Supplementary Online Content

Gregg NM, Kim AE, Gurol ME, et al. Incidental cerebral microbleeds and cerebral blood flow in elderly individuals. *JAMA Neurol*. Published online July 13, 2015. doi:10.1001/jamaneurol.2015.1359.

eMethods.

eTable. CMB topography

eMethods

Inter-rater reliability assessment

For assessment of agreement at the level of individual CMBs, there was agreement for no microbleeds in 11 of the 20 subjects. The two readers identified a total of 23 distinct CMBs, with agreement on 17 of the 23 CMBs, for an overall agreement in 28 of 34 comparisons (17 individual CMBs plus 11 CMB negative subjects), and a kappa of 0.64.

Structural MRI

Structural T1 MPRAGE used parameters optimized for maximal contrast among grey matter, white matter, and cerebrospinal fluid. Data were acquired in the sagittal plane (echo time (TE) = 2.98, repetition time (TR) = 23, flip angle = 9 degrees, NEX = 1, slice thickness = 1.2 mm, no gap). This generated a 256 x 240 x 160 matrix with a 24 x 25.6 cm FOV. Data were first bias corrected using the unified segmentation approach in SPM8[1]. Hippocampal volumes were then determined from the bias-corrected images using the FIRST algorithm in the FSL software processing stream[2]. Concurrently, intracranial volumes were determined using structural T2 MR images, also processed using SPM8 unified segmentation. The resulting grey matter, white matter, and cerebrospinal fluid probability maps were summed and a subject-dependent voxel threshold that ranged from 0.1 to 0.3 was applied to generate a binary map. Intracranial volumes were calculated from binary maps encompassing brain tissue and CSF. Hippocampal values were normalized to intracranial volume.

ASL-MRI

The ASL perfusion data used a pulsed-ASL tagging scheme with a 3D-interleaved spiral fast spin echo readout with the following parameters: spatial resolution = 240 x 240 x 109 mm, TR = 4000 ms, TE = 18 ms, bandwidth = 2790 Hz/Px, slice thickness = 4 mm with 1 mm gap, 22 slices, 0.7 sec labeling duration, 45 msec acquisition time, and 1.2 sec delay time. Untagged control images were also acquired with parameters: spatial resolution = 240 x 240 x 109 mm, TR = 8000 ms, TE = 18 ms, bandwidth: 2790 Hz/Px, slice thickness: 4 mm with 1 mm gap, and number of slices: 22. ASL data were processed by the SPM8-(Statistical Parametric Mapping; http://www.fil.ion.ucl.ac.uk/spm/) based University of Pennsylvania script "Perf_reconstruct_SPM8.m"[3]. Images were realigned and mean control and tagged ASL images were coregistered to the high-resolution anatomical image. The coregistered images were smoothed using a 12-mm FWHM Gaussian filter. Subtraction images of tagged and control images were calculated using the sinc subtraction approach[4].

PET

The subject's head was immobilized to minimize head motion during the scan. PiB was injected intravenously (~15 mCi, over 20 s, specific activity ~1-2 Ci/μmol) and dynamic PET scanning was performed over 30 minutes (40-70 minutes post-injection, 6x5 min frames). Regional PiB uptake values were determined over the 50-70 minute post-injection period and normalized to injected dose and body mass to generate standardized uptake values (SUV). FDG was injected intravenously (~7 mCi) 2 hours after the start of the PiB scan.

The FDG-PET scan was acquired over 25 min (5x5 min frames) after a 35 min uptake period as the subjects rested quietly in a dimly lit room with their eyes open. The FDG-PET data were analyzed via summed images in a manner consistent with established methods[5,6]. The FDG data were summed over 40 to 60 min post-injection (4 frames) and SUV was obtained by normalizing tissue radioactivity concentration (μCi/ml) by injected dose (mCi) and body mass (in units of ml, making the approximation that 1 g equals 1 ml).

eTable. CMB Topography

Subjects with CMBs	21
Number of CMBs	54
Max single subject CMB count	10
Subjects with cortical CMBs	11
Number of cortical CMBs	20
Lobar CMB	
Parieto-occipital	21 (39%)
Cortical	12
Subcortical white	9
Frontal	11 (20%)
Cortical	3
Subcortical white	8
Temporal	11 (20%)
Cortical	5
Subcortical white	6
Deep and Infratentorial CMB*	
Cerebellar	6 (11%)
Deep	4 (7%)
Brainstem	1 (2%)

*Deep region includes basal ganglia and thalamus.

eReferences

1. Ashburner J, Friston KJ. Unified segmentation. *Neuroimage.* Jul 1 2005;26(3):839-851.

2. Patenaude B, Smith SM, Kennedy DN, Jenkinson M. A Bayesian model of shape and appearance for subcortical brain segmentation. *NeuroImage.* Jun 1 2011;56(3):907-922.

3. Wang J, Aguirre GK, Kimberg DY, Roc AC, Li L, Detre JA. Arterial spin labeling perfusion fMRI with very low task frequency. *Magnetic resonance in medicine : official journal of the Society of Magnetic Resonance in Medicine / Society of Magnetic Resonance in Medicine.* May 2003;49(5):796-802.

4. Aguirre GK, Detre JA, Zarahn E, Alsop DC. Experimental design and the relative sensitivity of BOLD and perfusion fMRI. *NeuroImage.* Mar 2002;15(3):488-500.

5. Minoshima S, Frey KA, Koeppe RA, Foster NL, Kuhl DE. A diagnostic approach in Alzheimer's disease using three-dimensional stereotactic surface projections of fluorine-18-FDG PET. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine.* Jul 1995;36(7):1238-1248.

6. Ziolko SK, Weissfeld LA, Klunk WE, et al. Evaluation of voxel-based methods for the statistical analysis of PIB PET amyloid imaging studies in Alzheimer's disease. *Neuroimage.* Oct 15 2006;33(1):94-102.