Evoked potentials in large-scale cortical networks elicited by TMS of the visual cortex

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Garcia JO, Grossman ED, Srinivasan R. Evoked potentials in large-scale cortical networks elicited by TMS of the visual cortex. J Neurophysiol 106: 1734–1746, 2011. First published June 29, 2011; doi:10.1152/jn.00739.2010.—Single pulses of transcranial magnetic stimulation (TMS) result in distal and long-lasting oscillations, a finding directly challenging the virtual lesion hypothesis. Previous research supporting this finding has primarily come from stimulation of the motor cortex. We have used single-pulse TMS with simultaneous EEG to target seven brain regions, six of which belong to the visual system (left and right primary visual area V1, motion-sensitive human middle temporal cortex, and a ventral temporal region), as determined with functional MRI-guided neuronavigation, and a vertex “control” site to measure the network effects of the TMS pulse. We found the TMS-evoked potential (TMS-EP) over visual cortex consists mostly of site-dependent theta- and alphaband oscillations. These site-dependent oscillations extended beyond the stimulation site to functionally connected cortical regions and correspond to time windows where the EEG responses maximally diverge (40, 200, and 385 ms). Correlations revealed two site-independent oscillations ~350 ms after the TMS pulse: a theta-band oscillation carried by the frontal cortex, and an alpha-band oscillation over parietal and frontal cortical regions. A manipulation of stimulation intensity at one stimulation site (right hemisphere V1-V3) revealed sensitivity to the stimulation intensity at different regions of cortex, evidence of intensity tuning in regions distal to the site of stimulation. Together these results suggest that a TMS pulse applied to the visual cortex has a complex effect on brain function, engaging multiple brain networks functionally connected to the visual system with both invariant and site-specific spatiotemporal dynamics. With this characterization of TMS, we propose an alternative to the virtual lesion hypothesis. Rather than a technique that simulates lesions, we propose TMS generates natural brain signals and engages functional networks.

transcranial magnetic stimulation; electroencephalogram; vision

TRANSCRANIAL MAGNETIC STIMULATION (TMS) studies are often conceived as creating a very fast, focal, and reversible interruption of neural processes at the site of stimulation, effectively disabling neural function or introducing neural noise (Harris et al. 2007), and have been likened to creating a virtual lesion (Amassian et al. 1989; Pascual-Leone et al. 2000). However, evidence inconsistent with this hypothesis demonstrates that the action of TMS is much more complex than simply disabling a region of cortex (e.g., Sack et al. 2006). Brain mapping studies using concurrent imaging techniques (e.g., PET, EEG) have measured responses to the TMS pulse distal from the stimulation site, typically in regions believed to be functionally connected to the stimulation site (functional (f)MRI: Bohning et al. 1998; Baudewig et al. 2001; PET: Fox et al. 1997; Laird et al. 2008; Paus et al. 1997; and single-photon emission computed tomography (SPECT): Okabe et al. 2003). For example, TMS over the frontal eye field engages a whole network of brain areas as measured by PET, including regions in occipital and parietal cortex (Paus et al. 1997). Simultaneous EEG measurements have shown TMS-induced brain activity that spreads across cortex very quickly (Paus et al. 2001b) and travels as far as to the opposite hemisphere within 30 ms following stimulation (Ilmoniemi et al. 1997). Thus the action of the TMS pulse at one location apparently engages large-scale cortical networks, not just the focal region that receives the stimulation.

There are a number of mechanisms that determine how TMS engages cortical networks to elicit neural responses. One consideration is the underlying morphology of the stimulation site, which is differentially impacted by the TMS pulse depending on a number of factors. For example, the orientation of the cortical grey matter relative to the coil is one of these factors where the horizontal cells at the fundus of the sulcus and surface of the gyrus (Day et al. 1989) and pyramidal cells in the sulcal wall are thought to be preferentially stimulated (Fox et al. 2004). Further, anisotropy and heterogeneity of the brain tissue underlying the stimulation site can be expected to significantly impact the spatial distribution of the induced electrical field (De Lucia et al. 2007).

Although detailed finite element (FEM) models have been used to estimate the induced electrical field at the stimulation site (Salvador et al. 2010; Silva et al. 2007), such models still do not account for the synaptic interactions of the stimulated neurons within the functional network engaged by the cascade of neural firing following a TMS pulse. The functional connectivity of the stimulation site constrains the excitation to the nodes that are dynamically linked to the stimulation region (Amassian et al. 1998; Ruohonen and Ilmoniemi 1999). A number of EEG studies have measured stimulation site-specific TMS-evoked potentials (TMS-EP) time locked to the TMS pulse onset with, for example, a wave of activity that spreads across cortex, rather than remaining localized to a particular region, following single-pulse TMS-EEG over motor cortex (Bonato et al. 2006). Simultaneous EEG studies have also measured the induced oscillations (i.e., modulations of power of ongoing EEG activity) from single TMS pulses, with resulting transient oscillations apparent in the alpha and beta bands (Fuggetta et al. 2005; Ilmoniemi et al. 1997; Paus et al. 2001a; Rosanova et al. 2009; Van Der Werf and Paus 2006). Thus EEG recordings are consistent with the idea that TMS pulses stimulate other cortical regions with spatiotemporal properties that reflect networks functionally connected to the stimulation site.
In this study, we seek to characterize the spatiotemporal dynamics of visual responses to single TMS pulses using simultaneous EEG recordings. Previous studies investigating functional connectivity within the visual system have limited the regions and measurements used. For example, functional connectivity has been previously investigated, finding visual cortical responses are affected by TMS to frontal and parietal cortical areas (Ruff et al. 2008, 2009). We expand this literature by measuring emergent site-invariant and site-specific networks from visual cortical stimulation. We applied TMS over three functionally distinct visual areas [primary visual areas (V1-V3), motion-sensitive human middle temporal cortex (hMT+), and a ventral temporal (VT) region of each hemisphere], and a single unrelated “control” area (the vertex). Because the behaviorally perceptible impact of TMS is also stimulation intensity specific (e.g., the induction of phosphenes and scotomas, Kammer 1999; Kastner et al. 1998), we also measured the differences observed as a function of stimulation intensity.

Our results show that the evoked potential is dominated by low frequency theta and alpha oscillations (all <12 Hz) and is spectrally similar across stimulation of different areas of the visual system and the vertex. We measured stimulation-site specific (or “local”) responses, distributed mainly over the parietal and occipital cortex in the first 100 ms after stimulation. At longer time scales, we observed theta and alpha oscillations that were globally distributed and spatially similar across stimulation sites. A manipulation of stimulation intensity at one stimulation site (right hemisphere V1-V3) revealed sensitivity to the stimulation intensity at different regions of cortex. Together these results suggest that a TMS pulse applied to the visual cortex has a complex effect on brain function, engaging multiple brain networks functionally connected to the visual system with both invariant and site-specific spatiotemporal dynamics.

METHODS AND MATERIALS

Participants

Eight individuals (5 men, 3 women, aged 20–33) participated in both the TMS-EEG and functional (f)MRI portions of the experiment. All observers have normal or corrected-to-normal vision and gave informed, written consent as approved by the University of California, Irvine Institutional Review Board.

Procedures

fMRI. Before participating in the TMS-EEG portion of the experiment, subjects participated in an MRI/fMRI session to 1) acquire anatomical images of the individual’s brain, and 2) to localize the regions to be stimulated. Averaged fMRI activity and estimated stimulation sites are shown in Fig. 1. Briefly, the early visual areas (V1-V3 region) were identified via a traveling wave analysis (Engel et al. 1994). Subjects viewed 24-s sequences of a high-contrast, contrast-reversing checkerboard wedge (15° wide, 11.1° maximum length) rotating at a rate of 1 cycle every 24 s. The wedge made nine complete rotations within each scan, and each traveling wave scan was repeated twice. Phase-encoding maps were generated by visualizing the lags of the correlated [thresholded at P < 0.05, false discovery rate (FDR) corrected] neural response on the individual’s inflated cortical sheet in BrainVoyager (Brain Innovations). The early visual cortex was identified on the basis of meridian border reversals near the right and left occipital pole.

The VT regions were identified from the same traveling wave analysis as the lateral retinotopically organized brain areas (e.g., Larsson and Heeger 2006) approximately ventral and anterior to the LO-1, LO-2 maps (see Fig. 1 for approximations of the site of stimulation). The hMT+ was localized as the brain area on the ascending branch of the inferior occipital sulcus that was more activated during 14-s intervals of RDK optic flow motion (randomly switching between inward and outward motion) compared with 14-s intervals of stationary dots. Statistical threshold for significance was set using the FDR as a correction for family-wise error rate (P < 0.01, FDR corrected), which controls the proportion of expected false positives and has the advantage of being adaptive to the signal levels in data while still correcting for multiple comparisons (Genovese et al. 2002).

Finally, the vertex stimulation site was defined by skull landmarks rather than fMRI activity. It was set to the midpoint between the nasion and inion in the anterior-posterior direction, and between the tragus of the left and right ears, as is quite typical in EEG studies.

TMS-EEG. After the fMRI localization experiments, subjects participated in the TMS-EEG phase of the experiment on a separate day. Stimulation sites were targeted via a frameless stereotaxic guidance system operated in conjunction with Advanced Source Analysis software (Advanced Neurotechnology, The Netherlands). Subjects were seated in a comfortable chair ~120 cm from the monitor with their heads fixed in a chinrest to minimize movement. A three-pronged axis device was secured to a fixed position on the subject’s head and served to register a landmark position (0,0,0), together with fiducial landmarks (tragus of the outer ear, tip of the nose), that identified a coordinate system specific to the subject’s head. These landmarks were coregistered to the individual’s high-resolution anatomical brain images, which then allows targeting of the desired site of stimulation. The coil position was aligned based on maximizing the overlap between the region of maximum current induction and the localization of visual areas based on fMRI. Coil orientation was determined based on previous psychophysical literature where certain orientations maximally impact performance and create a magnetic field perpendicular to the sulcus of interest (e.g., Amassian et al. 1994; Brasil-Neto et al. 1992; Meyer et al. 1991). For early visual cortex, the coil was oriented parallel to the coronal view (e.g., paddle pointing up), while for MT and VT stimulation the coil was oriented parallel to the midline (e.g., paddle pointing back). These orientations show consistency between behavioral measures used to find these areas and fMRI-guided neuronavigation TMS (Campana et al. 2002; Sack et al. 2009).

![Stimulation Site](image.png)

Fig. 1. Stimulation sites. Head model created from an average MRI from all participants. Functional (f)MRI activity derived from a normalized activity is shown within the sample brain, P < .001, false discovery rate (FDR). Stimulation sites are shown as filled circles. V1-V3, primary visual areas; hMT+, motion-sensitive human middle temporal cortex; VT, ventral temporal region; R, right; L, left.
interval surrounding the pulse to remove any quick shifts in amplitude due to the artifact editing procedure. Automated artifact editing based on amplitude thresholds was used to eliminate 26 channels likely contaminated with artifacts, leaving 102 channels. Percentiles were calculated for each set of 75 trials for each site and subject. Channels and trials that were beyond the 95th percentile were discarded from the analysis. If a channel contained artifact in half of subjects, this channel was completely removed from the analysis. These measures assured that channels containing artifact from the TMS pulse and high amounts of blink artifact were discarded from the analysis. To avoid any residual artifact including eye blinks and subject movement, median evoked potentials were calculated for each stimulation site, channel, and subject. The evoked potentials were band-pass filtered using a Butterworth filter with 2-dB attenuation at 2 and 40 Hz. A total of 56 recordings of evoked potentials (8 subjects × 7 stimulation sites) was used for further analysis.

ANOVA and correlation analysis. To determine the regions that are significantly different across stimulation site, the TMS-EP was subjected to a 2 (hemisphere) × 3 (region) ANOVA across subjects, excluding the vertex stimulation site. We also computed an ANOVA separately for each stimulation hemisphere to measure the differences and the homogeneity of each stimulation hemisphere. To quantify the similarity of the TMS-EP between stimulation sites, we calculated the correlation coefficient between the spatial patterns at each time point. Simple effects were also calculated to isolate the regions driving the main effects.

Wavelet analysis. The majority of spectral power was carried by the evoked power in the TMS-EP, so a continuous wavelet transform of the TMS-EP was calculated with a complex Morlet wavelet with a two-cycle bandwidth for scales between 4 and 40 Hz. The similarity of the spatial pattern of these complex Wavelet coefficients was quantified with a squared correlation coefficient.

Head model creation and deblurring of EEG. For visualization and source analysis, all anatomical MRIs collected from each individual subject were transformed into a common space (Talairach and Tournoux 1988) and then averaged. Electrode positions were also transformed and then fit to the nearest vertex on the derived head model mesh image. A BEM head model was constructed using the Matlab Toolbox for volume conduction modeling developed at the Helsinki University of Technology (Stenroos et al. 2007). The head model consisted of three ellipsoids fit to the subject averaged, segmented MRI, representing the scalp and inner and outer boundaries of the skull (Srinivasan et al. 2007). The thickness of each layer was uniform across the elliptical surfaces; electrode positions were fit to the scalp ellipse. The use of a uniform thickness skull is essential to avoid generating errors due to thickness variations in the skull layer (Nunez and Srinivasan 2006; Srinivasan et al. 2007). The spatial distribution of the EEG signals was deblurred by using the BEM head model to estimate the potentials on the surface of the brain ellipse following the procedure given by Babloni et al. (1997). These estimates of cortical potential have been shown to be closely related to surface Laplacian methods (Babiloni et al. 1997; Nunez and Westdorp 1994; Nunez and Srinivasan 2006). Topographic maps show the de-blurred EEG map projected (nearest neighbor) to the cortical surface of a single subject’s normalized (Talairach and Tournoux 1988) brain. Brodmann areas were then found by finding the peak response within a potential cluster and finding the nearest Brodmann area to that peak. Note that this estimate of cortical potential does not disambiguate sources in adjacent gyral and sulcal surfaces.

RESULTS

TMS-Induced Oscillations and the Resonant Structure of the Recording Site

We applied single pulses of TMS to three functionally dissociated regions of visual cortex (in both hemispheres)

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2006; Schenk et al. 2005; Silvanto et al. 2005). Coil orientation for vertex stimulation was parallel to the axial plane with the paddle pointed back.

For each region, 75 single pulses of stimulation at 55% machine intensity output (E-field, 297 V/m) were applied to each stimulation site, no closer than 4+ apart, parameters that are regarded as safe (Rossi et al. 2009). Because we targeted visual cortex in this study, we used visual phosphophes as a guide for stimulation intensity as opposed to motor responses (as the 2 are not correlated), which has been the standard for several years of TMS-psychophysical investigations (Stewart et al. 2001). Stimulation intensity was set to be ~80% of the phosphophes threshold, based on the subset of subjects that experienced phosphophes when pulsed over early visual cortex (3/8). The remainder of the subjects (5/8) did not experience phosphophes; this is most likely the lower bound of phosphophes induction across subjects. All seven regions were collected within two sessions, with the order of stimulation blocked by targeted area randomized across subjects, and subjects wore earplugs to minimize the auditory “click” of the TMS.

Three subjects participated in an additional control experiment with four levels of stimulation intensity of right V1, ranging from 30 to 70% stimulation intensity with corresponding E-field estimates of 162–378 V/m.

Materials

TMS-EEG. The TMS-EEG experiments were conducted in the TMS/EEG Laboratory in the Human Neuroscience Laboratory at the University of California, Irvine. Stimulation was conducted with a MagStim Model 200 Monophasic Stimulator P/N 3010–00 equipped with a figure-of-eight coil (70 mm, P/N 3190–00). EEG measurements were made with TMS-compatible EEG system (ANT) fitted with a Waveguard cap system using small Ag/AgCl electrodes and a standard 128 channel, high-input impedance amplifier. The EEG signals were recorded with an online average reference and digitized at 1,024 Hz.

fMRI. Neuroimaging data were collected on a 3T Philips Intera Achieva Magnetic Resonance system located at the University of California, Irvine. Visual displays were projected with a Christie DLV1400-DX DLP projector controlled by a G4 dual-processor Macintosh computer equipped with Matlab (Mathworks) and the Psychophysics Toolbox (Brainard 1997; Pelli 1997). Subjects viewed animations used in functional localization of the visual areas via a periscope mirror mounted on an eight-channel birdcage headcoil, directed at a screen positioned behind the subject’s head.

For each individual we collected high-resolution (T1-weighted, MPRAGE) whole-brain anatomical images used for coregistration of the functional data. Functional data were collected using single-shot T2*-weighted parallel imaging (SENSE reduction factor R = 1.5; gradient EPI, TE = 30 ms, flip angle = 70°, AP phase-encoding, interleaved acquisition) with slices that covered nearly the entire brain (32 ACPC-aligned slices, 1.8 × 1.8 × 4 mm voxels, TR = 20.00 ms).

Analysis

Segmentation. Raw EEG data (containing the pulse artifact) were subjected to a principal component’s analysis for segmentation into epochs for each stimulation site in each subject. The maxima from the largest component of each data set was used to segment each block into 75, 2-s epochs, starting 1 s before pulse. Each trial was normalized by the SD of the 512 samples before the stimulation to standardize variability in EEG amplitudes across recording sessions and subjects. Due to the amplifier artifact introduced by TMS, the 4 samples before the pulse and 16 (15.6 ms) after the pulse were removed from the epochs. Only for visualization, these samples were later replaced with a forward autoregressive moving average prediction of the contaminated data from the intervals directly preceding the TMS pulse and a mild Savitzky-Golay smoothing filter over the
and the vertex. Figure 2 shows the resulting TMS-EP at all EEG channels with the channel closest to the stimulation site demarcated in red (using the 10–20 international naming convention, these stimulation sites were closest to electrodes O1/O2, PO5/PO6, PPO9H/PPO10H, and CZ, respectively).

Several features common across stimulation sites are clearly visible from the time course of the TMS-EP. The TMS-EP shows peaks in the potential at roughly 40, 200, and 385 ms across stimulation sites. In each case, the strongest responses are recorded distal to the electrode closest to the stimulation site. We measured the lowest amplitude oscillations from...
Spatial Correlations Reveal a Site-Invariant Response

To quantify the spatial similarities between the oscillations evoked at each stimulation site, the spatial pattern of the TMS-EP for each stimulation site was correlated with every other site at each time point. Figure 4 displays the results from this analysis. For convenience, we only show the correlation with the pair of early stimulation sites (LV1/RV1) as these capture the essential effects. Directly after the TMS pulse,
within the window containing the first peak of the TMS-EP (40 ms; Fig. 2), the correlations between stimulation sites clearly separate by hemisphere. The evoked response for left hemisphere stimulation is positively correlated with other left hemisphere stimulation sites and negatively correlated with right hemisphere stimulation sites and vice versa for right hemisphere stimulation. Vertex stimulation is uncorrelated with all the other stimulation sites at this time point.

These correlation estimates also reveal two time points where the spatial pattern across all stimulation sites are highly similar, maximally at 116 and 292 ms after the TMS pulse, with an average $r = 0.82$, measured between all possible pairs of stimulation regions. The high correlations fall on the decline of the peaks of the TMS-EP; however, inspection of the source distribution shows that many sources are still reliably higher than baseline EEG ($z > 1.96$). This site-invariant signal has a periodicity of $\sim 150 – 170$ ms and is primarily observed within the dorsal parietal/frontal and posterior occipital cortex, with a spatial distribution reminiscent of the P300 evoked potential (Sutton et al. 1965; Basar et al. 1984).

Correlations of Evoked Power

A wavelet analysis was carried out on the TMS-EP to characterize its frequency content within the theta (4–6 Hz), alpha (8–12 Hz), beta (15–28 Hz), and gamma (30–40 Hz) bands and the correlation between the spatial distributions of wavelet coefficients observed with different stimulation sites. Summed over channels, most of the power of the TMS-EP was found in the theta (69% of total power) and alpha (22% of total power) bands. Correlations were calculated between the complex valued (magnitude and phase) wavelet coefficients, and the results are presented as squared correlation coefficients ($r^2$). Only the theta and alpha bands exhibited significant correlation of the spatial patterns between stimulation sites. Figure 5B shows the power in the theta and alpha to peak shortly (40 ms) after stimulation and decline over the following 700 ms. In contrast, the correlations (Fig. 5A) peak at $\sim 350$ ms for all stimulation sites except VT cortex (both hemispheres). The widespread activity within the first 40 ms following VT stimulation, and the overlap between this network and that of right early visual cortex stimulation, may account for the earlier correlation apparent for the left VT/right VT stimulation (see Fig. 2). Finally, spatial patterns of this oscillatory activity reveal a theta-band oscillation focused in the dorsal parietal/frontal cortex, and an alpha-band oscillation primarily driven by frontal and posterior parietal regions (Fig. 5C).

Differential “Intensity-Tuning” Distal to the Stimulation Sites

Previous EEG studies have isolated specific components of the TMS-evoked response as being invariant to changes in stimulation (e.g., a result of exposure to an external event rather than injection of current into the brain, per se), with others being modulated by stimulation intensity (e.g., Kahkonen et al. 2005). To test intensity dependence of our observed responses, we have stimulated right hemisphere early visual cortex (RV1) in three subjects and measured the brain response to four different levels of stimulation intensity 30, 50,
The idea of distal and long-lasting oscillations as a result of TMS directly challenges the so-called virtual lesion hypothesis. Previous evidence against this hypothesis has come from simultaneous TMS-EEG studies of motor cortex, showing the propagation of neural activity following a single pulse of TMS (e.g., Bonato et al. 2006; Ilmoniemi et al. 1997; Paus et al. 2001b) or from other concurrent functional neuroimaging studies that measure neural activity in distant brain sites (fMRI:...
Bohning et al. 1998; Baudewig et al. 2001; PET: Fox et al. 1997; Laird et al. 2008; Paus et al. 1997; and SPECT: Okabe et al. 2003). Our results extend this literature by providing evidence that the time course of the cortical response to a single pulse of TMS is sufficiently long (≈700 ms) to impact multiple cognitive functions and that stimulating one region of cortex has broad impact on a network of brain areas. Critically, by showing that the responses have a complex interaction with stimulation intensity, we argue that our measurements reflect TMS-induced cortical oscillations, rather than endogeneous activity.

Site Specificity of Single-Pulse TMS

TMS induces responses in networks that depend on the stimulation site. The correlation analyses as well as the raw evoked potential showed site-specific and large network oscil-
lutions unique for each stimulation site. The implication is that
the entire network of brain regions engaged by TMS is depend-
ent on connectivity of that site to the rest of the brain. For
visual cortex stimulation, we observed a theta-band oscillation
in occipital and parietal cortex, with the specific spatial distri-
bution depending mainly on the hemisphere of the stimulation
site. We also observed stimulation over the more VT site to
engage additional ventral cortical areas.

Most striking in these findings were the mirrored spatial
distributions for stimulation of corresponding cortical sites
(e.g., left and right V1) of each hemisphere where the elicited
parietal activity was always contralateral to the stimulation
site, and this effect is only dependent on hemisphere of stim-
ulation rather than any specific stimulation site in visual cortex.
Also, the activity near the site of stimulation was negatively
correlated across hemispheres. These oscillations were appar-
ent at 40 ms and had a time course that spans =500 ms. The
strong hemispheric structure of the observed responses is
perhaps not so surprising, since the three nodes of the visual
system that we stimulated are heavily interconnected and part
of an integrated system with common targets in parietal and
frontal cortex.

Interestingly, there were few occipital regions that show
significant differences between stimulation sites; the most
striking differences may be the comparison between the MT
stimulation and VT stimulation. MT stimulation resulted in
a very low amplitude response, where VT stimulation resulted
in widespread high magnitude responses, especially throughout
lateral occipital cortex. hMT+, the human homologue to mon-
key MT, has been the subject of study for decades, and its
response to visual information is determined by several prop-
erties of the stimulus, including size, speed, direction, location,
and binocular disparity (for review, see Born and Bradley
2005). The VT region, overlapping with the lateral occipital
complex, is less clearly mapped. VT cortex is known to be
involved in object recognition (e.g., Grill-Spector et al. 2001),
visual categorization (Thompson-Schill et al. 1999), and object
learning (Op de Beeck et al. 2006). Whereas MT is a relatively
small brain area engaged early in perceptual processing, the
lateral occipital complex is a large, homogenous region with
a range in tuning to natural objects and invariances to low-level
properties of the visual scene. This complexity would imply a
need for greater connectivity across the cortex, especially
lateral occipital and temporal cortex, as we have observed.

In contrast to the great deal of similarity of the occipital and
parietal responses to the three stimulation sites of each hemi-
sphere, we observed frontal and prefrontal responses that
depended strongly on the stimulation site primarily driven by
responses to VT stimulation. Thus we find evidence of distinct
large-scale functional connectivity of VT to distinct frontal and
prefrontal targets.

Site-Invariant Networks

The first site-invariant network we observed is the periodic
signal suggested by the spatial correlations in Fig. 4. The
periodic signal beginning 116 ms after TMS emerges on the
fall of the TMS-EP and has a spatial distribution reminiscent of
the P300 often observed in visual oddball studies (Sutton et al.
1965); however, the timing of this periodic signal is inconsis-
tent with the P300, which is not repeated, as is shown here at
116 and 292 ms. Instead, this periodic signal is quite unique to
TMS. Similar in spatial pattern is the increased power within
the theta band observable 300 ms after TMS for all of the
stimulation sites (Fig. 5). A stimulation site-invariant alpha
oscillation also appeared following the pulse. With the timing
alone, it may be suggested that these oscillations could reflect
an endogeneous theta/alpha rebound, as typically observed
several hundred milliseconds after a visual stimulus following
alpha blocking (e.g., Sauseng et al. 2005). However, our
intensity varying analysis does not support this interpretation.

The simulation-site invariant responses provide further evi-
dence of the global origins of many scalp EEG signals (Nunez
2000; Nunez and Srinivasan 2006). The cortex is characterized
by extensive connectivity by white-matter (corticocortical and
callosal) fiber systems that are both specific and diffuse (Brait-
enberg and Schuz 1991; Nunez 1995). These fiber systems
create large-scale neuronal networks, which are believed to
give rise to large-scale coherent oscillations such as spatially
coherent alpha rhythm, observable over the whole head with
EEG electrodes. Mathematical models suggest that these osci-
lations emerge from the delays imposed by the white-matter
connectivity (Nunez 2000; Nunez and Srinivasan 2006) rather
than intrinsic properties of the cortical circuits. TMS at one site
is propagated to many cortical regions via corticocortical and
callosal fiber systems over the cortex and apparently engages
global oscillatory modes in theta and alpha bands.

Previous TMS-EEG studies have also found an alpha-band
oscillation following TMS to occipital and parietal cortex.
Through neural entrainment with repetitive (r)TMS, enhance-
ment of this alpha network has even been found to be behav-
iorally relevant, modulating visual input processing (Romei
et al. 2010) and improving working memory capacity (Sauseng
et al. 2009). Single pulses of TMS have also previously been
found to elicit alpha activity with occipital stimulation (Rosan-
ova et al. 2009). Our results are consistent with the engage-
ment of these alpha band networks.

Complex Interaction with Stimulus Intensity Suggests
Functional Network Responses at Distant Sites

Although previous researchers have used intensity indepen-
dence as a diagnostic for an endogenous mechanism, other
factors such as the sensation of the pulse on the scalp or the
auditory evoked response may also linearly scale with inten-
sity. Our results show that brain regions distal to the stimula-
tion site do not have a monotonic response to stimulation
intensity. It is known that TMS affects only a subset of neurons
at the site of stimulation due to orientation or cycle of the
refractory period (Fitzpatrick and Rothman 2000); however,
the synaptic interactions are yet unknown. It seems plausible
that a change in stimulation intensity increases the probability
that electrical current of the induced field will cause an action
potential. If a “distant” network connected to the stimulation
site is engaged when large assemblies of neurons are active
within the stimulation site, then an increase in stimulation
intensity will more likely engage this functional network. It
should be noted that distant in the brain is actually not very
distant at all. It has been shown that a single synapse may reach
every other neuron in the brain in relatively few hops or jumps, reminiscent of a small world network (Bassett and Bullmore 2006).

If the probability of a neuron firing increases with stimulation intensity, then this alone would predict that higher stimulation intensity will engage larger networks with an increasing electrical field; however, we also know that the brain has compensatory mechanisms and inhibitory networks that may also be engaged, suppressing some networks and enhancing others. The complex excitation/inhibition interactions within the brain could act as gates, blocking the synthetic visual system response when engaged by a specific stimulation intensity or allowing its spread at another stimulation intensity.

Hemispheric Asymmetries

Interestingly, our results show asymmetries within both the magnitude of the evoked potential (compare left and right VT) and also the ANOVA comparing each region constrained to only one hemisphere, where the right hemisphere stimulation sites appear to be more homogenous than the left hemisphere stimulation sites. While hemispheric asymmetries are the hallmark of several cognitive phenomena (e.g., orienting in spatial attention), we have no a priori reason to expect lateralization in the connectivity from early visual cortex or extrastriate areas of the visual system. Moreover, we found that the resulting oscillations from TMS had more power when applied over the right hemisphere of any given stimulation site. This could reflect higher connectivity of the right hemisphere regions to the rest of the brain compared with the left hemisphere or simply that the anatomical characteristics of the regions stimulated in the left hemisphere were not as optimal (e.g., relative to coil orientation) to elicit the highest response possible. As has been proposed for cortical regions responsible for language comprehension and production, the differences observed could reflect differences in the functional connectivity of left and right visual cortex (Hustler and Galuske 2003).

Implications in psychophysical studies of TMS

It has been known for more than a decade that single pulses of TMS applied to visual cortex may disrupt processing on a particular task (e.g., letter identification, Corthout et al. 1999; motion discrimination, Hotson and Anand 1999). It has also been shown that the time course of such impairments is quite complex (Laycock et al. 2007) and depends on the site of stimulation. Our results speak to these psychophysical investigations of visual processing in several ways. First, it should be noted that these psychophysical studies do not explicitly rely on the virtual lesion hypothesis. Rather than declaring a specific brain region task relevant, we can interpret their findings to declare that brain networks engaged by stimulating the specific brain region are task relevant. While conducting a psychophysical investigation of visual processing, researchers have declared a stimulation region to be task relevant when the behavioral effect is present when stimulating one region over a nonrelevant control region. Our results suggest that the use of vertex stimulation results in an entirely distinct evoked potential that does not engage occipital or parietal cortex for ≥200 ms following stimulation. However, given our results and the high probability of cross-talk between brain regions, it seems unreasonable to perfectly find two nonoverlapping networks in time and space. This suggests that contrasts between stimulation sites that are functionally connected (e.g., V1/VT or V1/MT) are complex to interpret because they engage both common and distinct cortical networks.

Psychophysical research using single-pulse TMS over visual areas of the brain often uncovers multiple windows in time for which TMS impacts performance, both before (>100 ms) stimulus onset and well after (>100 ms) stimulus onset (e.g., Hotson and Anand 1999). In an attempt to understand these temporal dependencies, dynamic theories of visual processing argue that awareness is strongly influenced by the feedback component of visual analysis (e.g., Lamme and Roelfsema 2000). Pascual-Leone and Walsh (2001) hypothesized that the feedforward and feedback processes of visual analysis could be explored by phosphenes induction (bright flashes of light induced by TMS over visual cortex). Their results suggest that backprojections from MT to V1 are necessary for awareness of moving phosphenes, and they speculate that reentry of information (Edelman 1989) may be a general principle of visual awareness (Pascual-Leone and Walsh 2001). The correlations shown here, however, would suggest that time points of 40, 200, and 385 ms would have maximal differentiation between stimulation sites and time points corresponding to 116 and 292 ms would have little difference across stimulation sites. Based on the amplitude of the TMS-EP, we would predict the maximal behavioral impact to be very shortly after the pulse, perhaps at 40 ms. On the basis of these findings, we suggest that it may be possible to use both time and space in generation of a psychophysical control for TMS studies. An ideal study to test the reliance of a brain area on a task would have at least two “visual” brain regions (and a vertex control) and two time points of test. With our results, we would predict two sites would produce the common psychophysical effects at 116 ms and may be compared with the results at 40 ms, producing the effect size at the region of task interest that is not a general impact of TMS (116 ms) but still different across sites (40 ms). Future research will assess whether the oscillations found here are behaviorally relevant and align with the multiple time windows of processing these psychophysical studies often report.

Generality of Our Results to Other Stimulation Protocols

Our results have been based on single pulses of TMS from a monophasic coil. It is yet unknown what effects a rTMS or a biphasic coil will have on the TMS-induced oscillations reported here. rTMS sends single trains of TMS pulses very quickly at a region of the cortex. It seems reasonable to speculate, however, that the frequency of the pulse will interact with the global networks reported here in very specific ways and show a significant tuning to rate of stimulation with rTMS. With similar reasoning, rTMS has been paired with a 10-Hz flickering stimulus that entrains neurons and results in a brain response at the flicker frequency (Johnson et al. 2010). These researchers show that rTMS biases task-related activity, interacting with the networks created by neuronal entrainment.

Further, the monophasic coil induces a current in only one direction, while a biphasic coil induces a sinusoidal current. The difference between biphasic and monophasic TMS is becoming increasingly studied (Corthout et al. 2001; Kammer et al. 2001; Niehaus et al. 2000; Arai et al. 2007) but is much
more prominent in the rTMS literature. It seems reasonable to assume that a biphasic coil will have effects on the results shown here. It has been shown behaviorally that a monophasic coil has a larger effect on the motor evoked potential than a biphasic coil (Araì et al. 2005). Given the variable responses due to intensity of stimulation as well as region stimulated, it seems likely that a biphasic TMS pulse will also create unknown interactions with the reported networks. Future research is needed to determine the relationship between the local/global oscillations reported here and coil type.

Mechanisms of TMS

A recent surge in publications has attempted to uncover the inter-regional effects of single pulse TMS (Ruff et al. 2006). Most studies that have used concurrent neuroimaging devices have shown a nonlocalized effect of TMS, and our results provide more evidence for this nonlocal interpretation of the mechanism of TMS. We find that TMS engages multiple networks of brain regions in at least two different frequency bands, most likely reflecting the underlying connectivity of that brain region to multiple brain networks. Contrary to common assertions in the application of TMS, the effects of TMS are not very local at all.

Most recently, the virtual lesion hypothesis has been challenged by several researchers who believe TMS is not simply injecting noise into the brain system impacted by the magnetic field but instead traces the resonant frequency of the brain region stimulated (Rosanova et al. 2009). Through inspection of the frequencies >10 Hz within our data, we see consistency with those conclusions where we see that posterior occipital stimulation results in lower frequency activity (∼11 Hz) than vertex stimulation (∼13 Hz); however, since that research did not provide quantitative analysis of the oscillation <10 Hz, we cannot directly speak to the consistency with the relatively low-frequency responses we report here. We also note that their study investigated brain regions that belonged to very different functional networks (occipital, parietal, and frontal) while the purpose of our study was to contrast oscillatory activity elicited by functionally distinct areas of the occipital cortex within the visual system.

Lastly, it is worth noting that the more stimulation site-specific oscillations we measured share some characteristics of a visual-evoked response. Rather than single-pulse TMS creating a virtual lesion, we suggest that TMS is injecting another “stimulus” (consistent with the network engaged) into the brain at specific points in time; this stimulus engages other brain regions to form functional networks.

Conclusions

TMS-induced oscillations trace the multiple functional networks associated with the stimulation site. Robust effects of TMS include global resonances, elicited by any of the stimulation sites we investigated. Together, these findings are in agreement with growing evidence that the “virtual lesion” hypothesis should be revised or abandoned. By targeting a specific brain network, one may use simultaneous neuroimaging or EEG to uncover the functional network of that brain region and the network for which it belongs, researchers may use TMS to track modifications in this network as a function of different cognitive constraints (e.g., attention, visual discrimination) or as a function of disease or aging.

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DISCLOSURES

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REFERENCES


