Supplementary material

Methods

Scanning protocol:

The MRI scanning protocol consisted of pre- and post-gadolinium 3DT1 scans (TR= 9.8 ms, TE= 4.6 ms, flip angle 8°, voxelsize 1.15 x 1.15 mm, 130 slices of 1.2 mm), a sagittal acquired 3D FLAIR (TR= 4800 ms, TE= 279 ms, flip angle 40°, voxelsize 1.11 x 1.11 mm, 310 slices of 1.12 mm), a DWI (TR= 2685 ms, TE= 72 ms, flip angle 90°, voxelsize 1.96 x 2.44 mm, 25 slices of 5 mm), a SWI (TR= 45 ms, TE= 31 ms, flip angle 13°, voxelsize 0.78 x 0.78 mm, 140 slices of 0.8 mm) and a T2 scan (TR= 4744 ms, TE= 80 ms, flip angle 90°, voxelsize 0.43 x 0.48 mm, 48 slices of 3 mm).

Measurements:

General features of small-vessel disease were evaluated on baseline scans. Brain atrophy was defined as central (enlargement of the ventricles) or peripheral (enlargement of the gyri) and rated on a subjective scale of 0 to 3 (none/moderate/severe).[1] The volume of WMLs was automatically assessed using a previously described method,[2] using tools of the FSL software package (Functional MRI of the Brain [FMRIB] Software Library v5, www.fmrib.ox.ac.uk/fsl).[3] In short, 3DT1 and FLAIR images were brain extracted using the brain extraction tool and co-registered using FMRIB's linear image registration tool to the MNI152 standard space. WML volumes in a conservative MNI white matter mask were automatically identified using a threshold of three standard deviations above the mean FLAIR signal intensity.[2] Presence of periventricular WMLs and deep WMLs were also separately evaluated using the Fazekas scale.[4] Enlarged perivascular spaces (EPVS) were assessed in the

basal ganglia (BG) and semiovale center (SC) on T2 and T1-weighted images, and rated according to a 4-point semi-quantitative score (none/mild/moderate/frequent).[5] Lacunar infarcts were defined as parenchymal defects <20 mm, hyperintense on T2-weighted images and hypointense with hyperintense rim on FLAIR images, located in the basal ganglia, thalamus, internal or external capsule, or brainstem.

References methods

1. Farrell C, Chappell F, Armitage PA, et al. Development and initial testing of normal reference MR images for the brain at ages 65-70 and 75-80 years. *Eur Radiol.* 2009;19(1):177-83. doi:https://doi.org/10.1007/s00330-008-1119-2.

2. Hulsey KM, Gupta M, King KS, et al. Automated quantification of white matter disease extent at 3 T: comparison with volumetric readings. *J Magn Reson Imaging.* 2012;36(2):305-11. doi:https://doi.org/10.1002/jmri.23659.

3. Jenkinson M, Beckmann CF, Behrens TE, et al. Fsl. *Neuroimage.* 2012;62(2):782-90. doi:https://doi.org/10.1016/j.neuroimage.2011.09.015.

4. Fazekas F. MR signal abnormalities at 1.5 T in Alzheimer's dementia and normal aging. *AJR Am J Roentgenol.* 1987:351-6. doi:https://doi.org/10.2214/ajr.149.2.351.

5. 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.* 2015;39(3-4):224-31. doi:https://doi.org/10.1159/000375153.

Supplementary Table 1 Clinical findings during Baseline and Follow-up

	Baseline (n=29)	**Follow-up (n=17)**
Retinopathy[1] (yes, n(%))	19/20 (95)	16/17 (94)
Features of focal or global brain dysfunction (yes, n(%))	11/29 (38)	11/17 (65)
Internal organ dysfunction[2] (yes, n(%))	6/29 (21)	8/17 (47)

[1] 20 mutation carriers were aware of their mutation status and were tested for vascular retinopathy.

[2] Internal organ dysfunction was defined as organ dysfunction (liver, kidney and thyroid disease and anemia) requiring treatment.

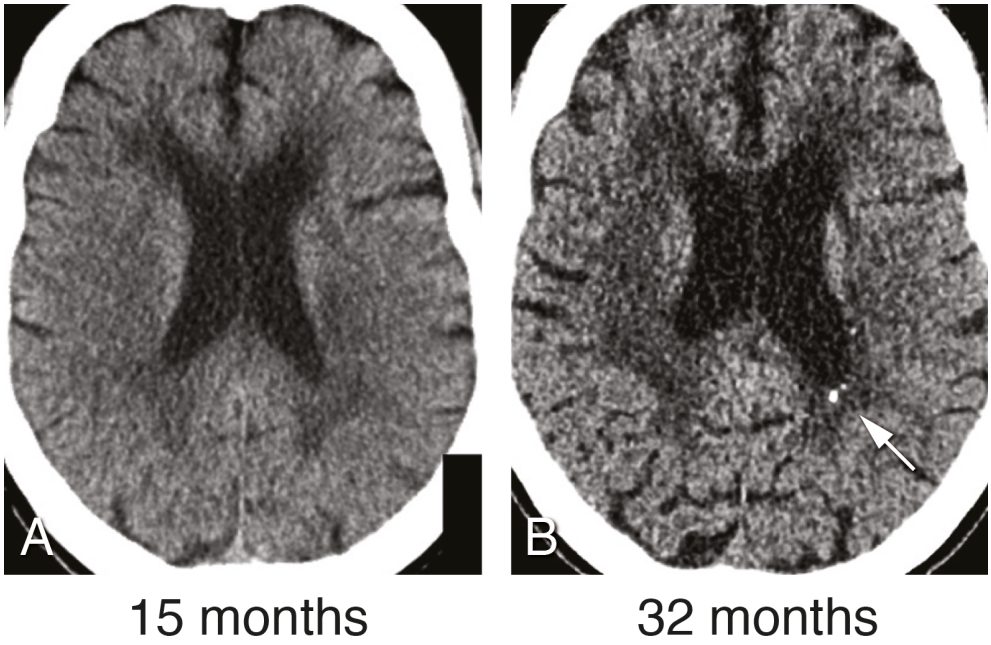
Supplementary Table 2 Imaging findings in mutation carriers with progression at follow-up

MC	Age at Baseline*	Time Follow up (Months)	Nr of Baseline Lesions	Nr of New Lesions	Pseudo Tumor	Maximum Contrast Enhancement (Months)	Maximum Diffusion Restriction (Months)	New Infratentorial Lesions
1	60	31	9	1	Yes	31	23	-
2	60	36	5	6	-	37	22	-
3	50	28	2	2	-	16	-	-
4	55	28	1	1	-	19	-	Yes
5	60	25	6	3	-	25	25	Yes
6	45	33	0	4	Yes	6	6	Yes
7	45	33	2	3	-	17	16	-
8	55	31	7	1	-	31	31	-
9	65	26	0	1	-	-	-	-
10	40	27	1	5	-	-	-	Yes
11	55	31	10	1	-	32	32	-
12	60	24	0	0	-	-	-	Yes

MC=mutations carrier

*Age was rounded to the nearest half-decade to protect anonymity

Supplementary Figure 1 Development of focal calcifications on CT images



CT images acquired in the mutation carrier from Figure 5 at 15 and 32 months. No calcifications are present at 15 months. At 32 months development of focal calcifications is seen, corresponding with the SWI artefacts seen at 31 months (Figure 5).