsatility (protocol in online Supplementary Appendix 2). We assess BBB integrity using dynamic contrast-enhanced (DCE)-MRI and gadolinium-based contrast agent (gadobutrol) injection,^{11,43} unless eGFR <30 ml/min. We assess CVR using a blood oxygenation level dependent (BOLD) MRI sequence, during which participants inhale air with intermittent-added CO₂ (12-min paradigm alternating 2 min air and 3 min 6% CO₂) through a tight-fitting facemask, described previously.^{13,44} Arterial, venous and CSF pulsatility are measured using phase contrast MRI sequences.^{14,44} We measure CBF using major arterial phase contrast flow measures obtained during pulsatility measurements (and arterial spin labelling where feasible).

We process MRI computationally using well-validated methods to assess intracranial volume, CSF, normal-appearing white and grey matter, WMH volumes, index and prior stroke lesion volumes, lacunes, microbleeds and perivascular space metrics.^{45,46} We visually quantify index and prior stroke lesions (location, type), WMH (baseline, change), lacunes (number, location), perivascular spaces, microbleeds, siderosis, superficial and deep brain volume loss, according to STRIVE criteria using validated scales.^{2,47-51} See online Supplementary Appendix 2 for image processing

methods description including advanced neuroimaging data.

Retinal imaging

We assess vision (Logmar cabinet, Sussex Vision) and use Spectralis OCT2[®] with Optical Coherence Tomography Angiography (OCTA) (Heidelberg Engineering) at each visit, imaging retinal vessels, retinopathy, nerve fibre layer thickness, choroid OCTA, intra-retinal and sub-retinal fluid. We computationally process retinal and arteriolar widths, branching patterns, complexity,⁵² nerve fibre layer thickness and microvessels on OCTA using well-validated tools.⁵³ See online Supplementary Appendix 2 for processing details.

Systemic vascular measures

We record BP at three standard points during the baseline visit (Omron) and measure arterial velocities through pulse wave velocity and pulse wave analysis using a tonometric device (*SphygmoCor*[®], AtCor Medical/Vicorder, Skidmore Medical) held over the carotid and radial pulses while supine. We provide a 24-h ambulatory BP monitoring device (SpaceLabs Medical) to most and encourage all to submit self-monitored blood pressure recordings. We repeat BP measurements at all study visits.

Biochemical, haematological, cardiovascular and imaging investigations

We document routinely collected index stroke investigation results including serum haematology and biochemistry, electrocardiogram, echocardiography and carotid Doppler ultrasound. We collect 18 ml venous blood at baseline for inflammatory and endothelial function markers. We store 20 ml urine for inflammatory marker analysis and 5 ml for albumin-to-creatinine ratio.

Endpoints

The primary endpoint is the proportion of SVD lesions that regress, progress or appear de novo in the year after stroke. The secondary endpoints are: (a) BBB integrity; (b) CVR; (c) intracranial vascular/CSF pulsatility; (d) WMH, PVS, lacunes and microbleeds; (e) white matter structural integrity measured with diffusion tensor and T_1 parameters and (f) incidence of reported symptoms including neuropsychiatric and cognitive symptoms, recurrent stroke, transient ischaemic attacks and cardiac events.

Statistical analysis

Sample size calculation. In a previous study with one year of longitudinal imaging follow-up at this centre, 10.6% of participants had a de novo lesion on MRI at follow-up.²⁸ In the same study, 65% had a mean 5.5 ml WMH volume increase and 35% had a mean 6.6 ml decrease.⁵⁴ A sample of 250 participants would be required to detect WMH change in the year after stroke, with significance 0.05 and power 0.90 in univariate analysis. We aim to recruit up to total 300 participants which allows for loss to follow-up.⁵⁴

Proposed analyses. In our primary analyses, we will use linear mixed effects models adjusted for age, vascular risk factors and baseline SVD burden to estimate the effect of cerebrovascular structure/function on SVD lesion progression and regression.

Secondarily, we will use similar models and other approaches (e.g. stratifying by low vs. high SVD burden, stroke subtype) to quantify associations of the following factors with lesion change: cognitive test scores and incident cognitive impairment; symptom test scores and incident symptoms; functional status; life course factors; lifestyle factors; blood pressure; systemically measured vascular stiffness; retinal measures; and inflammatory and endothelial function markers.

Discussion

The MSS-3 is a detailed prospective observational study which will advance our knowledge of how detailed measures of small vessel dysfunction and changing lesions relate to comprehensive symptom, cognitive, retinal, early life and lifestyle factors, whether some individuals are more vulnerable than others to the effects of small vessel dysfunction and whether a single candidate measure could best differentiate abnormal from normal-appearing brain tissue by stage and severity of SVD.

This study is novel in its concurrent use of advanced neuroimaging techniques to measure CVR, BBB leakage, CBF and vascular/CSF pulsatility: the first time these measures have been performed contemporaneously, alongside an unprecedented comprehensive assessment of symptoms and signs as they relate to these measures and lesion changes across multiple time-points. We build on previous studies,^{8,55} establishing a well-phenotyped profile of the dynamic natural history of SVD, capturing this rich dataset in the sub-acute post-stroke phase, monitoring the vulnerable brain at risk for early disease accumulation.^{16,56} Our systematic approach is a template for application to future research studies, designed to optimally assess

rates of disease progression and regression, translatable to other SVD presentations including mild cognitive impairment.

This study will fill an existing gap of longitudinal imaging studies evaluating symptoms such as apathy, fatigue, anxiety, delirium, sleep disturbance and emotional lability, contributing to the detection of preclinical SVD states. We will identify ‘red flags’ to the presence and progression of SVD, so that we may intercept disease earlier, even before it develops, rather than in patients presenting with overt brain dysfunction, e.g. stroke, dementia. We will gain insight into novel preventative and therapeutic targets by uncovering the nature of and factors associated with lesion regression. This study will deepen our understanding of SVD, essential to future prevention and treatment of stroke and vascular dementia.

To date, we have recruited 116 participants. The baseline visit lasts 6.5h, and most are willing to attend three further visits, each lasting two hours, with positive participant feedback, demonstrating the feasibility of applying this design at other centres.

Trial registration

ISRCTN 12113543

Declaration of conflicting interests

The author(s) declared the following potential conflicts of interest with respect to the research, authorship, and/or publication of this article: UC, MS, MJT, GB, AM, OH, CM, FND and JMW hold academic grants from government and charitable funding agencies, outlined below.

Funding

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work is supported by: the UK Dementia Research Institute which receives its funding from DRI Ltd, funded by the UK MRC, Alzheimer’s Society and Alzheimer’s Research UK; the Fondation Leducq Network for the Study of Perivascular Spaces in Small Vessel Disease (16 CVD 05); Stroke Association ‘Small Vessel Disease-Spotlight on Symptoms (SVD-SOS)’(SAPG 19\100068; The Row Fogo Charitable Trust Centre for Research into Aging and the Brain; Stroke Association Garfield Weston Foundation Senior Clinical Lectureship (FND) (TSALECT 2015/04); NHS Research Scotland (FND); Stroke Association Post-Doctoral Fellowship (SW) (SAPDF 18/100026); British Heart Foundation Edinburgh Centre for Research Excellence (RE/18/5/34216); NHS Lothian Research and Development Office (MJT); European Union Horizon 2020, PHC-03-15, project No666881, ‘SVDs@Target’ (MS,GB); Chief Scientist Office of Scotland

Clinical Academic Fellowship (UC) (CAF/18/08); Stroke Association Princess Margaret Research Development Fellowship (UC) (2018); Medical Research Scotland studentship (AM) (PhD-1165-2017); College of Medicine and Veterinary Medicine, University of Edinburgh scholarship, as part of the Wellcome-funded Translational Neuroscience PhD programme (OH); MRC Doctoral Training Programme in Precision Medicine (CM) (MR/R01566X/1). The Research MR scanners are supported by the Scottish Funding Council through the Scottish Imaging Network, A Platform for Scientific Excellence (SINAPSE) Collaboration; the 3T scanner is funded by the Wellcome Trust (104916/Z/14/Z), Dunhill Trust (R380R/1114), Edinburgh and Lothians Health Foundation (2012/17), Muir Maxwell Research Fund and the University of Edinburgh.

Informed consent

Written informed consent is obtained from all subjects before the study.

Ethical approval

Ethical approval for this study was obtained from South East Scotland Research Ethics Committee (Ref 18/SS/0044) on 31 May 2018. NHS Lothian Research & Development approved this study on 31 May 2018 (Ref 2018/0084).

Contributorship

UC: recruitment, data collection/management, study design, study coordination; DJG, WH: data collection/management, study coordination; IM, MB, MJT, MS, GB, SMM, ES, CM, AGM, IH: advanced neuroimaging techniques advice/design; TM, SW, KH, CH: retinal imaging techniques advice/design, data collection; MCVH, LB, MS, MJT: image analysis techniques advice/design; OH: cognitive test protocol advice, data collection; RB, EB: laboratory processing advice; DJ: data management; CA: advice regarding study design; FC: advice/study design, data management, statistical analysis plan; FD: recruitment, funding, study design, supervision, clinical oversight; JMW: conception, funding, ethics and regulatory approvals, study design, data collection, all supervision and governance, drafting and final editing of text. All authors also prepared, revised and approved the final manuscript.

Guarantor


JMW.

Acknowledgements


We thank the participants, their families, radiographers at Edinburgh Imaging Facility Royal Infirmary of Edinburgh and the Stroke Research Network at the University of Edinburgh.

ORCID iDs

Una Clancy  <https://orcid.org/0000-0002-8062-7537>

Olivia KL Hamilton  <https://orcid.org/0000-0002-5874-0058>

Lucia Ballerini  <https://orcid.org/0000-0002-1956-4012>

Joanna M Wardlaw  <https://orcid.org/0000-0002-9812-6642>

Supplemental material

Supplemental material for this article is available online.

References

1. Wardlaw JM, Smith C and Dichgans M. Small vessel disease: mechanisms and clinical implications. *Lancet Neurol* 2019; 18: 684–696.
2. Wardlaw JM, Smith EE, Biessels GJ, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. *Lancet Neurol* 2013; 12: 822–838.
3. Sudlow CL and Warlow CP. Comparable studies of the incidence of stroke and its pathological types: results from an international collaboration. International Stroke Incidence Collaboration. *Stroke* 1997; 28: 491–499.
4. Gorelick PB, Scuteri A, Black SE, et al. Vascular contributions to cognitive impairment and dementia: a statement for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke* 2011; 42: 2672–2713.
5. Sweeney MD, Montagne A, Sagare AP, et al. Vascular dysfunction – the disregarded partner of Alzheimer’s disease. *Alzheimer’s Dementia* 2019; 15: 158–167.
6. Georgakis MK, Dering M, Wardlaw JM, et al. WMH and long-term outcomes in ischemic stroke: a systematic review and meta-analysis. *Neurology* 2019; 92: e1298–e1308.
7. Wardlaw JM, Chappell FM, Hernandez MDCV, et al. White matter hyperintensity reduction and outcomes after minor stroke. *Neurology* 2017; 89: 1003–1010.
8. van Leijssen EMC, van Uden IWM, Ghafoorian M, et al. Nonlinear temporal dynamics of cerebral small vessel disease: the RUN DMC study. *Neurology* 2017; 89: 1569–1577.
9. Ramirez J, McNeely AA, Berezuk C, et al. Dynamic progression of white matter hyperintensities in Alzheimer’s disease and normal aging: results from the Sunnybrook Dementia Study. *Front Aging Neurosci* 2016; 8: 62.
10. Cho AH, Kim HR, Kim W, et al. White matter hyperintensity in ischemic stroke patients: it may regress over time. *J Stroke* 2015; 17: 60–66.
11. Wardlaw JM, Makin SJ, Hernández MDCV, et al. Blood-brain barrier failure as a core mechanism in cerebral small vessel disease and dementia: evidence from a cohort study. *Alzheimer’s Dementia* 2017; 13: 634–643.
12. Blair GW, Doubal FN, Thrippleton MJ, et al. Magnetic resonance imaging for assessment of cerebrovascular reactivity in cerebral small vessel disease: a systematic review. *J Cereb Blood Flow Metabol* 2016; 36: 833–841.
13. Blair GW, Thrippleton MJ, Shi Y, et al. Intracranial hemodynamic relationships in patients with cerebral small vessel disease. *Neurology* 2020; 94(21): e2258–e2269.
14. Shi Y, Thrippleton MJ, Blair GW, et al. Small vessel disease is associated with altered cerebrovascular pulsatility but not resting cerebral blood flow. *J Cereb Blood Flow Metab* 2020; 40: 85–99.
15. Conklin J, Silver FL, Mikulis DJ, et al. Are acute infarcts the cause of leukoaraiosis? Brain mapping for 16 consecutive weeks. *Ann Neurol* 2014; 76: 899–904.
16. Lee EJ, Kang DW and Warach S. Silent new brain lesions: innocent bystander or guilty party? *J Stroke* 2016; 18: 38–49.
17. Ter Telgte A, Wiegertjes K, Gesierich B, et al. Contribution of acute infarcts to cerebral small vessel disease progression. *Ann Neurol* 2019; 86: 582–592.
18. Stewart R, Godin O, Crivello F, et al. Longitudinal neuroimaging correlates of subjective memory impairment: 4-year prospective community study. *Br J psychiatry* 2011; 198: 199–205.
19. Pinter D, Ritchie SJ, Doubal F, et al. Impact of small vessel disease in the brain on gait and balance. *Sci Rep* 2017; 7: 41637.
20. Hollocks MJ, Lawrence AJ, Brookes RL, et al. Differential relationships between apathy and depression with white matter microstructural changes and functional outcomes. *Brain* 2015; 138: 3803–3815.
21. Saini M, Ikram K, Hilal S, et al. Silent stroke: not listened to rather than silent. *Stroke* 2012; 43: 3102–3104.
22. Choi SH, Na DL, Chung CS, et al. Diffusion-weighted MRI in vascular dementia. *Neurology* 2000; 54: 83–89.
23. Chowdhury D, Wardlaw JM and Dennis MS. Are multiple acute small subcortical infarctions caused by embolic mechanisms? *J Neurol Neurosurg Psychiatry* 2004; 75: 1416–1420.
24. Wardlaw JM, Allerhand M, Doubal FN, et al. Vascular risk factors, large-artery atheroma, and brain white matter hyperintensities. *Neurology* 2014; 82: 1331–1338.
25. Gardener H, Scarmeas N, Gu Y, et al. Mediterranean diet and white matter hyperintensity volume in the Northern Manhattan Study. *Arch Neurol* 2012; 69: 251–256.
26. Backhouse EV, McHutchison CA, Cvorov V, et al. Early life risk factors for cerebrovascular disease: a systematic review and meta-analysis. *Neurology* 2017; 88: 976–984.
27. Wardlaw JM, Doubal F, Armitage P, et al. Lacunar stroke is associated with diffuse blood-brain barrier dysfunction. *Ann Neurol* 2009; 65: 194–202.
28. Makin SD, Doubal FN, Dennis MS, et al. Clinically confirmed stroke with negative diffusion-weighted imaging magnetic resonance imaging: longitudinal study of clinical outcomes, stroke recurrence, and systematic review. *Stroke* 2015; 46: 3142–3148.
29. van Rooij FG, Vermeer SE, Goraj BM, et al. Diffusion-weighted imaging in transient neurological attacks. *Ann Neurol* 2015; 78: 1005–1010.
30. Krupp LB, LaRocca NG, Muir-Nash J, et al. The fatigue severity scale: application to patients with multiple sclerosis and systemic lupus erythematosus. *Arch Neurol* 1989; 46: 1121–1123.

31. Spitzer RL, Kroenke K, Williams JB, et al. A brief measure for assessing generalized anxiety disorder: the GAD-7. *Arch Intern Med* 2006; 166: 1092–1097.
32. Radloff LS. The CES-D scale: a self-report depression scale for research in the general population. *Appl Psychol Meas* 1977; 1: 385–401.
33. Buysse DJ, Reynolds CF 3rd, Monk TH, et al. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. *Psychiatry Res* 1989; 28: 193–213.
34. Kaufer DI, Cummings JL, Ketchel P, et al. Validation of the NPI-Q, a brief clinical form of the Neuropsychiatric Inventory. *J Neuropsychiatr Clin Neurosci* 2000; 12: 233–239.
35. American Psychiatric Association. *Diagnostic and statistical manual of mental disorders (DSM-5)*. 5th ed. Washington, DC: American Psychiatric Publishing, 2013.
36. Marin RS, Biedrzycki RC and Firinciogullari S. Reliability and validity of the Apathy Evaluation Scale. *Psychiatr Res* 1991; 38: 143–162.
37. Jorm AF. A short form of the Informant Questionnaire on Cognitive Decline in the Elderly (IQCODE): development and cross-validation. *Psychol Med* 1994; 24: 145–153.
38. Mulligan AA, Luben RN, Bhaniani A, et al. A new tool for converting food frequency questionnaire data into nutrient and food group values: FETA research methods and availability. *BMJ Open* 2014; 4: e004503.
39. Makin SDJ, Mubki GF, Doubal FN, et al. Small vessel disease and dietary salt intake: cross-sectional study and systematic review. *J Stroke Cerebrovasc Disease* 2017; 26: 3020–3028.
40. Makin SD, Doubal FN, Shuler K, et al. The impact of early-life intelligence quotient on post stroke cognitive impairment. *Eur Stroke J* 2018; 3: 145–156.
41. van Swieten JC, Koudstaal PJ, Visser MC, et al. Interobserver agreement for the assessment of handicap in stroke patients. *Stroke* 1988; 19: 604–607.
42. Duncan PW, Bode RK, Lai SM, et al. Rasch analysis of a new stroke-specific outcome scale: the Stroke Impact Scale. *Arch Phys Med Rehabil* 2003; 84: 950–963.
43. Heye AK, Thrippleton MJ, Armitage PA, et al. Tracer kinetic modelling for DCE-MRI quantification of subtle blood-brain barrier permeability. *NeuroImage* 2016; 125: 446–455.
44. Thrippleton MJ, Shi Y, Blair G, et al. Cerebrovascular reactivity measurement in cerebral small vessel disease: rationale and reproducibility of a protocol for MRI acquisition and image processing. *Int J Stroke* 2018; 13: 195–206.
45. Hernandez MDCV, Armitage PA, Thrippleton MJ, et al. Rationale, design and methodology of the image analysis protocol for studies of patients with cerebral small vessel disease and mild stroke. *Brain Beh* 2015; 5: e00415.
46. Ballerini L, Lovreglio R, Hernandez MDCV, et al. Perivascular spaces segmentation in Brain MRI using optimal 3D filtering. *Sci Rep* 2018; 8: 2132.
47. Association between brain imaging signs, early and late outcomes, and response to intravenous alteplase after acute ischaemic stroke in the third International Stroke Trial (IST-3): secondary analysis of a randomised controlled trial. *Lancet Neurol* 2015; 14: 485–496.
48. Cordonnier C, Potter GM, Jackson CA, et al. Improving interrater agreement about brain microbleeds: development of the Brain Observer MicroBleed Scale (BOMBS). *Stroke* 2009; 40: 94–99.
49. Fazekas F, Niederkorn K, Schmidt R, et al. White matter signal abnormalities in normal individuals: correlation with carotid ultrasonography, cerebral blood flow measurements, and cerebrovascular risk factors. *Stroke* 1988; 19: 1285–1288.
50. Potter GM, Chappell FM, Morris Z, et al. Cerebral perivascular spaces visible on magnetic resonance imaging: development of a qualitative rating scale and its observer reliability. *Cerebrovasc Dis (Basel, Switzerland)* 2015; 39: 224–231.
51. Wardlaw JM and Sellar R. A simple practical classification of cerebral infarcts on CT and its interobserver reliability. *Am J Neuroradiol* 1994; 15: 1933–1939.
52. Doubal FN, MacGillivray TJ, Patton N, et al. Fractal analysis of retinal vessels suggests that a distinct vasculopathy causes lacunar stroke. *Neurology* 2010; 74: 1102–1107.
53. MacGillivray TJ, Trucco E, Cameron JR, et al. Retinal imaging as a source of biomarkers for diagnosis, characterization and prognosis of chronic illness or long-term conditions. *Br J Radiol* 2014; 87: 20130832.
54. Chappell FM, Hernandez MDCV, Makin SD, et al. Sample size considerations for trials using cerebral white matter hyperintensity progression as an intermediate outcome at 1 year after mild stroke: results of a prospective cohort study. *Trials* 2017; 18: 78.
55. Raz N, Yang YQ, Rodrigue KM, et al. White matter deterioration in 15 months: latent growth curve models in healthy adults. *Neurobiol Aging* 2012; 33: 429.e1–429.e5.
56. Kang DW, Latour LL, Chalela JA, et al. Early and late recurrence of ischemic lesion on MRI: evidence for a prolonged stroke-prone state? *Neurology* 2004; 63: 2261–2265.