Behavioral/Systems/Cognitive

Occipital Transcranial Magnetic Stimulation Has Opposing Effects on Visual and Auditory Stimulus Detection: Implications for Multisensory Interactions

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Multisensory interactions occur early in time and in low-level cortical areas, including primary cortices. To test current models of early auditory–visual (AV) convergence in unisensory visual brain areas, we studied the effect of transcranial magnetic stimulation (TMS) of visual cortex on behavioral responses to unisensory (auditory or visual) or multisensory (simultaneous auditory–visual) stimulus presentation. Single-pulse TMS was applied over the occipital pole at short delays (30–150 ms) after external stimulus onset. Relative to TMS over a control site, reactions times (RTs) to unisensory visual stimuli were prolonged by TMS at 60–75 ms poststimulus onset (visual suppression effect), confirming stimulation of functional visual cortex. Conversely, RTs to unisensory auditory stimuli were significantly shortened when visual cortex was stimulated by TMS at the same delays (beneficial interaction effect of auditory stimulation and occipital TMS). No TMS-effect on RTs was observed for AV stimulation. The beneficial interaction effect of combined unisensory auditory and TMS-induced visual cortex stimulation matched and was correlated with the RT-facilitation after external multisensory AV stimulation without TMS, suggestive of multisensory interactions between the stimulus-evoked auditory and TMS-induced visual cortex activities. A follow-up experiment showed that auditory input enhances excitability within visual cortex itself (using phosphene-induction via TMS as a measure) over a similarly early time-window (75–120 ms). The collective data support a mechanism of early auditory–visual interactions that is mediated by auditory-driven sensitivity changes in visual neurons that coincide in time with the initial volleys of visual input.

Key words: **multisensory; auditory; visual; primary visual cortex; transcranial magnetic stimulation; TMS; crossmodal**

Introduction

Detecting and integrating information across the senses is an advantageous mechanism for reducing perceptual ambiguity (MacLeod and Summerfield, 1990) and enhancing stimulus detection/localization (Stein and Meredith, 1993). The neural mechanisms subtending multisensory integration have traditionally been ascribed to higher-order brain areas and to occur late in sensory processing (Jones and Powell, 1970). Recent findings challenge this view, showing multisensory effects within low-level cortices and at early latencies (Ghazanfar and Schroeder, 2006). Studies in nonhuman primates show multisensory effects within primary and adjacent auditory cortices (Ghazanfar et al., 2005; Kayser et al., 2005, 2007; Schroeder and Foxe 2005; Lakatos et al., 2007). In humans, supra-additive nonlinear responses to multisensory stimuli occur within 100ms poststimulus onset (Giard and Peronnet, 1999; Foxe et al., 2000) and within primary (Martuzzi et al., 2007) or near-primary cortices (Foxe et al., 2002; Murray et al., 2005). These and similar studies indicate that neural processing in low-level cortices can be modulated by interactions between the senses at specific, early poststimulus time periods.

When characterizing the neural mechanisms mediating these early multisensory effects, it is important to consider their timing and origin. For example, it is important to consider whether multisensory interactions coincide with the initial feedforward inputs in an area or with later feedback activity. The early onset of cortical responses in humans to auditory (15–20 ms peak latency) (Liegeois-Chauvel et al., 1994) relative to visual stimulation (60–90 ms peak latency) (Martinez et al., 1999) makes it conceivable that auditory processes can impact feedforward visual processes within nominally visual cortices (Schroeder et al., 2004). This notion receives anatomical support from the demonstration in monkeys of direct, monosynaptic projections from primary and adjacent auditory cortices to early visual cortex (V1/V2) (Falchier et al., 2002; Rockland and Ojima, 2003). In additional support, there is evidence of auditory-driven responses at 48 ms within inferior parietal cortex of humans (Schroeder et al., 2004)

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that represent a potential basis for modulation of early sensory processing within visual cortex. Given this framework, auditory inputs are particularly well poised to impact visual processing not only at the lowest cortical levels but potentially at poststimulus latencies as short as 20 ms.

The present study investigated mechanisms of auditory–visual integration within the visual cortex through the application of single-pulse transcranial magnetic stimulation (TMS) over the occipital pole and at variable poststimulus delays (30–150 ms) while participants performed a simple detection task involving unisensory (auditory or visual) or multisensory (auditory–visual) external stimuli. In this way, we evaluated both the qualitative consequence of TMS and its temporal dynamics separately for each stimulus condition, revealing critical intervals for auditory integration within visual areas. We show that occipital TMS has opposing effects on visual and auditory stimulus detection over identical time periods. In a follow-up experiment, we further demonstrate that external auditory stimuli enhance visual cortex excitability as measured by phosphene induction via TMS (Silvanto et al., 2006; Bestmann et al., 2007; Ramos-Estebanez et al., 2007).

Materials and Methods

Participants. Fifteen neurologically normal volunteers participated in the study (four woman; mean age, 29.4 years; range, 25–40 years). All subjects had normal hearing and normal or corrected-to-normal vision. All participants gave written informed consent to the study approved by the Ethical Committee of the Geneva University Hospital. Seven of these participants also took part in the follow-up experiment.

TMS apparatus. A 70 mm figure-eight coil (maximum field strength, 2.2 T) and a Magstim Rapid^2 Transcranial Magnetic Stimulator were used (Magstim Company, Spring Gardens, UK). In both experiments of this study, the coil was placed over the occipital pole centered 3 cm above the inion with the handle pointing upward. The midline position and orientation of the coil ensured that we were stimulating the central representation and both hemispheres equally. External stimulus presentation, TMS pulse delivery, and behavioral response collection were controlled by E-prime (E-Prime 1.1; Psychology Software Tools, Pittsburgh, PA).

Main experiment: stimuli and task. Task-relevant sensory events consisted of auditory (A), visual (V), or simultaneous auditory–visual (AV) multisensory stimuli. The visual stimulus was a white disc presented on a gray background subtending 1.2° in diameter and was positioned at 3° vertically below the central fixation cross on a CRT computer monitor (75 cm viewing distance). The auditory stimulus was a 900 Hz pure tone [75 dB sound pressure level (SPL) at the ear; 44,100 kHz sampling rate] presented through two loudspeakers located on each side of the computer monitor and in horizontal alignment with the visual stimulus. Being delivered through a pair of speakers, the tone was perceived at the midline and thus colocalized with the visual stimulus. We opted to use a pure tone stimulus because previous research has shown that core (i.e., primary) auditory cortices respond best to pure tones, whereas belt (i.e., nonprimary) cortices respond best to complex tones and white noise bursts (Rauschecker et al., 1997).

Stimuli were presented for 14 ms duration either alone (A or V) or simultaneously (AV) as multisensory pairs with equal probability across these three stimulus conditions. The intertrial interval was varied pseudorandomly from 3000 to 5000 ms in steps of 500 ms to avoid anticipation of the stimulus onsets. Participants were instructed to make a button press with their right index finger as quickly as possible after stimulus detection, while maintaining central fixation. In this way, attention was continuously allocated to both sensory modalities. Subjects were seated in a comfortable chair in a dimly lit room.

External stimuli were paired with the delivery of a single TMS pulse centered over the occipital pole or to a control site (vertex in accordance with the standard electrode position Cz) at nine different time-delays relative to stimulus onset (30–150 ms, in steps of 15 ms). TMS was applied at a fixed intensity (70% of maximum stimulator output). Trials including TMS were randomly intermixed with trials excluding TMS to have a baseline measure of performance. Reaction times in the absence of TMS pulses (baseline trials) did not differ as a function of experimental blocks that varied whether TMS was applied to the occipital pole (target site) or vertex (control site) (V, $t_{(14)} = -0.99$, $p = 0.34$; A, $t_{(14)} = -0.65$, $p = 0.53$; AV, $t_{(14)} = -0.04$, $p = 0.97$). There was a total of 450 trials per stimulation site (occipital vs vertex) resulting in 15 repetitions per stimulus condition (three external stimulus conditions by 10 TMS delays, including the no-TMS baseline). Each subject completed 18 blocks of trials (nine per TMS stimulation site), with each block lasting ~3 min. Blocks involving occipital and vertex TMS stimulation were alternated within subjects, and block order was counterbalanced across subjects. We chose to alternate occipital and vertex TMS blocks, rather than presenting occipital and vertex TMS in succession, to minimize the risk of within-subject differences in baseline performance between test and control stimulation because of fatigue. Breaks were encouraged between blocks to maintain high concentration and minimize fatigue.

Postexperimental session: phosphene determination and perceptual training. After the above experimental session, all participants were asked whether they had perceived visual sensations other than the presented white disc during the experiment. TMS over the occipital pole can induce illusory visual percepts (called phosphenes) if applied at sufficiently high intensities (Meyer et al. 1991; Cowey and Walsh, 2000), but often requires several perceptual training sessions (Kammer et al., 2005b; Ramos-Estebanez et al., 2007). None of participants reported having seen phosphenes or any other illusory visual sensations, except one.

Subsequently, all participants underwent a training session to evaluate whether TMS could induce phosphenes at the experimental stimulation site and for quantification of individual phosphene threshold (PT), if applicable. Four of the 15 subjects perceived phosphenes during this training session in which they had their eyes closed and if stimulated below the intensity used in the experimental session (PTs, mean, 59.8%; range, 58–62% of maximum stimulator output). Six subjects were only able to perceive a phosphene with their eyes closed when stimulated at higher intensities (PTs, mean, 76%; range, 71–90%). The remaining five participants did not see any phosphenes or other illusory visual sensations throughout the training session at any TMS intensity.

Follow-up experiment. To further address the issue of multisensory interactions in early visual areas, we tested for auditory-induced modulation of visual cortex excitability by using phosphene perception as a probe measure of cortical excitability, similar to previous studies (Silvanto et al., 2006; Bestmann et al., 2007; Ramos-Estebanez et al., 2007). We retested those subjects who perceived phosphenes and were available to take part in additional sessions ($n = 6$) and one participant who initially did not perceive phosphenes but agreed to undergo additional training after which he also reliably perceived a phosphene (total $n = 7$).

Subjects were blindfolded and TMS was applied over the occipital pole at constant subthreshold TMS intensity (85% of individually defined PTs). We quantified the frequency of phosphene perception as a function of TMS delay with respect to auditory stimulus onset. We also applied TMS alone to establish a baseline frequency at which phosphenes were perceived. These trials were randomly intermixed with the auditory stimulus-TMS pairings. Auditory stimulation was identical to the previous experimental session, as were the TMS site, the delays between auditory stimulus onset and TMS (i.e., 30–150 ms, in steps of 15 ms) and the intertrial intervals. Fifteen trials were assessed per subject and condition. Participants were instructed to make a button-press with their right index finger whenever phosphenes were perceived and with the right middle finger whenever no phosphenes were perceived. All participants ($n = 7$) reliably perceived phosphenes with TMS above phosphene threshold.

Control experiment. In the main and follow-up experiment, we presented auditory stimuli at an intensity level (75 dB) well below those inducing a startle response. However, to further rule out the possibility that changes in cortical excitability in response to auditory stimulation may be caused by a startle reflex (possibly associated with widespread cortical activation), we looked for acoustic startle responses to the 900 Hz pure tone (14 ms duration, 44,100 kHz sampling rate) at the intensity

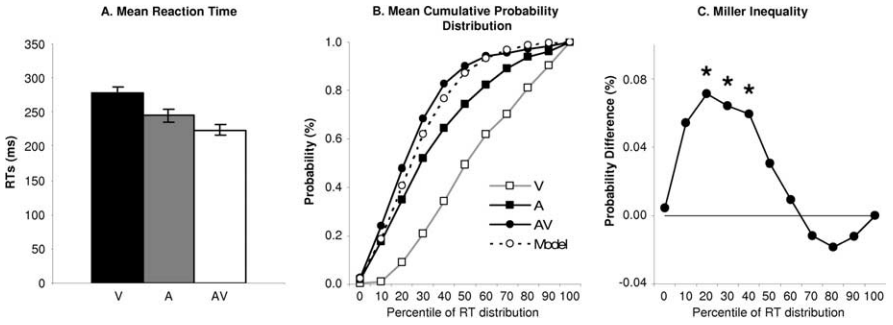

Figure 1. ***A***, Mean RTs (SEM indicated) to external stimuli at baseline (no TMS) as a function of stimulus-type. ***B***, Group averaged cumulative probability distribution for each stimulus condition and Miller's (1982) model predictions. ***C***, Reaction times to multisensory stimuli exceeded prediction of probability summation over the fastest part of the distribution (indicated by positive values). *$p < 0.05$.

used during the previous experiments (75 dB) as well as three other intensities (65, 85, and 95 dB SPL at the ear).

Six subjects (four having participated in the main and follow-up experiment and two new subjects) were asked to fixate a central cross on a gray CRT computer screen while the auditory stimulus was presented randomly at varying intensities through the two loudspeakers (30 trials per intensity). The intertrial interval varied randomly between 5 and 9 s. All other aspects of the experimental setting were identical to the main and follow-up experiments, including monitor and loudspeaker placements relative to the subject (75 cm distance). The startle response was measured by recording the eyeblink reflex (Blumenthal et al., 2005). For this purpose, we recorded electromyographic (EMG) activity of the orbicularis oculi muscle bipolarly from two electrodes beneath the right eye (van Boxtel et al., 1998; Blumenthal et al., 2005). Electrode impedance was kept below 10 kOhm. EMG activity was recorded at a sampling rate of 5000 Hz (Brainamp MR system; BrainProducts, München, Germany).

Data analyses. Trials with RTs faster than 100 ms and the 5% of trials with slowest RTs were not considered in the analysis to exclude outliers associated with stimulus anticipation and lack of attention, respectively. The fixed lower cutoff was chosen in accordance with previous studies using simple reaction time paradigms (Iacoboni and Zaidel, 1995; Jeeves and Moes, 1996; Marzi et al., 1998). As the cutoff for the upper outliers was more variable across previous studies, we used a percentile criterion for its definition (Clark-Carter, 1997). To verify that multisensory stimulus presentation facilitated reaction times relative to either unisensory condition, baseline data (i.e., in the absence of TMS and from all 18 blocks of trials) were submitted to a repeated-measure ANOVA using stimulus condition (A, V, AV) as the within-subject factor. Faster reaction times to the multisensory condition would be evidence of a redundant signals effect. Moreover, we performed an analysis using Miller's race-model inequality (Miller, 1982), which assesses whether reaction time facilitation exceeds predictions of probability summation and provides a psychophysical benchmark for invoking the occurrence of neural response interactions. Details of this analysis can be found in previously published studies by our group (Murray et al., 2005; Zampini et al., 2007). Briefly, Miller's inequality contrasts the probability of an RT of a given speed under multisensory conditions with the sum of the probabilities of an equally fast RT under unisensory conditions (minus the joint probability of this occurrence). Our group and others have consistently demonstrated such facilitation in excess of probability summation under highly similar experimental conditions (Molholm et al., 2002; Martuzzi et al., 2007), indicating that the redundant signals effect most likely results from neural response interactions.

To parcel out any unspecific effect of TMS that could contaminate response times (for instance because of the TMS-clicks), we calculated a relative RT measure by subtracting mean RTs between occipital and vertex stimulation (RT occipital minus RT vertex) for each condition and TMS delay (as well as the no-TMS trials), separately. These values were submitted to a repeated-measure ANOVA with the within-subject factors of stimulus condition (A, V, AV) and TMS-delay (noTMS, 30, 45, 60, 75, 90, 105, 120, 135, and 150 ms). Where appropriate, *post hoc* comparisons were performed, consisting of simple tests across conditions per TMS-delay (to explore the significant condition by TMS-delay interaction), as well as one-sample *t* tests against zero and paired *t* tests (to evaluate for a given TMS delay the effect of occipital TMS relative to control/vertex stimulation and relative to noTMS/baseline; within-condition comparisons).

For the follow-up experiment, the percentage of trials on which phosphenes were perceived were submitted to a repeated-measure ANOVA using TMS-delay as the within-subject factor (noTMS, 30, 45, 60, 75, 90, 105, 120, 135 and 150 ms). When appropriate, ANOVA results were completed by *t* tests.

Offline EMG signal processing in the control experiment consisted of bandpass filtering (20–500 Hz) (van Boxtel et al., 1998), rectifying of the signal (Blumenthal et al., 2005) and integration of the EMG activity from −100 to 0 ms before the auditory stimulus (to obtain baseline values) and from +20 to 120 ms after the stimulus (area under curve) where eyeblink activity was maximal. Data were then subjected to a repeated-measure ANOVA with the factor sound-intensity (baseline, 65, 75, 85, 95 dB), followed by *post hoc* paired *t* tests where appropriate.

Results

Mean RTs to A, V, and AV multisensory stimuli in the absence of any TMS pulse (i.e., baseline, no TMS) are shown in Figure 1*a*. A repeated measures ANOVA revealed a significant main effect of stimulus condition ($F_{(2,28)} = 38.8$; $p < 0.00001$). Follow-up contrasts confirmed that this effect was attributable to the facilitation of RTs in the multisensory (223.8 ± 7.2 ms, mean ± SEM) versus either visual (278.1 ± 8.2 ms; $t_{(14)} = 8.4$; $p < 0.00001$) or auditory (245.0 ± 9.4; $t_{(14)} = 5.4$; $p < 0.0001$) conditions. Reaction times to auditory and visual unisensory stimuli also differed ($t_{(14)} = 4.5$; $p < 0.001$). Additionally, there was violation of the race model inequality over the fastest 40% of the RT distribution (Fig. 1*b*,*c*). This violation is indicative of a redundant signals effect exceeding predictions based on simple probability summation and suggests that neural response interactions are occurring before motor response initiation. These findings thus replicate previously observed effects (Raab, 1962; Miller, 1982; Schröger and Widmann, 1998; Molholm et al., 2002; Martuzzi et al., 2007). For each participant, we then calculated the RT difference between the auditory unisensory condition (which was on average the faster unisensory condition) and the multisensory condition, to have a metric of the RT benefit from multisensory stimulation in the absence of TMS. On average, the multisensory condition produced an RT facilitation of 21.3 ± 4.1 ms, which is approximately equivalent to that obtained by Martuzzi et al. (2007).

RT differences between occipital and control (vertex) TMS stimulation for each of the nine TMS pulse delays and the three external stimulus conditions (A, V, AV) are shown in Figure 2. In addition, baseline values are shown from trials on which no TMS was applied. Visual inspection of these plots suggests that occipital TMS had differential effects on RTs as a function of the sensory modality stimulated and TMS-pulse delay. Specifically, RTs to visual stimuli were impaired at within 60–75 ms TMS-pulse delay (Fig. 2, black line), in keeping with previous studies demonstrating a visual "suppression" effect (Amassian et al., 1989; Beckers and Hömberg, 1991; Masur et al. 1993; Corthout et al., 1999; Kastner et al., 1998, Kammer et al., 2005a), whereas those to auditory stimuli were facilitated at the same delays (Fig. 2, gray line). No noticeable effect on multisensory stimulus processing

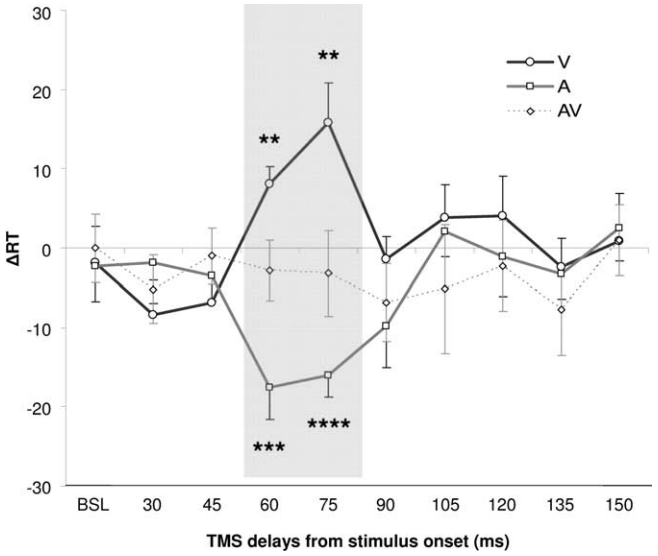

Figure 2. RTs (SEM indicated) to external stimuli are represented at baseline (BSL, no TMS) and for each TMS delay from stimulus onset (30–150 ms) as a function of stimulus type (A, V, AV). The curves represent changes in RTs caused by occipital TMS (visual cortex stimulation) relative to control (vertex) stimulation ($\Delta RT = RT_{occipital\ TMS}$ minus $RT_{vertex\ TMS}$). An RT benefit for external auditory stimuli (A) when combined with stimulation of visual cortex at TMS-delays of 60–75 ms was observed (gray curve, negative deflection). This coincided in time with a RT cost to external visual stimuli attributable to TMS at the same site (visual suppression, black curve, positive values). The shaded area represents the time window of coincident effects. $^{**}p < 0.01$; $^{***}p < 0.001$; $^{****}p < 0.0001$.

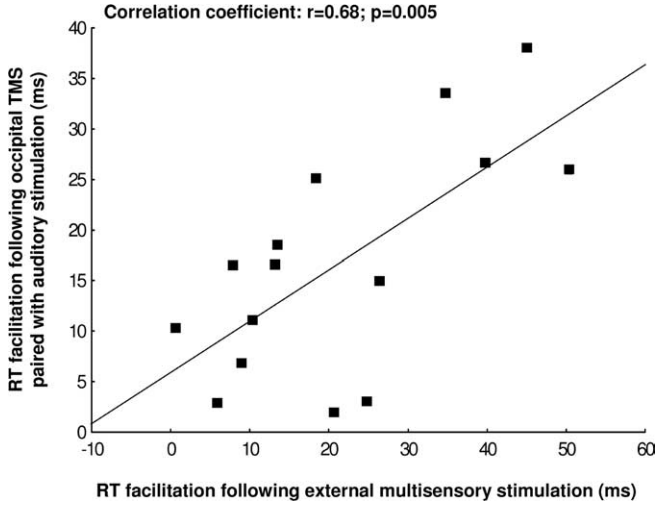

Figure 3. Scatterplot of the correlation between RT facilitations in this study. The *x*-axis plots the RT facilitation after multisensory stimuli in the absence of TMS (AV minus A). The *y*-axis plots the RT facilitation after occipital TMS in combination with auditory stimulation (A-occipital TMS minus A-vertex TMS).

was evident (Fig. 2, dotted line). To statistically assess these observations, these values were submitted to a repeated measures ANOVA, using TMS delay (including baseline) and stimulus condition as within-subjects factors. There was a significant interaction between these factors ($F_{(18,252)} = 2.6$; $p = 0.0006$). Separate ANOVAs were then conducted for each delay to further explore this interaction. Main effects of stimulus condition were only observed at delays of 60 ms ($F_{(2,28)} = 14.3$; $p = 0.00005$) and 75 ms ($F_{(2,28)} = 11.8$; $p = 0.0002$), suggesting differential effects of TMS at these delays. Additional *post hoc* tests at these delays showed that the RT benefit and cost induced by occipital TMS in the A and V conditions, respectively, are significant (relative to control/vertex TMS; one-sample *t* tests against zero) at 60 ms (A, $t_{(14)} = -4.5$, $p = 0.0005$; V, $t_{(14)} = 3.9$, $p = 0.002$) and 75 ms (A, $t_{(14)} = -6.2$, $p = 0.00002$; V, $t_{(14)} = 3.3$, $p = 0.006$). In addition, RT benefits and costs were also significantly different from their respective baseline values (paired *t* tests against noTMS) both for the 60 ms (A, $t_{(14)} = -3.0$, $p = 0.009$; V, $t_{(14)} = 2.6$, $p = 0.02$) and 75 ms delay (A, $t_{(14)} = -3.1$, $p = 0.008$; V, $t_{(14)} = -3.6$, $p = 0.003$). No such effects were observed for the AV-condition (all $p > 0.23$). These within-condition comparisons thus substantiate that occipital TMS facilitated RT to auditory stimuli and slowed RT to visual stimuli at specific TMS delays. Critically, these RT benefits to auditory stimuli caused by occipital TMS were observed after normalization to RTs to auditory stimuli under control TMS (Δ A-occipital minus A-vertex), and can thus be interpreted to result from auditory–"visual" interactions because of stimulus-evoked auditory and TMS-induced visual cortex stimulation. This overall pattern of results indicates that there is a period (Fig. 2, gray box) during which the behavioral consequences of occipital TMS depend on the sensory modality of external inputs.

An additional analysis tested whether the magnitude of the redundant signals effect obtained for multisensory stimulation in the absence of TMS (i.e., -21.3 ± 4.1 ms) differed from the RT facilitation observed for the combination of auditory stimuli and occipital TMS (mean RT facilitation across the 60ms and 75ms TMS delays used for analysis). No difference was observed (-16.8 ± 2.9 ms; $t_{(14)} = -1.5$; $p = 0.14$). The RT facilitation for detecting auditory stimuli that was induced by occipital TMS thus matched the RT facilitation obtained with pure external AV stimulation. We then tested whether the magnitude of these facilitation effects correlated across subjects. There was a significant correlation between the extent of RT facilitation after multisensory stimulation and the extent of RT facilitation following from the combination of occipital TMS and auditory stimuli (Pearson correlation: $r_{(13)} = 0.68$; $p = 0.005$; Spearman rank order correlation: $r_{(13)} = 0.62$, $p = 0.01$) (Figure 3). No such correlation was observed between the extent of RT facilitation after multisensory stimulation and the extent of RT impairment following from the combination of occipital TMS and visual stimuli.

The above pattern of effects did not change when the four subjects with strongest visual cortex excitability who showed the lowest phosphene threshold (below the TMS output intensity) were excluded from the analysis (interaction term $F_{(18,180)} = 1.7$; $p < 0.05$). The ANOVAs conducted separately for each delay remained significant for both the 60 ms (main effect of stimulus condition, $F_{(2,20)} = 8.9$; $p = 0.002$), and 75 ms delays ($F_{(2,20)} = 10.2$; $p = 0.0009$). The *post hoc* t tests showed a significant auditory RT benefit and visual RT cost at 60 ms (A, $t_{(10)} = -4.0$, $p = 0.003$; V, $t_{(10)} = 2.6$, $p = 0.02$) and 75ms-delay (A, $t_{(10)} = -4.5$, $p = 0.001$; V, $t_{(10)} = 3.9$, $p = 0.003$). Finally, the correlation between the extent of RT facilitation after multisensory stimulation and the extent of RT facilitation following from the combination of occipital TMS and auditory stimuli also remained significant (Pearson correlation: $r_{(9)} = 0.75$, $p = 0.007$; Spearman rank order correlation: $r_{(9)} = 0.65$, $p = 0.03$). This rules out the possibility that our findings were solely driven by these four subjects.

The results of the follow-up experiment that examined changes in visual cortex excitability combined with auditory stimulation (probed through phosphene induction by occipital

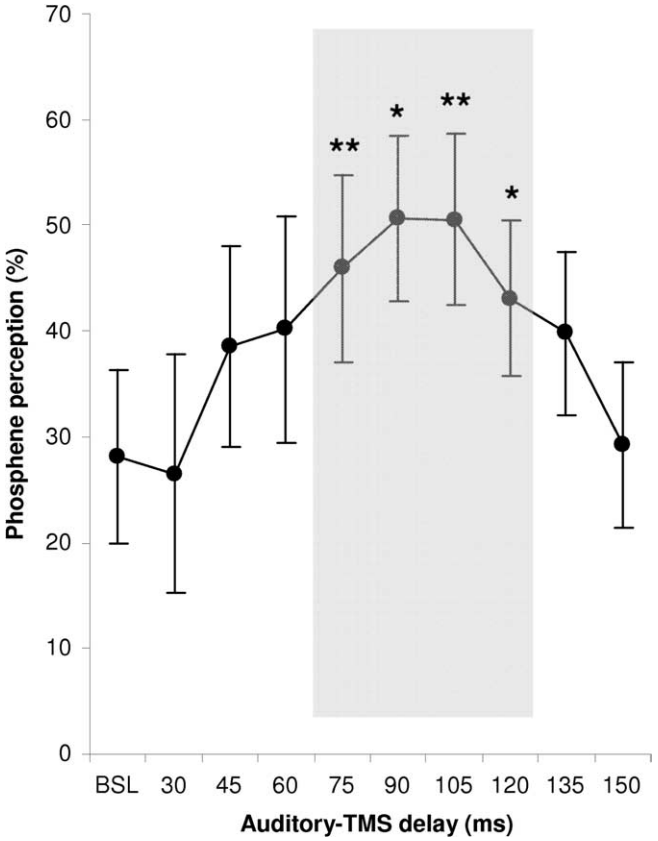

Figure 4. Percentage of perceived phosphenes caused by occipital TMS as a function of TMS-pulse delay from the brief auditory stimulus. TMS was applied below phosphene threshold (85% PT), latter defined at baseline (BSL; i.e., in the absence of any auditory stimulus). The shaded area represents the window of significantly increased visual cortex excitability by auditory input (i.e., phosphene perception > BSL; $*p < 0.05$; $**p < 0.01$).

TMS) are illustrated in Figure 4. Percentage of induced phosphenes significantly differed as a function of delay from auditory stimulus onset (one-way ANOVA; main effect, $F_{(9,54)} = 3.6$; $p = 0.001$) with maximal values (highest increase in visual cortex excitability) occurring at a delay of 90 ms postauditory stimulus onset. In comparison to baseline (TMS without auditory stimulus), phosphene perception (and, hence, visual cortex excitability) was significantly enhanced by auditory stimulation 75–120 ms before TMS delivery (all t values >3.045; $p < 0.023$), fitting the critical window for TMS-induced auditory–"visual" interactions identified above (Fig. 2).

Finally, the control experiment on reflexive eyeblink responses as a function of four sound intensity levels (65, 75, 85, and 95 dB) revealed that the auditory stimulus intensity used in the main and follow-up experiment (75 dB) was well below the intensity level inducing an acoustic startle response. EMG activity of the orbicularis oculi muscle at baseline (i.e., in the 100 ms before auditory stimulus onset) was on average 319.9 ± 36.2 μV · s. The EMG activity in response to the auditory stimuli was comparable in amplitude to this baseline value across all sound-levels except the loudest one (65 dB, 326.1 ± 40.5 μV · s; 75 dB, 308.2 ± 45.9 μV · s; 85 dB, 353.8 ± 37.7 μV · s; 95 dB, 495.3 ± 74.9 μV · s). The overall ANOVA revealed a main effect of sound intensity ($F_{(4, 20)} = 8.9$; $p = 0.0003$), which was explained by a reflexive startle (eyeblink) response to the 95 dB sound (paired t tests against baseline, $t = 3.7$, $p = 0.015$) that was absent for the other sound-levels (all $p > 0.57$). The 95 dB sound is well above the test level used (75 dB) for which no significant startle response was found.

Discussion

The present study examined the mechanisms of auditory–visual convergence and interactions in visual cortex using single-pulse TMS. Occipital TMS had opposing effects on behavioral responses to visual and auditory stimuli over identical poststimulus time windows. Stimulation of visual cortex by TMS was confirmed by the fact that RTs to visual stimuli were slowed when TMS was applied 60–75 ms later (Amassian et al., 1989). In contrast, RTs to auditory stimuli were facilitated when an identical TMS pulse was applied at the same temporal delays. The magnitude of this facilitation equaled and correlated with the magnitude of the redundant signals effect observed after multisensory stimulation without TMS. We propose that multisensory interactions between the external auditory stimulus and the central (TMS-induced) visual cortex stimulation underlie this facilitation, despite the absence of any external visual stimulus and any centrally induced visual percept (i.e., phosphenes). The timing of this facilitation revealed a short period of increased sensitivity for integration of multisensory inputs. To test the hypothesis that the auditory stimulus enhanced visual cortex excitability, we conducted a follow-up experiment wherein participants more often reported perceiving phosphenes, thought to originate from early visual cortex (V1/V2) (Cowey and Walsh, 2000; Kammer et al., 2005b), when TMS was preceded 75–120 ms beforehand by auditory stimuli. These collective results support the existence of an early critical period for producing facilitative multisensory interactions and further suggest that auditory and visual inputs are converging within the occipital cortex while nonetheless generating divergent profiles of activity.

This is the first study, to our knowledge, of opposing behavioral effects of occipital TMS on responses to visual versus nonvisual sensory inputs. The time window (i.e., 60–75 ms) of these effects is in line with previous studies revealing visual suppression by occipital TMS (Amassian et al., 1989), and is traditionally believed to correspond to the peak of the first volley of feedforward input in visual cortex (Martinez et al., 1999). Although transient disruption of brain function predominates in TMS research, there are also reports of functional improvements (Seyal et al., 1995; Walsh et al., 1998; Hilgetag et al., 2001; Grosbras and Paus, 2002, 2003; Kobayashi et al., 2004; Thut et al., 2005), similar to the RT facilitation we observed. Several studies further showed that stimulation of a given area can induce opposing (disruptive or facilitative) effects depending on task or behavioral context (Walsh et al., 1998; Grosbras and Paus, 2002, 2003). The mechanisms invoked to explain beneficial TMS-effects are a disinhibition of unstimulated brain areas whose function are normally suppressed by the TMS target site (Seyal et al., 1995; Walsh et al., 1998) or a neural priming effect caused by TMS before a critical period (Grosbras and Paus, 2002, 2003). Also, it has been shown that qualitatively different TMS effects can result from modulation of the neural activation state via previous experience (Silvanto et al., 2007). Could these mechanisms explain our data? We can discount disinhibition of auditory areas by occipital TMS because the facilitative effect on auditory stimulus detection should have been strongest when TMS was applied at early delays closer in time to the latency of initial cortical auditory processing ($\ll$60ms). This was clearly not the case with our results. We would also argue against a priming effect of occipital TMS on auditory detection, because

the TMS pulse was applied after auditory stimulus onset. Our data indicate that rather it was the external stimuli (visual vs auditory) that differentially set (primed) occipital cortex, so that a subsequent TMS pulse inducing a visual "signal" had a differential behavioral outcome depending on the external (visual or auditory) input context, if applied within a narrow time window between 60–75 ms poststimulus onset. Although "visual" induction by TMS resulted in interference if paired with an external visual input, it resulted in integration and thus facilitation if paired with an external auditory input, probably through multisensory interactions. We can only speculate as to the neurophysiological mechanisms underlying these opposing effects. There is anatomical evidence that the laminar distributions of direct auditory and visual inputs into the occipital pole differ. Whereas the feedforward thalamocortical visual inputs terminate predominantly within layer 4 (cf. Felleman and Van Essen, 1991), the direct monosynaptic inputs from primary and near-primary auditory cortices into the occipital pole terminate predominantly within superficial layers and are consistent with a functionally modulatory and lateral profile (Rockland and Ojima, 2003). It is therefore conceivable that the external visual and auditory stimuli evoked different laminar response profiles in visual cortex that may have differentially conditioned the fate of the subsequent TMS-induced "visual" signal (i.e., the way this signal integrates or not into the externally evoked, physiological meaningful pattern of activity). In line with the notion of different response profiles within V1 and opposing effects to auditory and visual stimuli, it has been shown previously that responsiveness of V1/V2 is enhanced to an auditory stimulus when this is presented ~60–70 ms after a multisensory auditory–visual stimulus (Watkins et al., 2006) and attenuated to a visual stimulus preceded by the same multisensory auditory–visual event (Meylan and Murray, 2007).

The present results do not allow us to draw conclusive inferences regarding the origin of auditory inputs to the occipital lobe thought to subserve the present effects. However, it is intriguing that occipital TMS should have such a time-locked impact on auditory RTs. That is, if the auditory input into the occipital lobe was already a highly processed signal, one might expect minimal impact of occipital TMS, presumably because multiple and parallel auditory-driven circuits would already be active and would be initiating RTs independently of events within the occipital pole. However, the robust effects of occipital TMS on auditory RTs and moreover, their correlation with the RT facilitation ordinarily observed under multisensory conditions, would suggest that events within the occipital pole are indeed playing an integral role in generating behaviorally relevant auditory–visual interactions. We therefore favor a model of interactions between relatively unprocessed sensory signals at low-levels of the cortical hierarchy. Direct anatomical connections between primary auditory and visual cortices may be the basis for the observed effects. Several laboratories have now independently identified monosynaptic projections between these cortices in nonhuman primates (Falchier et al., 2002; Rockland and Ojima, 2003; Cappe and Barone, 2005). Previous support for the functional significance of these pathways has been provided by the observation of short-latency (<80ms) auditory-evoked responses over occipital sites in patients with late-onset blindness that were linked to auditory-evoked visual sensations (Rao et al., 2007).

An alternative viewpoint, which we cannot unequivocally exclude, is that auditory inputs are rerouted via the parietal cortices en route to occipital cortices. Auditory-driven responses have been observed within parietal areas at ~50 ms poststimulus onset (Schroeder et al., 2004). This early response has been suggested to represent highly processed auditory information that is theoretically available at early visual processing stages (Schroeder et al., 2004). The parietal cortex would thus be a plausible candidate for early, auditory-driven feedback modulation of visual cortex sensitivity (Macaluso and Driver, 2005).

Finally, it could be argued that our effects might follow from an acoustic startle response to the suprathreshold auditory stimulus, which goes along with widespread cortical activation that is potentially mediated through the ascending reticular activating system (McDowell et al., 2006). Such an explanation seems unlikely. First, the auditory stimulus duration we used was short and its volume was below the typical threshold required to evoke startle responses (Blumenthal, 1996; Brown et al., 1991; Grillon and Baas, 2003), confirmed here through our control experiment. Second, although acoustic startle responses may go along with changes in cortical excitability, they would not be expected to be limited to specific latencies coinciding with visual input given that they would likely generalize to other cortical areas. Third, a previous TMS study of tactile influences on TMS-induced phosphene perception showed that the likelihood of perceiving a visual phosphene (induced by subthreshold TMS) is greatest when the tactile pulse precedes occipital TMS by 60ms (Ramos-Estebanez et al., 2007), matching the peak-latency of increased visual cortex excitability we observed. Notably, their use of subthreshold tactile stimulation, which did not reach perceptual awareness, excluded startle responses as a basis for increased cortical excitability (namely, enhanced phosphene perception).

Although occipital TMS facilitated RT to auditory stimuli and slowed RT to visual stimuli relative to control (vertex) TMS and the baseline (noTMS), there was no such effect on RTs to external multisensory AV stimulation. Intuitively, one might have expected a further enhancement of RTs in the AV condition, as three events co-occurred. However, the data (Fig. 2) support not even a small RT facilitation in the AV condition. Possible reasons are that opposing effects were mutually annihilated under this condition or, alternatively, that performance was already at ceiling levels.

In summary, we provide the first demonstration that multisensory auditory–visual interactions occurring during stages of initial sensory transmission within low-level visual cortices are behaviorally relevant, extending previous evidence for early nonlinear neuronal response interactions (Giard and Peronnet, 1999; Molholm et al., 2002) or multisensory effects within primary visual cortices (Martuzzi et al., 2007).

References

Amassian VE, Cracco RQ, Maccabee PJ, Cracco JB, Rudell A, Eberle L (1989) Suppression of visual perception by magnetic coil stimulation of human occipital cortex. Electroencephalogr Clin Neurophysiol 74:458–462.

Beckers G, Hömberg V (1991) Impairment of visual perception and visual short term memory scanning by transcranial magnetic stimulation of occipital cortex. Exp Brain Res 87:421–432.

Bestmann S, Ruff CC, Blakemore C, Driver J, Thilo KV (2007) Spatial attention changes excitability of human visual cortex to direct stimulation. Curr Biol 17:134–139.

Blumenthal TD (1996) Inhibition of the human startle response is affected by both prepulse intensity and eliciting stimulus intensity. Biol Psychol 44:85–104.

Blumenthal TD, Cuthbert BN, Filion DL, Hackley S, Lipp OV, van Boxtel A (2005) Committee report: guidelines for human startle eyeblink electromyographic studies. Psychophysiology 42:1–15.

Brosch M, Selezneva E, Scheich H (2005) Nonauditory events of a behavioral procedure activate auditory cortex of highly trained monkeys. J Neurosci 25:6797–6806.

Brown P, Rothwell JC, Thompson PD, Britton TC, Day BL, Marsden CD (1991) New observations on the normal auditory startle reflex in man. Brain 114:1891–1902.

Cappe C, Barone P (2005) Heteromodal connections supporting multisensory integration at low levels of cortical processing in the monkey. Eur J Neurosci 22:2886–2902.

Clark-Carter D (1997) Doing quantitative psychological research. From design to report. East Sussex, UK: Psychology.

Corthout E, Uttl B, Walsh V, Hallett M, Cowey A (1999) Timing of activity in early visual cortex as revealed by transcranial magnetic stimulation. Neuroreport 10:2631–2634.

Cowey A, Walsh V (2000) Magnetically induced phosphenes in sighted, blind and blindsighted observers. Neuroreport 11:3269–3273.

Falchier A, Clavagnier S, Barone P, Kennedy H (2002) Anatomical evidence of multimodal integration in primate striate cortex. J Neurosci 22:5749–5759.

Felleman DJ, Van Essen DC (1991) Distributed hierarchical processing in the primate cerebral cortex. Cereb Cortex 1:1–47.

Foxe JJ, Morocz IA, Murray MM, Higgins BA, Javitt DC, Schroeder CE (2000) Multisensory auditory-somatosensory interactions in early cortical processing revealed by high-density electrical mapping. Brain Res Cogn Brain Res 10:77–83.

Foxe JJ, Wylie GR, Martinez A, Schroeder CE, Javitt DC, Guilfoyle D, Ritter W, Murray MM (2002) Auditory-somatosensory multisensory processing in auditory association cortex: an fMRI study. J Neurophysiol 88:540–543.

Ghazanfar AA, Schroeder CE (2006) Is neocortex essentially multisensory? Trends Cogn Sci 10:278–285.

Ghazanfar AA, Maier JX, Hoffman KL, Logothetis NK (2005) Multisensory integration of dynamic faces and voices in rhesus monkey auditory cortex. J Neurosci 25:5004–5012.

Giard MH, Peronnet F (1999) Auditory–visual integration during multimodal object recognition in humans: a behavioral and electrophysiological study. J Cogn Neurosci 11:473–490.

Grillon C, Baas J (2003) A review of the modulation of the startle reflex by affective states and its application in psychiatry. Clin Neurophysiol 114:1557–1579.

Grosbras MH, Paus T (2002) Transcranial magnetic stimulation of the human frontal eye field: effects on visual perception and attention. J Cogn Neurosci 14:1109–1120.

Grosbras MH, Paus T (2003) Transcranial magnetic stimulation of the human frontal eye field facilitates visual awareness. Eur J Neurosci 18:3121–3126.

Hilgetag CC, Theoret H, Pascual-Leone A (2001) Enhanced visual spatial attention ipsilateral to rTMS-induced "virtual lesions" of human parietal cortex. Nat Neurosci 4:953–957.

Iacoboni M, Zaidel E (1995) Channels of the corpus callosum. Evidence from simple reaction times to lateralized flashes in the normal and the split brain. Brain 118:779–788.

Jeeves MA, Moes P (1996) Interhemispheric transfer time differences related to aging and gender. Neuropsychologia 34:627–636.

Jones EG, Powell TP (1970) An anatomical study of converging sensory pathways within the cerebral cortex of the monkey. Brain 93:793–820.

Kammer T, Puls K, Strasburger H, Hill NJ, Wichmann FA (2005a) Transcranial magnetic stimulation in the visual system. I. The psychophysics of visual suppression. Exp Brain Res 160:118–128.

Kammer T, Puls K, Erb M, Grodd W (2005b) Transcranial magnetic stimulation in the visual system. II. Characterization of induced phosphenes and scotomas. Exp Brain Res 160:129–140.

Kastner S, Demmer I, Ziemann U (1998) Transient visual field defects induced by transcranial magnetic stimulation over the occipital lobe. Exp Brain Res 118:19–26.

Kayser C, Petkov CI, Augath M, Logothetis NK (2005) Integration of touch and sound in auditory cortex. Neuron 48:373–384.

Kayser C, Petkov CI, Augath M, Logothetis NK (2007) Functional imaging reveals visual modulation of specific fields in auditory cortex. J Neurosci 27:1824–1835.

Kobayashi M, Hutchinson S, Theoret H, Schlaug G, Pascual-Leone A (2004) Repetitive TMS of the motor cortex improves ipsilateral sequential simple finger movements. Neurology 62:91–98.

Lakatos P, Chen CM, O'Connell MN, Mills A, Schroeder CE (2007) Neuronal oscillations and multisensory interaction in primary auditory cortex. Neuron 53:279–292.

Liegeois-Chauvel C, Musolino A, Badier JM, Marquis P, Chauvel P (1994) Evoked potentials recorded from the auditory cortex in man: evaluation and topography of the middle latency components. Electroencephalogr Clin Neurophysiol 92:204–214.

Macaluso E, Driver J (2005) Multisensory spatial interactions: a window onto functional integration in the human brain. Trends Neurosci 28:264–271.

MacLeod A, Summerfield Q (1990) A procedure for measuring auditory and audio-visual speech-reception thresholds for sentences in noise: rationale, evaluation, and recommendations for use. Br J Audiol 24:29–43.

Martinez A, Anllo-Vento L, Sereno MI, Frank LR, Buxton RB, Dubowitz DJ, Wong EC, Hinrichs H, Heinze HJ, Hillyard SA (1999) Involvement of striate and extrastriate visual cortical areas in spatial attention. Nat Neurosci 2:364–369.

Martuzzi R, Murray MM, Michel CM, Thiran JP, Maeder PP, Clarke S, Meuli RA (2007) Multisensory interactions within human primary cortices revealed by BOLD dynamics. Cereb Cortex 17:1672–1679.

Marzi CA, Miniussi C, Maravita A, Bertolasi L, Zanette G, Rothwell JC, Sanes JN (1998) Transcranial magnetic stimulation selectively impairs interhemispheric transfer of visuo-motor information in humans. Exp Brain Res 118:435–438.

Masur H, Papke K, Oberwittler C (1993) Suppression of visual perception by transcranial magnetic stimulation: experimental findings in healthy subjects and patients with optic neuritis. Electroencephalogr Clin Neurophysiol 86:259–267.

McDowell JE, Brown GG, Lazar N, Camchong J, Sharp R, Krebs-Thomson K, Eyler LT, Braff DL, Geyer MA (2006) The neural correlates of habituation of response to startling tactile stimuli presented in a functional magnetic resonance imaging environment. Psychiatry Res 148:1–10.

Meyer BU, Diehl R, Steinmetz H, Britton TC, Benecke R (1991) Magnetic stimuli applied over motor and visual cortex: influence of coil position and field polarity on motor responses, phosphenes, and eye movements. Electroencephalogr Clin Neurophysiol Suppl 43:121–134.

Meylan RV, Murray MM (2007) Auditory–visual multisensory interactions attenuate subsequent visual responses in humans. Neuroimage 35:244–254.

Miller J (1982) Divided attention: evidence for coactivation with redundant signals. Cogn Psychol 14:247–279.

Molholm S, Ritter W, Murray MM, Javitt DC, Schroeder CE, Foxe JJ (2002) Multisensory auditory–visual interactions during early sensory processing in humans: a high-density electrical mapping study. Brain Res Cogn Brain Res 14:115–128.

Murray MM, Molholm S, Michel CM, Heslenfeld DJ, Ritter W, Javitt DC, Schroeder CE, Foxe JJ (2005) Grabbing your ear: rapid auditory-somatosensory multisensory interactions in low-level sensory cortices are not constrained by stimulus alignment. Cereb Cortex 15:963–974.

Raab D (1962) Statistical facilitation of simple reaction times. Trans NY Acad Sci 24:574–590.

Rao A, Nobre AC, Alexander I, Cowey A (2007) Auditory evoked visual awareness following sudden ocular blindness: an EEG and TMS investigation. Exp Brain Res 176:288–298.

Ramos-Estebanez C, Merabet LB, Machii K, Fregni F, Thut G, Wagner TA, Romei V, Amedi A, Pascual-Leone A (2007) Visual phosphene perception modulated by sub-threshold cross-modal sensory stimulation. J Neurosci 27:4178–4181.

Rauschecker JP, Tian B, Pons T, Mishkin M (1997) Serial and parallel processing in rhesus monkey auditory cortex. J Comp Neurol 382:89–103.

Rockland KS, Ojima H (2003) Multisensory convergence in calcarine visual areas in macaque monkey. Int J Psychophysiol 50:19–26.

Schroeder CE, Foxe JJ (2005) Multisensory contributions to low-level, "unisensory" processing. Curr Opin Neurobiol 15:454–458.

Schroeder CE, Molhom S, Lakatos P, Ritter W, Foxe JJ (2004) Human-simian correspondence in the early cortical processing of multisensory cues. Cogn Process 5:140–151.

Schröger E, Widmann A (1998) Speeded responses to audiovisual signal changes result from bimodal integration. Psychophysiology 35:755–759.

Seyal M, Ro T, Rafal R (1995) Increased sensitivity to ipsilateral cutaneous stimuli following transcranial magnetic stimulation of the parietal lobe. Ann Neurol 38:264–267.

Silvanto J, Lavie N, Walsh V (2006) Stimulation of the human frontal eye fields modulates sensitivity of extrastriate visual cortex. J Neurophysiol 96:941–945.

Silvanto J, Muggleton NG, Cowey A, Walsh V (2007) Neural adaptation reveals state-dependent effects of transcranial magnetic stimulation. Eur J Neurosci 25:1874–1881.

Stein BE, Meredith MA (1993) The merging of the senses. Cambridge, MA: MIT.

Thut G, Nietzel A, Pascual-Leone A (2005) Dorsal posterior parietal rTMS affects voluntary orienting of visuospatial attention. Cereb Cortex 15:628–638.

van Boxtel A, Boelhouwer AJ, Bos AR (1998) Optimal EMG signal bandwidth and interelectrode distance for the recording of acoustic, electrocutaneous, and photic blink reflexes. Psychophysiology 35:690–697.

Walsh V, Ellison A, Battelli L, Cowey A (1998) Task-specific impairments and enhancements induced by magnetic stimulation of human visual area V5. Proc Biol Sci 265:537–543.

Watkins S, Shams L, Tanaka S, Haynes JD, Rees G (2006) Sound alters activity in human V1 in association with illusory visual perception. Neuroimage 31:1247–1256.

Zampini M, Torresan D, Spence C, Murray MM (2007) Auditory-somatosensory multisensory interactions in front and rear space. Neuropsychologia 45:1869–1877.