Supplemental material

MRI-guided TMS positioning.

We positioned the TMS coil on the locations of the subjects' scalp corresponding to the target cortical coordinates by using MRI guided information. Here we describe in more detail the technical aspects of this procedure.

Structural MRI. MRI head scans were obtained in all subjects with a Siemens Magnetom Allegra 3T with the following parameters: repetition-time 7.92 ms, echo-time 2.4 ms, inversion-time 910 ms, flip angle 15°, pixel bandwidth 128 Hz, 1 mm³ isotropic voxel size. We used MDEFT (Modified Driven Equilibrium Fourier Transform), a 3D inversion recovery gradient echo imaging sequence optimized to yield a high contrast-to-noise ratio at 3T (Deichman et al., 2004). To determine the location of the selected cortical sites on the individual brain, the structural MR image of each subject was first co-registered and normalized to the MNI space by using the linear registration tool (FLIRT) available in FSL (FMRIB, Oxford, UK). The MNI coordinates of the target cortical sites were projected on the subject's normalized brain image, and the resulting projection was checked for a possible mislocalization due to the normalization procedures. This verification is especially critical for brain regions characterized by appreciable inter-individual variability, such as the TPJ (Ozcan et al., 2005; Van Essen, 2005). When we detected an incongruence in the localization of the target site, we made slight adjustments of its coordinates based on identified anatomical landmarks: for instance, the Sylvian fissure, the supramarginal and superior temporal gyri for TPJ sites. The target sites defined in MNI coordinates were then re-transformed in the coordinates of the original MRI image using the co-registration transformation parameters. We finally determined the position of five different anatomical reference points on the subject's head in image coordinates: nasion, left or right incisurae intertragicae, lateral and
medial margin of the eyes orbits, mid point of the upper lip. The coordinates of these points were used to perform the transformation from the MRI image to physical space necessary to position the TMS coil over the corresponding sites on the subject’s scalp.

**TMS positioning.** Prior to each TMS experiment, we co-registered the MRI image in the physical space. To this end, the physical coordinates corresponding to the five anatomical reference points we had previously identified in the MRI image (see above) were acquired on the subject’s head by means of a stylus (Optotrak 6 markers probe) instrumented with six infrared emitting markers. These markers were monitored at 100 Hz by the Optotrak 3020 (Northern Digital Inc., Waterloo, Canada). Subjects were asked to keep the head fixed while we sampled the position of the five reference points; sampling was restarted if head displacements exceeded 2 mm. During this procedure, subjects were generally able to limit head displacements to less than 1.5 mm (mean translation of each marker). Physical coordinates of the reference points were then obtained by averaging three samples and these values were used to compute the six parameters of the rigid-body transformation linking image and physical reference frames. The six parameters were least-squares fitted following the procedure described by Neggers et al. (2004). The mean correspondence registration error (Neggers et al., 2004) was 3.8 mm (± 0.7-mm SD).

Using the previously defined rigid-body transformation, we computed the physical coordinates of the cortical sites. In order to guide the coil positioning, both the head and the coil positions were monitored at 100-Hz with the Optotrak system. To this end, six infrared emitting markers spaced evenly between the anterior (frontal) and lateral (temporal) side of the scalp were attached to the lycra cap donned by the subject. Five other markers were placed on the coil to monitor its position and orientation. A custom-made real-time graphical
interface helped to place the coil hotspot within a target volume of 7-mm radius centered on a given cortical target site. The coil was held firmly in place by means of a multiple degrees of freedom mechanical arm (Magic Arm, Manfrotto Italy); for the hMT/V5+ site, the coil was held tangential to the skull with the handle pointing backwards at about 90° to the spinal axis, while for the TPJ site the coil was held with the handle pointing upwards and medially. During each experimental session, we monitored the head and the coil positions at 10Hz, so as to detect any possible displacement of the coil hotspot outside the target volume.

We validated the accuracy of our positioning procedure *a posteriori* by means of an off-line analysis of the coil position data acquired during each experiment. Inaccuracies in the sampling of the reference point positions on the subject’s head in both MR image and physical coordinates were modeled as three-dimensional isotropic Gaussian distributions with SD=1.5 mm. This value was based on the average variability observed when repeating the physical sampling of reference points on the subject’s head. The SD observed when repeating sampling in the MR image equals about 1 mm. Uncertainty related to head displacement tracking proved to be negligible, due to the fact that co-registration occurs between sets of reference points placed on a rigid body. In order to provide an estimate of the accuracy of coil placement, we computed the variability of the centre position of the target region obtained when adding noise to the coordinates of the reference points measured in both MR image and physical reference frames. For a given target position, variability was determined subject by subject, adding random noise sampled from the above mentioned distributions to both physical and image coordinates and computing the corresponding target position in physical coordinates. The spatial distribution of 1000 perturbed target positions was computed as the 95% confidence ellipsoid centered on the average target position. We found that the axes of the confidence ellipsoid were always < 4 mm. Finally, an estimate of
the overall geometrical distortions was obtained from the ratio of distances computed in MR image and physical coordinates between pairs of reference points (and averaged over all possible pairs within a given subject). We found that the mean ratio between image to physical distance across all subjects was 1.03 voxel/mm (± 0.03 voxel/mm SD).

Localization and validation of TMS sites

The ideal locations of the target stimulation sites were determined a priori in normalized stereotatic space (MNI) on the basis of previous fMRI studies. For TPJ, we delimited a region of interest from the mean brain volume activated jointly by visual gravitational motion (using the contrast \([g \text{ trials} > -g \text{ trials}]\)) and vestibular caloric stimulation (using the contrast \([\text{left ear stimuli} + \text{right ear stimuli}] > \text{baseline}\)) in the fMRI study of Indovina et al. (2005). The findings of Indovina et al. (2005) have subsequently been replicated in additional studies performed by our group, yielding an overall database of 69 subjects (Maffei et al., 2007; Miller et al., 2008). The activation volume includes a complex of peri-Sylvian cortical areas in the insula, parietal operculum, supramarginal gyrus and posterior segment of the superior temporal gyrus. These areas are considered the core of the vestibular cortical network because they can be activated independently by direct vestibular stimulation (caloric or galvanic, see Bense et al., 2001; Bottini et al., 2001; Dieterich et al., 2003; Indovina et al., 2005). Moreover their direct electrical cortical stimulation can elicit vestibular sensations, such as body tilts (Blanke et al., 2000; Kahane et al., 2003). The regions deep within the Sylvian fissure (such as the insula) cannot be easily accessed with TMS. Thus, for the present TMS study we defined the target TPJ stimulation sites around two bilateral activation foci located on the surface of the supramarginal gyrus (BA 40) at MNI coordinates : ±62, -36, 32 and ±61, -29, 21.

With regard to area hMT/V5+, there is extensive neuroimaging literature defining its extent and functional properties. We used two independent methods to delimit the region of interest
corresponding to hMT/V5+, based on the available literature. First, we referred to the hMT/V5+ borders outlined in the visuotopic cortical partitioning scheme available in Caret (Van Essen, 2005). Second, we carried out an Activation Likelihood Estimate (ALE) meta-analysis (Turkeltaub et al., 2002) to obtain a probabilistic activation map starting from 52 activation foci reported by 28 visual motion studies as belonging to hMT/V5+. Then we centered the bilateral target sites at the MNI coordinates (±52, -71, 0 mm) of the activation foci reported by Orban et al. (2003) and verified post-hoc that stimulation sites at least fell within both the probabilistic activation map and the borders defined by Caret’s scheme. We found that in all cases these criteria were satisfied.
REFERENCES


