**Appendix A.**

**Supplemental Methods**

Subjects

Ten healthy young subjects between the ages of 20 and 44 (average age 31.0) were recruited for the study. Six were male (ages 20 to 44) and four were female (ages 21 to 36). Nine of 10 were right-handed. Subjects did not have any known neurological illnesses at time of enrollment. The study was approved by the University of Iowa Human Subjects Office and Institutional Review Board. All subjects signed a written consent and participated voluntarily.

MRI Acquisition

Subjects obtained 7-Tesla MRI structural brain scans within 30 days of participation. T1-weighted MRI sequences were obtained on a 7T GE MR950 scanner; sequence parameters varied slightly in an ongoing effort to optimize image quality at 7T. To harmonize image sequences and improve performance in Brainsight reconstruction software (Rogue Research, Montreal, Quebec), images were resampled to 1 mm isotropic voxels and the intensity range was truncated to standardize values from air-to-scalp-to-skull. Iterative tissue segmentation and N4 intensity inhomogeneity correction was applied to remove regional intensity differences throughout brain tissue, as per Tustison et al. (1) and Avants et al. (2). The processed images were loaded into Brainsight for neuronavigation-based measurements.

Determination of Scalp Targets using Beam F3 Method and 5.5 cm Rule

Three trained TMS technicians performed repeated scalp measurements on each subject at three time points over the course of 1 month, with at least 24 hours separating measurement sessions. The technicians also participated in the study as subjects and identified the stimulation targets on each other (n=162 total measurements, 81 at each scalp target). The Beam F3 and 5.5 cm rule target were both measured using standard measurement procedures (3-5) and plotted onto a Lycra swim cap with a marker. The Beam F3 method utilized freely available computer software (clinicalresearcher.org) and incorporated the adjustments as recommended by Mir-Moghtadaei et al. (6) The 5.5 cm rule motor hand knob was identified by visualization of thumb movement with the lowest stimulus dose necessary to induce movement in at least 3 of 5 consecutive stimulations separated by 6 seconds or more. A new swim cap was used for each measurement. Points were then registered on each subject’s anatomical image using the Brainsight neuronavigation system.

Comparison of Beam F3 Method and 5.5 cm Rule Scalp Targets

Distances between the targets as identified by the two methods were measured both on the scalp directly and using the Brainsight software. The anatomical images and associated coordinates on the surface of the head were warped from each subject’s native anatomical space into a common anatomical space (MNI152) and analyzed with Freesurfer. We first evaluated reliability between the Beam F3 method and the 5.5 cm rule. We separately evaluated inter-rater reliability relative to a group-averaged target coordinate and intra-rater reliability relative to a technician-specific average target coordinate. The Euclidean distances between each individual point and the average target coordinates were calculated. Euclidean distances were calculated in MNI152 space to control for variability introduced by head shape and size.In addition to reliability we calculated the average MNI coordinates that represent the scalp and brain coordinates from the Beam F3 and 5.5 cm methods. An MNI scalp reconstruction was created to show the spatial distribution of the Beam F3 and 5.5 cm rule targets, along with a group mean centroid for each method. Although the statistical analyses were run with all measured points, the average MNI coordinates were calculated after removal of any outliers, defined as 1.5 times above or below the interquartile range of the median, to minimize technician error in identification of the most representative valid Beam F3 and 5.5 cm rule coordinates.

Statistical Analysis

Generalized linear mixed models were used to test for differences in distance between F3 and 5.5 cm targeting methods. Random effects were included for subject and technician, with subject nested within technician. Mean ratios, 95% confidence intervals, and p-values were calculated to assess the comparison of targeting methods.

Converting Between Brain and Scalp Coordinates

Cortical targets were identified from technician-measured scalp targets by taking each individual coordinate on the scalp and moving it to the nearest vertex on the pial surface. Using Freesurfer, this involved computing a Euclidean distance from the scalp target to each pial vertex and selecting the vertex with the shortest distance. These coordinates were then transformed from the Freesurfer surface space to the subject’s volumetric space and subsequently to MNI152 space. In comparing our reported coordinates to those from other publications, we similarly adjusted all published coordinates to the pial and scalp surfaces for visualization purposes. For example, if a publication reported a coordinate off the pial surface, that point would first be adjusted to the nearest pial surface vertex, and from there to the nearest scalp vertex. Any coordinates reported in Talairach space were warped to MNI152 atlas space using a nonlinear registration computed with ANTs software (7-9); this involved application of a transform drawing upon the Washington University 711-2B Talairach atlas co-registered to MNI152 space. Then, the same process described above was used to identify the pial and scalp surface in Freesurfer.

**Appendix A References**

1. Tustison NJ, Avants BB, Cook PA, Zheng Y, Egan A, Yushkevich PA, et al. N4ITK: improved N3 bias correction. IEEE Trans Med Imaging. 2010;29(6):1310-20.

2. Avants BB, Tustison NJ, Wu J, Cook PA, Gee JC. An open source multivariate framework for n-tissue segmentation with evaluation on public data. Neuroinformatics. 2011;9(4):381-400.

3. George MS, Wassermann EM, Williams WA, Callahan A, Ketter TA, Basser P, et al. Daily repetitive transcranial magnetic stimulation (rTMS) improves mood in depression. Neuroreport. 1995;6(14):1853-6.

4. Pascual-Leone A, Catala MD, Pascual-Leone Pascual A. Lateralized effect of rapid-rate transcranial magnetic stimulation of the prefrontal cortex on mood. Neurology. 1996;46(2):499-502.

5. Beam W, Borckardt JJ, Reeves ST, George MS. An efficient and accurate new method for locating the F3 position for prefrontal TMS applications. Brain Stimul. 2009;2(1):50-4.

6. Mir-Moghtadaei A, Caballero R, Fried P, Fox MD, Lee K, Giacobbe P, et al. Concordance Between BeamF3 and MRI-neuronavigated Target Sites for Repetitive Transcranial Magnetic Stimulation of the Left Dorsolateral Prefrontal Cortex. Brain Stimul. 2015;8(5):965-73.

7. Van Essen DC. Windows on the brain: the emerging role of atlases and databases in neuroscience. Curr Opin Neurobiol. 2002;12(5):574-9.

8. Avants BB, Tustison NJ, Song G, Cook PA, Klein A, Gee JC. A reproducible evaluation of ANTs similarity metric performance in brain image registration. Neuroimage. 2011;54(3):2033-44.

9. Grabner G, Janke AL, Budge MM, Smith D, Pruessner J, Collins DL. Symmetric atlasing and model based segmentation: an application to the hippocampus in older adults. Med Image Comput Comput Assist Interv. 2006;9(Pt 2):58-66.