

Supplementary File 1 - METHODS

Peripheral vascular assessment

VASCULAR MEASURE	METHODS
Pulse wave velocity (PWV), m/s	Distance between carotid and femoral arteries measured and divided by time delay between carotid and femoral pulses, calculated by ECG-gated applanation tonometry (SphygmoCor; AtCor Medical, Sydney, Australia).
Flow mediated dilatation (FMD) of the brachial artery, %	Ultrasound imaging of brachial artery at rest, and following 5 min of forearm occlusion at 200mmHg. The maximum peak in diameter post cuff deflation compared to baseline was calculated (flow mediated dilatation, FMD, %) (Brachial Analyzer, MIA LLC, IA, USA).
Carotid intima media thickness (CIMT), mm	B-mode ultrasound and a 7MHz probe (Acuson, Sequoia, Siemens Medical Solutions) used to obtain images of the posterior intima media layer in two planes. Automatic border detection software (Siemens Syngo® Arterial Health Package) used to measure the carotid intima media thickness (CIMT) and the average of all readings used.

Methods - Composite scores in neuropsychological testing

The processing speed composite score was the mean of z-scores on the Symbol-Digit Modalities Test (SDMT, written version(1)) and Trailmaking Test Part A (2). The attention/working memory composite score was the mean of scale scores on the forward and backward Digit Span sub-tests from the Wechsler Adult Intelligence Scale (WAIS-IV)(3) and the Symbol Span sub-test from the Wechsler Memory Scale (WMS-IV)(4). The executive function composite score was the mean of z-scores on the Similarities sub-test from the WAIS-IV (3), FAS letter fluency test (5), Trailmaking Test Part B(2), and Stroop Neuropsychological Screening Test (6).

1. Smith A. Symbol Digit Modalities Test (SDMT). Torrance, CA:: WPS; 1982.
2. Tombaugh TN. Trail Making Test A and B: Normative data stratified by age and education. Archives of Clinical Neuropsychology. 2004;19(2):203-14.
3. Wechsler D. Wechsler Adult Intelligence Scale - Fourth UK Edition (WAIS-IV UK). London: Pearson Assessment; 2011.
4. Wechsler D. Wechsler Memory Scale - Fourth UK edition (WMS-IV UK). London: Pearson Assessment; 2011.
5. Tombaugh TN, Kozak J, Rees L. Normative Data Stratified by Age and Education for Two Measures of Verbal Fluency: FAS and Animal Naming. Archives of Clinical Neuropsychology. 1999;14(2):167-77.
6. Trenerry MC, B.; DeBoe J.; et al. Stroop Neuropsychological Screening Test (SNST). Lutz, FL: PAR, Inc; 1989.

Methods - MRI acquisition

SEQUENCE	ORIENTATION	DETAILS
T2-weighted FLAIR	Axial	repetition time/inversion time/echo time = 10000/2250/140ms, slice thickness = 5mm, interslice gap = 1.5mm, matrix 384 x 256, acquisition time = 3:20min
T1-weighted FSPGR BRAVO	Axial	repetition time/inversion time/echo time = 9.0/450/3.6 ms, slice thickness = 1mm, no interslice gap, flip angle= 12°, matrix 320 x 320, acquisition time = 4:28min
T2*-weighted susceptibility angiography (SWAN)	Axial	repetition time/echo time = 40/25 ms, slice thickness 3.6mm, slice gap = 1.8mm, flip angle 15°, matrix 320 x 224, acquisition time = 2:16min
Inhance 3D velocity MR angiography (MRA)	Sagittal	repetition time/echo time = 8.6/3.54 ms, slice thickness = 1.2mm, flip angle = 8°, matrix 320 x 224, acquisition time = 3:11min
3D Pseudocontinuous arterial spin labelling (ASL)*	Axial	repetition time = 4864ms, echo time = 10.1ms, labelling duration = 1500ms, post-labelling delay time = 2025ms, slice thickness 3.5mm, matrix 128 x 128, flip angle 155°, NEX 3.0, time = 4.42min.

*In the first three patients, a neurovascular head coil (8 channel) was used when obtaining the ASL scans at baseline.

Methods - Transcranial Doppler ultrasound

This was performed during this study but is not discussed in the main paper.

Details of how this was performed can be found in Moreton et al, Journal of Cerebral Blood Flow & Metabolism; 2018, Vol. 38(6) 1085-1095.

SUPPLEMENTARY FILE 2 - RESULTS

Table 1: Change over study period in neuropsychological measures with repeated measures comparison

	n ^a	Baseline	Year 1	Year 2	p value ^c
Neuropsychological measures					
Attention/Working memory, median [IQR], composite scale score ^b	20	9.2 [6.8 - 10.7]	9.2 [7.8 - 10.3]	9.6 [7.1 - 11.3]	0.196
Executive function, median [IQR], composite z-score	17	-0.3 [-0.9 - 0.1]	-0.2 [-1.2 to 0.1]	-0.3 [-1.8 - 0.5]	0.943
Trailmaking B minus A, median [IQR], seconds	19	35 [26 - 82]	47 [30 - 69]	46 [25 - 103]	0.841
Transcranial Doppler measures					
Mean flow velocity, cm/s, mean (SD)	13	42 (9)	41 (8)	44 (10)	0.354
Cerebrovascular reactivity, %, mean (SD)	13	19 (9)	19 (12)	15 (9)	0.288

^anumber where data available at all three time points. ^bStandard scale with mean = 10 and SD = 3. ^cNormally distributed data expressed as mean (SD) and comparisons made with repeated measures ANOVA. Non-parametric data expressed as median [IQR] and comparisons made with Friedman test.

Table 2: Baseline vascular and radiological markers in those obtaining the clinical composite outcome

Baseline value	Did not obtain composite outcome ^a	Obtained composite outcome	p value ^b
Number	17	5	-
Age, years	48 (12)	54 (6)	0.328
Grey matter CBF, ml/100g/min, mean (SD)	51 (10)	43 (9)	0.207
White matter CBF, ml/100g/min, mean (SD)	43 (7)	40 (10)	0.551
Subcortical hyperintensity CBF, ml/100g/min, mean (SD)	23 (5)	21 (2)	0.437
Brain CVR, %/CO ₂ , mean (SD)	8 (9)	3 (5)	0.474
CIMT, mm, mean (SD)	0.64 (0.1)	0.64 (0.1)	0.925
FMD, %, mean (SD)	4.1 (1.9)	3.3 (1.9)	0.552
PWV, m/s, mean (SD)	7.5 (1.0)	8.0 (1.4)	0.523
Systolic blood pressure, mmHg, mean (SD)	122 (11)	114 (10)	0.192
Lacunae, n, median [IQR]	4 [11]	14 [25]	0.109
Microbleeds, n, median [IQR]	0 [5]	3 [8]	0.249
Normalised subcortical hyperintensity volume, %, mean (SD)	6.7 (2.4)	11.3 (2.9)	0.002
Brain volume, L, mean (SD)	1.55 (0.08)	1.53 (0.13)	0.615

^aComposite outcome = incident stroke or incident new dementia or increase of modified Rankin Scale of one or more than one in patients with pre-existing

dementia. ^bIndependent t-test for normal data (mean, SD), Mann-Whitney U test for non-parametric data (median [IQR]).

Table 3: Baseline vascular and radiological markers in subjects in those with decline in attention/working memory

	No decline in attention/working memory	Decline in attention/working memory	p value ^a
Number	15	5	-
Age, years, mean (SD)	49 (11)	54 (10)	0.366
Grey matter CBF, ml/100g/min, mean (SD)	52 (6)	40 (8)	0.009
White matter CBF, ml/100g/min, mean (SD)	43 (4)	36 (8)	0.135^b
Subcortical hyperintensities ml/100g/min, mean (SD)	23 (4)	19 (3)	0.079
Brain CVR, %/CO ₂ , mean (SD)	8 (9)	7 (9)	0.967
CIMT, mm, mean (SD)	0.64 (0.1)	0.64 (0.1)	0.961
FMD, %, mean (SD)	3.4 (1.5)	4.9 (2.1)	0.153
PWV, m/s, mean (SD)	7.7 (1.0)	7.8 (1.3)	0.834
Systolic blood pressure, mmHg, mean (SD)	121 (9)	121 (15)	0.888
Lacunae, n, median (IQR)	5 [11]	17 [29]	0.257
Microbleeds, n, median (IQR)	1 [5]	3 [6]	0.553
Normalised subcortical hyperintensity volume, mean (SD)	6.8 (2.3)	11.1 (3.1)	0.004
Brain volume, mean (SD)	1.53 (0.1)	1.54 (0.1)	0.787

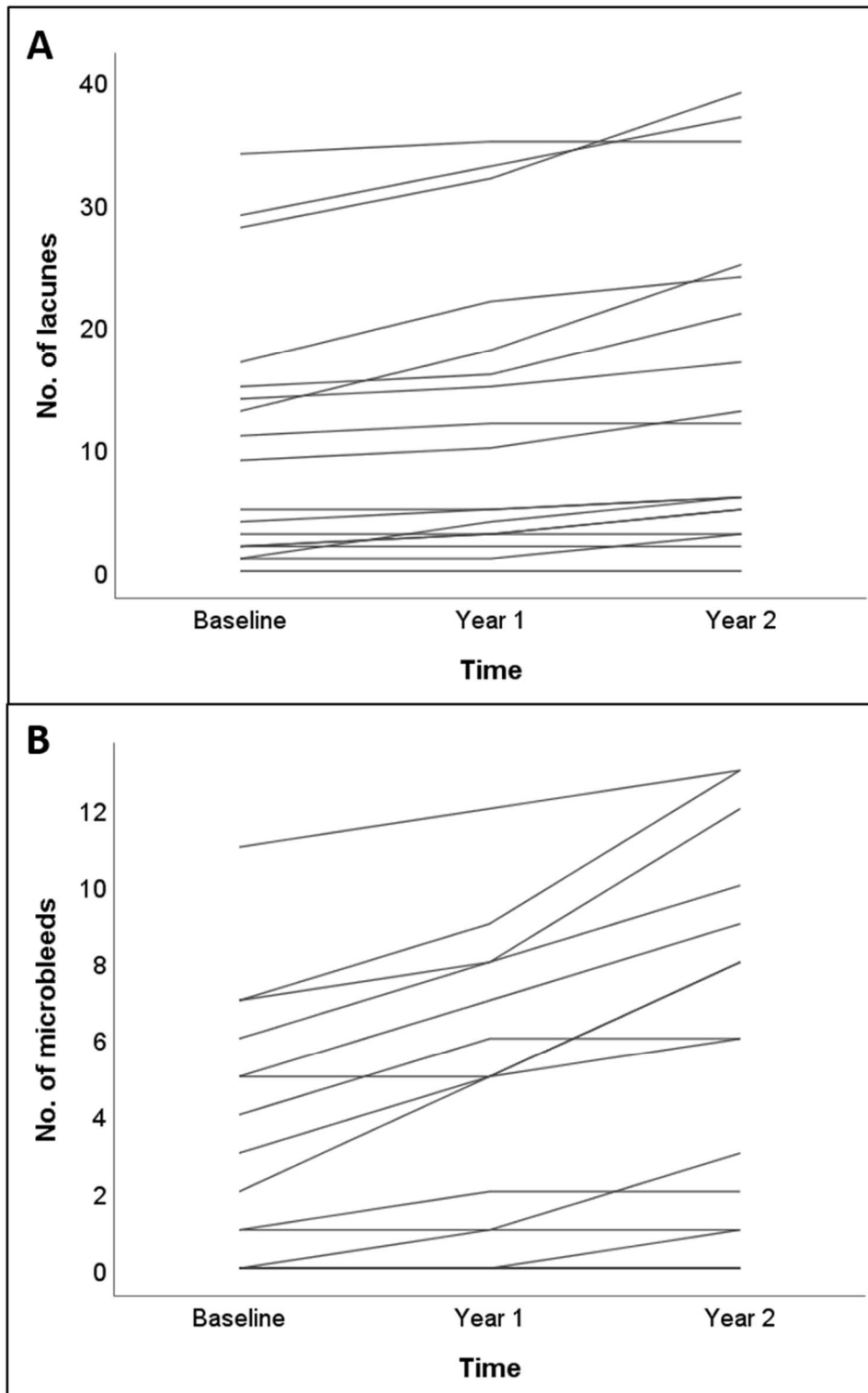
^aIndependent t-test for normal data (mean, SD), Mann-Whitney U test for non-parametric data (median [IQR]). ^bEqual variances not assumed

Table 4: Baseline vascular markers predicting change in structural MRI from baseline to year 2 - lacunes and microbleeds.

			Incident lacunes at 2 years	Incident microbleeds at 2 years
Spearman's rho	Age, years	Correlation Coefficient	.364	-.062
		Sig. (2-tailed)	.125	.795
		N	19	20
	Grey matter CBF, ml/100g/min	Correlation Coefficient	-.291	-.063
		Sig. (2-tailed)	.292	.824
		N	15	15
	Brain CVR, %/CO ₂	Correlation Coefficient	-.339	-.068
		Sig. (2-tailed)	.236	.818
		N	14	14
	CIMT, mm	Correlation Coefficient	.347	.127
		Sig. (2-tailed)	.146	.592
		N	19	20
	FMD, %	Correlation Coefficient	-.065	.144
		Sig. (2-tailed)	.818	.596
		N	15	16
	PWV, m/s	Correlation Coefficient	-.030	-.011
		Sig. (2-tailed)	.905	.966
		N	18	19

Figure 1: Spaghetti plots of change in MRI variables over two years.

(A) Number of lacunes; (B) number of microbleeds; (C) normalised subcortical hyperintensity volume; and (D) normalised brain volume. Each line shows an individual subject.



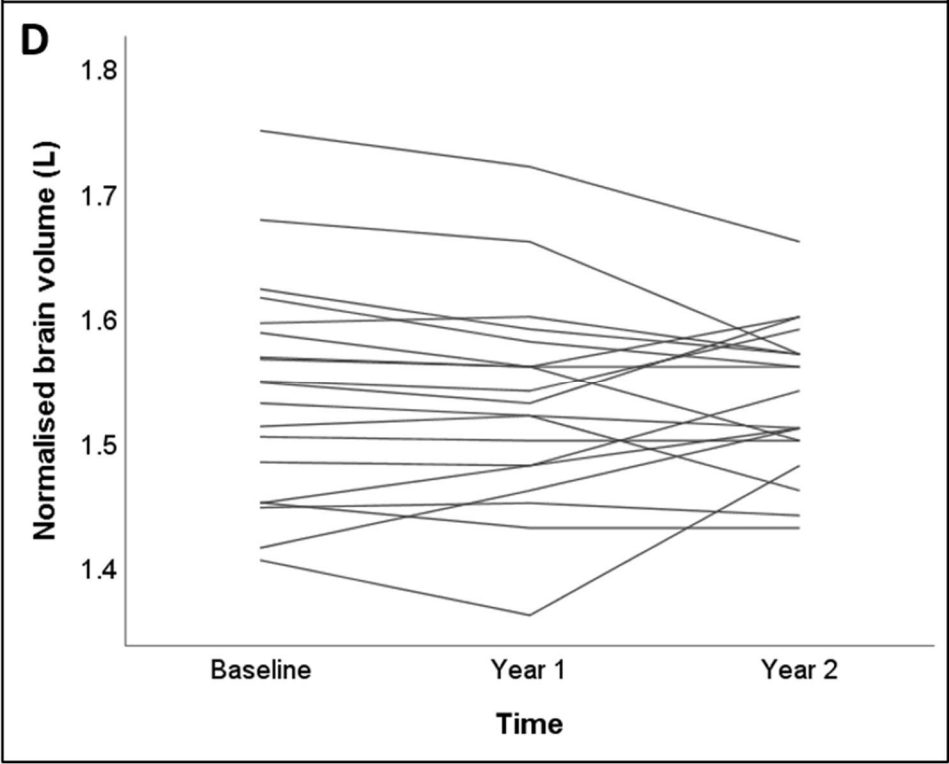
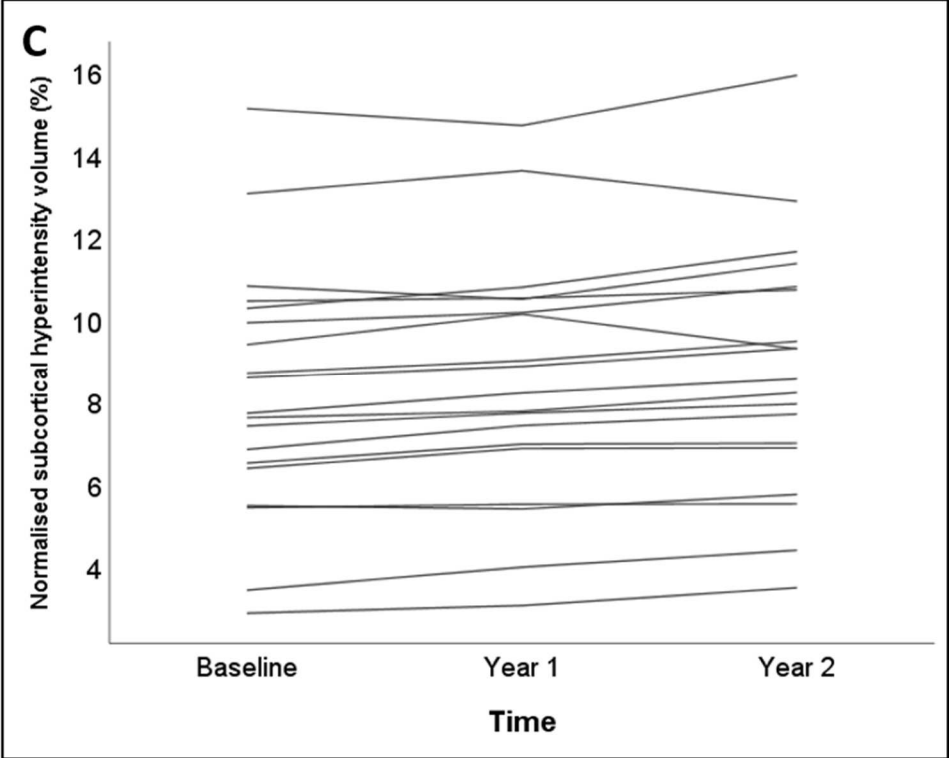


Figure 2: Spaghetti plots of change in key vascular and neuropsychological variables over two years

(A) Grey matter cerebral blood flow; (B) brain cerebrovascular reactivity measured by ASL; (C) carotid intima media thickness; and (D) processing speed. Each line represents an individual subject

