Effects of the Use of Multiple Scanners and of Scanner Upgrade in Longitudinal Voxel-based Morphometry and Diffusion Tensor Imaging Studies

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Purpose

Structural magnetic resonance (MR) imaging and diffusion tensor imaging (DTI) have been widely used to study gray matter morphology and the integrity of white matter tracts in healthy brains and in a variety of neurological diseases. There has recently been growing interest in longitudinal, multi-center studies such as the Alzheimer’s Disease Neuroimaging Initiative (ADNI) study. Longitudinal studies avoid some of the problems of secular trends and between-subject variation because each subject forms his or her own control. The statistical power to detect brain changes can, however, be limited by measurement errors. Previous studies have evaluated the effects of different scanners on cross-sectional or longitudinal results; however, we are not aware of any study that has simultaneously investigated the effects of inter-scanner variability on longitudinal morphometric and DTI results in relation to the effects of scanner upgrade. The purpose of this study was to evaluate the effects of inter-scanner variability (bias) and of scanner upgrade on longitudinal changes in regional gray matter volume and white matter diffusion properties.
Methods and Materials

Subjects

A total of 234 normal subjects (65 females and 169 males, mean age = 57 ± 9 years, age range = 40-84 years) were included in this study [1]. None of the subjects had a history of neuropsychiatric disorder including serious head trauma, psychiatric disorders, or alcohol/substance abuse or dependence. Mean Mini-Mental State Examination (MMSE) score was 29.6 ± 0.7 (range = 27-30). Each subject was scanned twice, at an interval of about 1 year (mean interval = 1.0 ± 0.1 years, range = 0.6-1.3 years). A board-certified radiologist reviewed all scans (including T1-weighted and T2-weighted images) and found no gross abnormalities such as infarct, hemorrhage, or brain tumor in any of the subjects. Fazekas score (range, 0-3) was 0 (absence) or 1 (caps, pencil-thin lining and/or punctuate foci) [2]. The ethical committee of the University of Tokyo Hospital approved the study. After a complete explanation of the study to each subject, written informed consent was obtained.

Imaging Data Acquisition

MR data were obtained on two 3.0-T Signa scanners (GE Medical Systems, Milwaukee, WI) with an 8-channel brain phased array coil. Both scanners were the exact same model, and were simultaneously upgraded from HDx to HDxt during the study period. Software alone was upgraded; hardware remained unchanged. The subjects were grouped as follows: Group A, baseline images were obtained on scanner 1 and follow-up images were obtained on scanner 1 (n = 74); Group B, baseline images were obtained on scanner 1 and follow-up images were obtained on scanner 2 (n = 49); Group C, baseline images were obtained on scanner 2 and follow-up images were obtained on scanner 1 (n = 58); and Group D, baseline images were obtained on scanner 2 and follow-up images were obtained on scanner 2 (n = 53) (Table 1). Of the 234 subjects, 165 underwent baseline and follow-up scans before upgrade, and the remaining 69 underwent a baseline scan before upgrade and a follow-up scan after upgrade. Of the 234 subjects, 19 and 10 subjects were excluded from voxel-based morphometry (VBM) and tract-based spatial statistics (TBSS) analysis, respectively, because of poor image quality (motion/artifact).

Table 1. Subject characteristics

<table>
<thead>
<tr>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Group D</th>
</tr>
</thead>
<tbody>
<tr>
<td>(1/1)</td>
<td>(1/2)</td>
<td>(2/1)</td>
<td>(2/2)</td>
</tr>
<tr>
<td>N</td>
<td>74</td>
<td>49</td>
<td>58</td>
</tr>
<tr>
<td>Age (yrs)</td>
<td>58 ± 10</td>
<td>57 ± 10</td>
<td>57 ± 10</td>
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<td>----------</td>
<td>---------</td>
<td>---------</td>
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</tr>
<tr>
<td>Sex</td>
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</tr>
<tr>
<td>male</td>
<td>12</td>
<td>15</td>
<td>26</td>
</tr>
<tr>
<td>Upgrade</td>
<td>(-)</td>
<td>(+)</td>
<td>(+)</td>
</tr>
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</table>

Note: Of the 234 subjects, 19 and 10 were excluded from VBM and TBSS analysis, respectively, because of poor image quality.

T1-weighted images were acquired using three-dimensional (3D) inversion recovery prepared fast spoiled gradient recalled acquisition in the steady state (IR-FSPGR) in 176 sagittal slices (repetition time = 5.3-5.4 ms; echo time = 1.7 ms; inversion time = 450 ms; flip angle = 15°; field of view = 250 mm; slice thickness = 1.0 mm with no gap; acquisition matrix = 256 x 256; number of excitations = 0.5; image matrix = 256 x 256; ASSET [Array Spatial Sensitivity Encoding Technique], acceleration factor = 2.0).

Diffusion tensor images were acquired using a single-shot spin-echo echo-planar sequence in 50 axial slices (repetition time = 13,200 ms; echo time = 62 ms; field of view = 288 mm; slice thickness = 3 mm with no gap; acquisition matrix = 96 x 96; number of excitations = 1; image matrix = 256 x 256; ASSET, acceleration factor = 2.0). Diffusion weighting was applied along 13 non-collinear directions with a \( b \)-value of 1,000 s/mm\(^2\) and a single volume was collected with no diffusion gradients applied \( (b_0) \).

**Voxel-based Morphometry (VBM)**

Image analysis was performed using statistical parametric mapping (SPM) 8 software (http://www.fil.ion.ucl.ac.uk/spm) developed in the Wellcome Department of Imaging Neuroscience, Institute of Neurology, University College London, running in MATLAB 7.13.0 (Mathworks, Sherborn, MA).

First, the IR-FSPGR images were corrected for spatial distortion due to gradient non-linearity using 'grad_unwarp' [3] and for intensity non-uniformity using the nonparametric non-uniform intensity normalization algorithm N3 [4]. The images were then segmented into gray matter, white matter, and cerebrospinal fluid using an integrated generative model (unified segmentation) [5]. The International Consortium for Brain Mapping (ICBM) gray matter, white matter, and cerebrospinal fluid templates were used as priors to segment the images. The Diffeomorphic Anatomical Registration Through Exponentiated
Lie Algebra (DARTEL) algorithm [6] was used to spatially normalize the segmented images. The normalized gray matter images were modulated to correct voxel signal intensity for volume displacement during normalization and reflect brain volume [7], and smoothed using an 8 mm kernel. The baseline gray matter images were then subtracted from the follow-up gray matter images.

Subtraction images were analyzed with SPM 8 employing the framework of the general linear model [8]. First, we identified areas with significant longitudinal changes in gray matter volume in each group (A-D). Scanner upgrade, age, and sex were included as covariates of no interest. Next, we identified areas where there was a significant effect of scanner upgrade on longitudinal changes. Significance levels for $t$ tests (one-tailed) were set at $p = 0.025$, corrected for multiple comparisons using the family-wise error (FWE) rate (voxel-level). We computed two $t$ contrasts (positive, negative) for $t$ tests. Only voxels with a volume greater than 0.1 were included in analyses.

**Tract-based Spatial Statistics (TBSS)**

Image analysis was carried out using tract-based spatial statistics (TBSS) 1.2 [9], part of FSL (FMRIB Software Library 4.1, http://www.fmrib.ox.ac.uk/fsl).

First, the raw diffusion data were corrected for eddy current distortion and head motion using FMRIB's Diffusion Toolbox (FDT) 2.0 [10], and corrected for spatial distortion due to gradient non-linearity using 'grad_unwarp' [3]. Following brain extraction using Brain Extraction Tool (BET) 2.1 [11], fractional anisotropy (FA) and mean diffusivity (MD) maps were created by fitting a tensor model to the diffusion data using FDT. All subjects' FA data were then aligned into Montreal Neurological Institute (MNI) 152 space using FMRIB's nonlinear registration tool (FNIRT) 1.0 [10], which uses a b-spline representation of the registration warp field. The FMRIB58 FA standard-space image was used as the target. Next, a mean FA image was created and thinned to create a mean FA skeleton representing the centers of all tracts common to the group. The mean FA skeleton image was thresholded at a FA value of 0.2 to prevent inclusion of non-skeleton voxels [9]. Each subject's aligned FA data were then projected onto this skeleton. The MD data were also aligned into MNI 152 space and projected onto the mean FA skeleton, using the FA data to find the projection vectors. Then, baseline skeleton-projected FA and MD data were subtracted from follow-up skeleton-projected FA and MD data, respectively.

Voxelwise analyses of subtraction images were performed using permutation-based, voxelwise non-parametric testing [12] (as implemented in the randomise tool, part of FSL). First, we identified areas with significant longitudinal changes in each group (A-D). Scanner upgrade, age, and sex were included as covariates of no interest. Next, we identified areas with a significant effect of scanner upgrade on longitudinal changes. Significance levels for $t$ tests (one-tailed) were set at $p = 0.025$, corrected for multiple comparisons using the FWE rate (voxel-level). We computed two $t$ contrasts (positive, negative) for $t$ tests. The number of permutations was 5000.
Results

Longitudinal Changes in Gray Matter Volume

Effects of Scanner

Voxel-based analysis of subtraction images revealed a number of regions with a significant increase or decrease in gray matter volume in groups in which the baseline and follow-up images were obtained on different scanners (B and C) (Fig. 1). There was a trend for opposite directions of change in these two groups (B and C). In groups in which both the baseline and follow-up images were obtained on the same scanner (A and D), however, only small regions showed significant longitudinal changes.
Fig. 1: VBM analysis of longitudinal (1-year) gray matter volume changes. The rows show an analysis for each group (A-D).
References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

Figure 2 shows beta images obtained from the above analysis, representing longitudinal changes in each group with the effects of scanner upgrade, age, and sex removed. Figure 3 shows histograms of the beta images (histogram bin width, 0.002; range, -0.1 to 0.1). In groups in which both the baseline and follow-up images were obtained on the same scanner (A and D), almost no voxels showed a change of less than -0.02 or more than 0.02 (A, 0.1%; D, 0.8%). In groups in which the baseline and follow-up images were obtained on different scanners (B and C), however, approximately 8% of voxels showed a change of less than -0.02 or more than 0.02 (B, 8.8%; C, 6.9%).
Fig. 2: VBM analysis of longitudinal (1-year) gray matter volume changes: beta images, representing longitudinal changes in each group with the effects of scanner upgrade, age, and sex removed.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
**Fig. 3**: VBM analysis of longitudinal (1-year) gray matter volume changes: histograms of beta images.

**References**: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

**Effects of Scanner Upgrade**

Voxel-based analysis of the subtraction images revealed a number of regions that showed a significant effect of scanner upgrade on longitudinal changes in gray matter volume (Fig. 4). There were both positive and negative changes due to scanner upgrade. The effects of upgrade of scanner 2 were similar to those of upgrade of scanner 1.
Fig. 4: VBM analysis of the effects of scanner upgrade on longitudinal (1-year) gray matter volume changes. The rows show the results of analysis of the effects of upgrade for each scanner.

References:
Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo, Japan

Figure 5 shows beta images obtained from the above analysis, representing changes due to scanner upgrade with the effects of group, age, and sex removed. Figure 6 shows histograms of the beta images (histogram bin width, 0.002; range, -0.1 to 0.1). Approximately 8% of voxels showed a change of less than -0.02 or more than 0.02 (upgrade of scanner 1, 7.6%; upgrade of scanner 2, 7.7%).
Fig. 5: VBM analysis of the effects of scanner upgrade on longitudinal (1-year) gray matter volume changes: beta images, representing changes due to upgrade of each scanner with the effects of group, age, and sex removed. 

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

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Standard Deviation Maps

Figure 7 shows standard deviations of longitudinal changes in gray matter volume for each group. Figure 8 shows histograms of the standard deviation maps (histogram bin width, 0.002; range, 0.0 to 0.2).
Fig. 7: Standard deviations of longitudinal (1-year) gray matter volume changes in each group (A-D).

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 8: Histograms of standard deviation maps of longitudinal (1-year) gray matter volume changes.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

Longitudinal Changes in White Matter FA and MD

Effects of Scanner

Voxelwise analysis of subtraction images revealed a number of regions with significant longitudinal changes in white matter FA and MD in groups in which the baseline and follow-up images were obtained on different scanners (B and C) (Figs. 9 and 10). There was a trend for opposite directions of change in these two groups (B and C). In groups in which both the baseline and follow-up images were obtained on the same scanner (A and D), there were no significant longitudinal changes except for a few voxels.
Fig. 9: TBSS analysis of longitudinal (1-year) white matter FA changes. The rows show an analysis for each group (A-D).

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 10: TBSS analysis of longitudinal (1-year) white matter MD changes. The rows show an analysis for each group (A-D).

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

Figures 11 and 12 show beta images representing longitudinal changes in each group with the effects of scanner upgrade, age, and sex removed. Figures 13 and 14 show histograms of the beta images (FA: histogram bin width, 0.002; range, -0.1 to 0.1; MD $[10^{-3} \text{ mm}^2/\text{s}]$: histogram bin width, 0.01; range, -0.5 to 0.5). In groups in which both the baseline and follow-up images were obtained on the same scanner (A and D), almost no voxels showed a change in FA of less than -0.02 or more than 0.02 (A, 0.1%; D, 0.4%) or a change in MD of less than -0.04$10^{-3}$ mm$^2$/s or more than 0.04$10^{-3}$ mm$^2$/s (A, 0.8%; D, 1.3%). In groups in which the baseline and follow-up images were obtained on different
scanners (B and C), however, approximately 7% of voxels showed a change in FA of less than -0.02 or more than 0.02 (B, 6.9%; C, 7.2%) and approximately 3% of voxels showed a change in MD of less than $-0.04 \times 10^{-3}$ mm$^2$/s or more than $0.04 \times 10^{-3}$ mm$^2$/s (B, 3.2%; C, 2.9%).

**Fig. 11:** TBSS analysis of longitudinal (1-year) white matter FA changes: beta images, representing longitudinal changes in each group with the effects of scanner upgrade, age, and sex removed.

**References:** Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 12: TBSS analysis of longitudinal (1-year) white matter MD changes: beta images, representing longitudinal changes in each group with the effects of scanner upgrade, age, and sex removed.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
**Fig. 13**: TBSS analysis of longitudinal (1-year) white matter FA changes: histograms of beta images.

**References**: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 14: TBSS analysis of longitudinal (1-year) white matter MD changes: histograms of beta images.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

Effects of Scanner Upgrade

Voxelwise analysis of the subtraction images revealed a number of regions with a significant effect of scanner upgrade on longitudinal changes in white matter FA and MD (Figs. 15 and 16). The effects of upgrade of scanner 2 were similar to those of upgrade of scanner 1.

Fig. 15: TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter FA changes. The rows show the results of analysis of the effects of upgrade for each scanner.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 16: TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter MD changes. The rows show the results of analysis for each scanner.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo, Tokyo, JP.

Figures 17 and 18 show beta images representing changes due to scanner upgrade with the effects of group, age, and sex removed. Figures 19 and 20 show histograms of the beta images (FA: histogram bin width, 0.002; range, -0.1 to 0.1; MD [×10^{-3} mm^2/s]: histogram bin width, 0.01; range, -0.5 to 0.5). Approximately 6% of voxels showed a change in FA of less than -0.02 or more than 0.02 (upgrade of scanner 1, 5.2%; upgrade of scanner 2, 6.3%), and approximately 5% of voxels showed a change in MD of less than -0.04×10^{-3} mm^2/s or more than 0.04×10^{-3} mm^2/s (upgrade of scanner 1, 4.6%; upgrade of scanner 2, 5.2%).
**Fig. 17:** TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter FA changes: beta images, representing changes due to upgrade of each scanner with the effects of group, age, and sex removed.

*References:* Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

**Fig. 18:** TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter MD changes: beta images, representing changes due to upgrade of each scanner with the effects of group, age, and sex removed.

*References:* Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 19: TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter FA changes: histograms of beta images.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 20: TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter MD changes: histograms of beta images.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP

Standard Deviation Maps

Figures 21 and 22 show standard deviations of longitudinal changes in white matter FA and MD, respectively, for each group. Figures 23 and 24 show histograms of the standard deviation maps (FA: histogram bin width, 0.002; range, 0.0 to 0.2; MD [x10^-3 mm^2/s]: histogram bin width, 0.01; range, 0.0 to 1.0)

Fig. 21: Standard deviations of longitudinal (1-year) white matter FA changes in each group (A-D).
Fig. 22: Standard deviations of longitudinal (1-year) white matter MD changes in each group (A-D).

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 23: Histograms of standard deviation maps of longitudinal (1-year) white matter FA changes.

References: Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
**Fig. 24:** Histograms of standard deviation maps of longitudinal (1-year) white matter MD changes.

**References:** Department of Radiology, Graduate School of Medicine, University of Tokyo - Tokyo/JP
Fig. 1: VBM analysis of longitudinal (1-year) gray matter volume changes. The rows show an analysis for each group (A-D).

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**Fig. 2:** VBM analysis of longitudinal (1-year) gray matter volume changes: beta images, representing longitudinal changes in each group with the effects of scanner upgrade, age, and sex removed.

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**Fig. 3:** VBM analysis of longitudinal (1-year) gray matter volume changes: histograms of beta images.

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**Fig. 4:** VBM analysis of the effects of scanner upgrade on longitudinal (1-year) gray matter volume changes. The rows show the results of analysis of the effects of upgrade for each scanner.

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Fig. 5: VBM analysis of the effects of scanner upgrade on longitudinal (1-year) gray matter volume changes: beta images, representing changes due to upgrade of each scanner with the effects of group, age, and sex removed.

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Fig. 6: VBM analysis of the effects of scanner upgrade on longitudinal (1-year) gray matter volume changes: histograms of beta images.

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**Fig. 20:** TBSS analysis of the effects of scanner upgrade on longitudinal (1-year) white matter MD changes: histograms of beta images.

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**Fig. 21:** Standard deviations of longitudinal (1-year) white matter FA changes in each group (A-D).

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Fig. 22: Standard deviations of longitudinal (1-year) white matter MD changes in each group (A-D).

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Fig. 23: Histograms of standard deviation maps of longitudinal (1-year) white matter FA changes.

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**Fig. 24:** Histograms of standard deviation maps of longitudinal (1-year) white matter MD changes.

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Conclusion

Using longitudinal (1-year) data obtained on two scanners of the exact same model, we examined the effects of inter-scanner variability (bias) and of scanner upgrade on longitudinal changes in regional gray matter volume and white matter diffusion properties. The results of the present study indicate that, even with scanners of the exact same model, the use of different scanners at different time points significantly influences longitudinal morphometric and DTI results. Regional gray matter volumes and white matter diffusion properties were relatively stable within the same scanner, but significantly different between the two scanners. Scanner upgrade had effects on longitudinal morphometric and DTI results comparable to those of using different scanners (of the exact same model) at different time points. To maintain consistency in longitudinal morphometric and DTI studies, scanner upgrade should be postponed if at all possible. The results of the present study are useful for planning not only longitudinal studies but also multi-center cross-sectional studies.
References

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