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# Causal Contributions of the SMA to Alertness and Consciousness Interactions

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## Abstract

Phasic alertness facilitates conscious perception through a fronto-striatal network, including the supplementary motor area (SMA). The functioning of the ventral attentional network has been related to the alerting system, overlapping with the ventral branch of the superior longitudinal fasciculus (SLF III). In this study, we use repetitive transcranial magnetic stimulation (rTMS) and a conscious detection task with near-threshold stimuli that could be preceded by an alerting tone to explore the causal implication of the SMA in the relationship between phasic alertness and conscious perception. Complementary to SMA stimulation, a sham and an active condition (left inferior parietal lobe; IPL) were included. Deterministic tractography was used to isolate the right and left SLF III. Behaviorally, the alerting tone enhanced conscious perception and confidence ratings. rTMS over the SMA reduced the alerting effect on the percentage of perceived stimuli while rTMS over the left IPL produced no modulations, demonstrating a region-specific effect. Additionally, a correlation between the rTMS effect and the integrity of the right SLF III was found. Our results highlight the causal implication of a frontal region, the SMA, in the relationship between phasic alertness and conscious perception, which is related to the white matter microstructure of the SLF III.

**Key words:** conscious perception, phasic alertness, rTMS, superior longitudinal fasciculus (SLF)

## Introduction

Only a small fraction of all the information reaching our senses is consciously perceived and can be reported, being in some cases the object of voluntary action. According to some influential models on conscious access (Dehaene and Changeux 2011), large-scale networks including parietal and frontal regions, together with the thalamo-cortical loop, amplify and maintain conscious contents to be efficiently processed. In order to reach consciousness, a sufficient level of vigilance and bottom-up activation from early sensory regions is required, along with top-down amplification from higher association cortices.

Attention and conscious perception are closely related processes, and the former is considered one of the pre-requisites of the latter (Posner 1994; Dehaene and Naccache 2001; Chica and Bartolomeo 2012). Nevertheless, attention is a heterogeneous construct. Posner and colleagues proposed that the attention system could be divided into alerting, orienting, and executive control. Among them, the alerting network mediates the ability to prepare and sustain attention to process high-priority signals (Posner and Petersen 1990; Petersen and Posner 2012). Two types of alertness have been described: tonic or intrinsic alertness refers to the ability to sustain a certain level of activation over a period of time, while phasic alertness is defined as the capacity to increase response readiness for a short period of time subsequent to a warning signal (Sturm and Willmes 2001).

Recent research has shown that attentional subsystems interact with conscious perception differently (Koch and Tsuchiya 2007; Wyart and Tallon-Baudry 2008; Chica et al. 2012;

Colás et al. 2017). Behavioral enhancements of conscious perception have been observed when manipulating phasic alertness. In neglect patients, the phasic increase of alertness temporally ameliorates visual consciousness deficits (Robertson et al. 1998). In healthy participants, a short auditory tone also improves conscious perception of near-threshold stimuli (Kusnir et al., 2011; Botta et al., 2014; Chica et al., 2016; Petersen et al., 2017), modulating perceptual sensitivity and not only response criterion.

At the neural level, the midbrain-thalamic-anterior cingulate cortex (ACC) circuit is related to both intrinsic and phasic alertness (Sturm and Willmes 2001). In a recent functional magnetic resonance imaging (fMRI) study from Chica et al. (2016), phasic alertness was manipulated by means of an auditory tone in order to explore its interaction with the conscious perception of near-threshold stimuli, and the underlying neural networks. The caudate, ACC, supplementary motor area (SMA), and frontal eye fields (FEFs) showed an interaction between phasic alertness and consciousness, as they were more activated for consciously perceived as compared with non-consciously perceived targets, especially when the alerting tone was absent. An interaction between phasic alertness and consciousness was also observed in the functional connectivity between the ACC and the SMA (Chica et al. 2016).

The SMA is implicated in preparation, selection, and planning of controlled motor responses to external stimuli (Luppino et al. 1993; Brass 2002). Phasic alerting seems to be mediated by the interaction of the midbrain-thalamus-ACC system with premotor areas for movement preparation, such as the SMA. Additionally, the activation of the SMA is more prominent the faster the behavioral response, suggesting a prime effect over motor selection and preparation areas through inputs from the anterior alerting system (Yanaka et al. 2010). Finally, the SMA shows a wide range of white matter connections with motor areas, but also with the cingulate cortex, and the inferior frontal gyrus (Vergani et al. 2014).

In addition to this fronto-striatal circuit, alertness activates frontal and parietal regions (Sturm et al. 1999, 2004; Coull et al. 2000, 2001; Clemens et al. 2011; Sturm and Willmes 2001). The fronto-parietal attentional network is often subdivided into the dorsal and ventral networks, which can be clearly segregated on the basis of their functional connectivity under task-free conditions and their responses profiles in different attentional paradigms (Corbetta et al. 2008; Leitao et al. 2015). The main components of the dorsal network are the FEFs, the superior parietal lobe, and the intraparietal sulcus. The ventral network is composed of the temporo-parietal junction and the ventral frontal cortex (Corbetta and Shulman 2002; Vossel et al. 2014). The ACC and the SMA have also been related to the ventral network (Yeo et al. 2011). It has been suggested that the ventral network constitutes an alerting system that acts as a circuit breaker when behaviorally relevant stimuli are detected (Corbetta and Shulman 2002; Corbetta et al. 2008; Kim 2014).

This segregation between dorsal and ventral networks is also present in white matter structural connectivity, which is mediated by the superior longitudinal fasciculus (SLF). The ventral branch of the SLF (SLF III) links regions of the inferior parietal lobe (IPL) and inferior frontal gyrus, overlapping with the ventral network (Thiebaut de Schotten et al. 2011; Parlatini et al. 2016). Current evidence relating phasic alertness with the SLF structure is scarce (see Klarborg et al. 2013). However, in a recent study, Chica et al. (2017) found that increased integrity of the left SLF III was predictive of the neural interactions observed between phasic alerting and conscious perception.

In the present study, we used repetitive transcranial magnetic stimulation (rTMS) to explore the causal contribution of the SMA to the interaction between phasic alertness and conscious perception (Chica et al. 2016). By means of the transient inhibition of the SMA, we expected to disrupt the association with ACC, a region that is too deep to be reached with TMS stimulation. We manipulated phasic alertness using an auditory signal and explored conscious reports of near-threshold stimuli. We hypothesized that the inhibition of the SMA might alter the benefit that the alerting cue produces over conscious perception. We also stimulated the left IPL as an active control to ensure that the observed effects were region-specific. In Chica et al. (2016), the IPL was more activated for consciously perceived as compared with non-consciously perceived targets, but did not show any interactions with alertness.

A second aim of the present study was to explore the link between the integrity of the SLF III and the rTMS effect over the SMA. We hypothesized a possible correlation between the integrity of this tact and the effects of phasic alertness over conscious perception. We sustain our predictions based on the overlap between the ventral attentional network (associated with the alerting effect) and the SLF III, and the anatomical link of this system with the SMA and the ACC (Vergani et al. 2014).

## Methods

### Participants

A sample of 32 right-handed volunteers [16 females, mean age 24 years, standard deviation (SD = 2.84)] took part in the study. One participant was eliminated from the analysis for conditions developed in between the 2 sessions that could interfere with the stimulation effects. Participants were unexperienced with the task and reported to have normal or corrected-to-normal vision and audition. Participants had no neurological or psychiatric conditions and followed all the safety requirements to undergo MRI and TMS studies (Rossi et al. 2012). They signed an informed consent to participate in the experiment and received a monetary compensation for their time and effort (10 Euros/hour). The study was reviewed and approved by the Ethics Committee of the University of Granada and was carried out according to the recommendations of Helsinki Declaration.

### Apparatus and stimuli

E-prime software was used to control the presentation of stimuli, timing operations, and data collection (Schneider et al. 2002). Visual stimuli were presented on a computer screen (Benq T903, 19″ wide, 1280 × 1024, 60 Hz) while participants sat at approximately 120 cm from the screen. Three horizontally aligned black boxes (63 mm height × 67 mm width) were displayed against a gray background (63 lm/m$^2$). The 2 lateral boxes were centered 60 mm to the left and right of a central fixation point (plus sing, 5 × 5 mm), placed inside of the central box. The target was a Gabor stimulus that could appear inside the lateral boxes. Matlab 8.1 (http://www.mathworks.com) was used to create 100 Gabor stimuli (4 cycles/deg. spatial frequency, 25 mm in diameter, SD of 3 mm), with a maximum and minimum Michelson contrast of 0.92 and 0.02, respectively. Target contrast was manipulated before the experimental task in order to adjust the percentage of consciously perceived targets to ~50% (see Procedure section). The auditory signal consisted of a tone of 20.05 Hz and 75.7 dB.

## Behavioral Procedure

In the experimental task (Fig. 1A), each trial began with the presentation of the fixation point for a random duration of 500, 1000, or 1500 ms. Participants were explicitly instructed to fixate the central plus sign throughout all the experiment. In half of the trials, the alerting tone was presented for 34 ms. The target was randomly presented for 50 ms either at the left or right location (50% at each location). Participants were required to perform 2 consecutive tasks using target information. In the first task (conscious detection task), they had to report if they consciously detected the appearance of the target by pressing a left situated key ("n") if the stimulus appeared in the left location and a right situated key ("m") if it appeared in the right location. They were asked to respond as accurately as possible (within the 1454 ms) and only when they were confident about their perception. If they had not seen the target, they had to press the "space bar". In the second task (confidence response task), participants had to rate the security of their response from 1 to 4 (1 = "no experience," 2 = "weak experience," 3 = "almost clear experience," and 4 = "clear experience") using 4 consecutive keys in the keyboard ("q," "w," "e," and "r"). They had a fixed time for each response (1454 ms and 1800 ms, respectively), making the total duration of the experimental task constant across participants (12 min). The experimental task consisted of 156 trials, 38.5% of which were catch trials (the target was not presented).
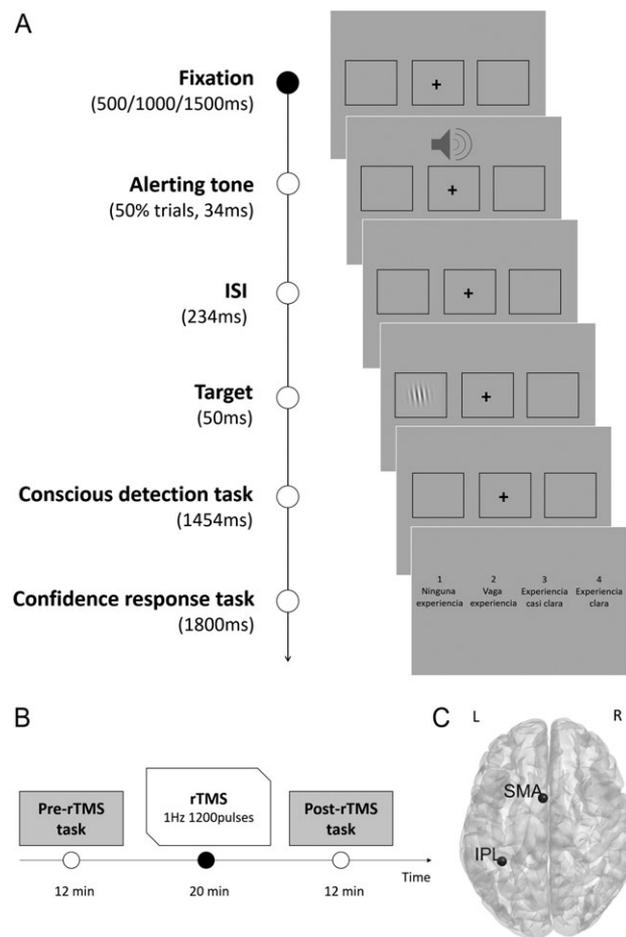
Before the experimental trials, Gabor contrast was titrated for each participant while they performed the above-described task. Titration began with a supra-threshold stimulus (Michelson contrast = 0.184), whose contrast was manipulated in successive blocks depending on the mean percentage of seen targets after every 14 trials. After each block, if participants reported seeing 63% or more targets, Gabors at the immediately following lower contrast level (Michelson contrast minus 0.009) were used during the next block of trials; besides, if the percentage of seen targets was equal or lower than 38%, the next block of trials used Gabors at the immediately following higher contrast level (Michelson contrast plus 0.009). The titration procedure stopped when target contrast yielded a percentage of seen targets ranging between ≥38% and ≤63% for 2 consecutive blocks of trials. This contrast value was used in the behavioral tasks before and after rTMS for the same session, but titration was repeated in the second session to ensure that the percentage of seen targets would be ~50% in both sessions.

## rTMS Procedure

Structural T1-weighted MRI scans were acquired for all participants at the Mind, Brain, and Behavior Research Center (CIMCYC), University of Granada. We used a 3-T Siemens magnetization prepared rapid gradient echo, flip angle = 7, repetition time (RT) = 2530 ms, echo time (TE) = 3.5 ms, slice thickness = 1 mm, and field of view (FOV) = 256 mm.

rTMS was delivered by means of a biphasic repetitive stimulator (Super Rapid 2, Magstim, Whitland UK) and a 70-mm TMS figure-of-eight coil (Magstim, Whitland UK) positioned at ~45° respect to the scalp. rTMS patterns consisted of 1200 pulses applied at 1 Hz with an inter-pulse interval of 1 sec (for a total duration of 20 min). Previous studies have suggested that this protocol transiently reduces cortical excitability within the stimulated sites outlasting for approximately 50–75% of the stimulation duration (Chen et al. 1997; Maeda et al. 2000; Muellbacher et al. 2000; Hilgetag et al. 2001; Wagner et al. 2007). The time window of reduced excitability in our study was then estimated in about 10–15 min (task duration was 12 min).

The TMS coil was controlled by a robotic arm (TMS Robot; Axilum Robotics) and a TMS neuronavigation system (Brainsight; Rogue Systems, Montreal, Canada) with the capacity to estimate and track in real time the relative position, orientation, and tilting of the coil on the sectional and 3D reconstruction of the participants MRI with a precision of 5 mm. As previously done elsewhere (Chica et al. 2011; Bourgeois et al. 2013), we used a fixed TMS intensity of 75% of the maximum stimulator output (MSO) throughout all the participants. All participants received stimulation at supra-threshold levels with respect to their individual motor thresholds (mean motor thresholds for the SMA group: 66% of MSO and for the IPL group: 63% of MSO).

The experiment consisted of 2 groups. One of them received rTMS over the SMA and the other over the left IPL (Fig. 1C). The coordinates of the SMA (MNI coordinates, $x = -3$, $y = 9$, $z = 60$) and IPL (MNI coordinates, $x = -36$, $y = -42$, $z = 42$) were extracted from a previous study by Chica et al. (2016). Although the SMA stimulation coordinates were lightly left-lateralized, given the spatial resolution of TMS, we assumed a bilateral stimulation of the SMA. Each group attended to 2 sessions in which participants performed the experimental task before and after receiving the offline rTMS protocol (Fig. 1B). In one session,



**Figure 1.** Experimental design. (A) Example of the stimuli presentation time line in the experimental task. (B) Illustration of the protocol followed in a session. (C) Stimulation site in each of the groups: SMA (MNI coordinates: $x = -3$, $y = 9$, $z = 60$) and left IPL (MNI coordinates: $x = -36$, $y = -42$, $z = 42$).

**Table 1** Mean Percentage of Seen Targets, FA, RTs (in ms), Confidence Ratings, a' and Beta" Values in the SMA and the Left IPL Groups, for Each Experimental Condition. Standard Error of the Mean is Presented in Parentheses

| | | | Seen Targets | FA | RTs | Confidence | A' | Beta" |
|---|---|---|---|---|---|---|---|---|
| **rTMS over SMA** | | | | | | | | |
| Sham | Pre | Tone | 43.00 (3.98) | 3.20 (1.13) | 707.44 (23.90) | 1.46 (0.13) | 0.83 (0.01) | 0.79 (0.07) |
| | | No tone | 28.43 (3.66) | 0.27 (0.27) | 754.45 (23.84) | 1.32 (0.12) | 0.82 (0.01) | 0.96 (0.02) |
| | Post | Tone | 38.90 (3.40) | 1.33 (0.65) | 638.08 (18.33) | 1.33 (0.12) | 0.83 (0.01) | 0.89 (0.05) |
| | | No tone | 24.90 (3.36) | 1.33 (0.71) | 732.21 (25.66) | 1.25 (0.10) | 0.80 (0.01) | 0.89 (0.04) |
| rTMS | Pre | Tone | 46.83 (4.36) | 4.00 (1.33) | 694.23 (23.67) | 1.52 (0.13) | 0.84 (0.02) | 0.76 (0.07) |
| | | No tone | 33.27 (4.54) | 1.07 (0.62) | 743.29 (23.32) | 1.24 (0.11) | 0.82 (0.01) | 0.92 (0.04) |
| | Post | Tone | 47.67 (6.72) | 3.33 (1.29) | 644.38 (24.29) | 1.46 (0.11) | 0.84 (0.02) | 0.65 (0.14) |
| | | No tone | 40.60 (7.33) | 1.40 (0.70) | 708.53 (22.52) | 1.18 (0.09) | 0.85 (0.02) | 0.73 (0.14) |
| **rTMS over left IPL** | | | | | | | | |
| Sham | Pre | Tone | 51.44 (3.33) | 3.88 (2.31) | 682.59 (36.61) | 1.79 (0.14) | 0.86 (0.01) | 0.83 (0.07) |
| | | No tone | 39.88 (4.75) | 4.38 (1.34) | 752.87 (40.57) | 1.66 (0.16) | 0.82 (0.01) | 0.74 (0.06) |
| | Post | Tone | 53.63 (4.80) | 5.63 (2.76) | 641.30 (37.71) | 1.74 (0.16) | 0.86 (0.01) | 0.76 (0.09) |
| | | No tone | 39.19 (4.47) | 3.75 (1.48) | 696.63 (36.40) | 1.64 (0.16) | 0.82 (0.01) | 0.78 (0.07) |
| rTMS | Pre | Tone | 52.19 (4.89) | 2.88 (0.82) | 701.71 (43.19) | 1.64 (0.14) | 0.86 (0.02) | 0.73 (0.10) |
| | | No tone | 38.94 (5.11) | 1.63 (0.48) | 758.36 (38.80) | 1.47 (0.13) | 0.81 (0.04) | 0.80 (0.08) |
| | Post | Tone | 51.38 (5.49) | 2.88 (1.19) | 644.82 (44.09) | 1.70 (0.16) | 0.86 (0.02) | 0.76(0.10) |
| | | No tone | 39.63 (5.54) | 2.13 (0.62) | 721.48 (45.25) | 1.46 (0.14) | 0.83 (0.02) | 0.81 (0.05) |

real rTMS was applied either to the SMA or the left IPL (depending on the group). In the other session, sham rTMS was applied by placing the coil in the same stimulation site while a recording of the original rTMS sound was reproduced by 2 hidden speakers. This procedure was effective in mimicking the clicking noise associated with the delivery of TMS pulses as none of the participants was able to distinguish between the real and sham rTMS session. The order of the real and sham rTMS sessions was counterbalanced between participants. The mean interval between both sessions was 10 days (SD = 8).

## Diffusion Tensor Imaging (DTI) Analysis

A total of 70 near-axial slices were acquired on a Siemens 3-T system using a sequence fully optimized for DTI of white matter, providing isotropic ($2 \times 2 \times 2$ mm) resolution and coverage of the whole head with a posterior-anterior phase of acquisition (ET = 88 ms and RT = 8400 ms). At each slice location, 6 images were acquired with no diffusion gradient applied and 60 diffusion-weighted images in which gradient directions were uniformly distributed in space. The diffusion weighting was equal to a $b$-value of 1500 s mm$^2$. In each slice, diffusion-weighted data were simultaneously registered and corrected for subject motion and geometrical distortion adjusting the gradient accordingly (ExploreDTI, http://www.exploredti.com).

Individual dissections of the tracts were carried out with the software TrackVis (http://www.trackvis.org). The ventral branch of the superior longitudinal fasciculus (SLF III on the left and right hemisphere) was isolated using a multiple region of interest approach. A frontal ROI around the white matter of the inferior frontal gyri and a ROI around the white matter of the parietal lobe were delineated. A no-part ROI in the temporal white matter was used to exclude streamlines of the arcuate fasciculus projecting to the temporal lobe (Thiebaut de Schotten et al. 2011; Rojkova et al. 2016). A new index employed as a surrogate for tract microstructural organization (mean Hindrance Modulated Orientational Anisotropy or HMOA; Dell'Acqua et al. 2013) was extracted from each dissected tract on the left and right hemisphere.

## Statistical Analysis

To test for the phasic alertness effect and its rTMS modulation, we analyzed the percentage of seen targets and the RTs in the conscious detection task, and the confidence rating in the confidence response task. We also analyzed participants' responses to the conscious detection task by using the signal detection theory (Abdi 2007). We computed the mean percentage of seen targets when the Gabor was presented (hits), and the mean percentage of seen targets when the Gabor was absent (false alarms; FA). We computed a non-parametrical index of perceptual sensitivity: A' = $0.5 + (((\text{Hits} - \text{FA}) * (1 + \text{Hits} - \text{FA}))/(4 * \text{Hits} * (1 - \text{FA})))$; and response criterion: Beta" = $(((\text{Hits} * (1 - \text{Hits})) - \text{FA} * (1 - \text{FA}))/(\text{Hit} * (1 - \text{Hit}) + \text{FA} * (1 - \text{FA})))$. A' values usually range between 0.5 (the signal cannot be distinguished from the noise) and 1 (perfect performance). For beta" values close to 1 indicate a conservative criterion, whereas values close to $-1$ indicate a non-conservative criterion (Stanislaw et al. 1999). For each dependent variable (see Table 1) and each group (SMA and left IPL), we conducted a repeated-measured analysis of variance (ANOVA) with the within participant factors of time (pre- and post-rTMS), TMS (rTMS and sham), and alerting (tone and no tone).

RTs shorter than 150 ms were considered outliers and were eliminated from the analysis (1% of the trials, SD = 0.08). Trials in which participants incorrectly reported the location of a present Gabor (0.3% of the trials, SD = 0.07) were also excluded. FA accounted for only 3% (SD = 0.16) of the catch trials.

In order to explore the role of the SLF III in the rTMS effect, we conducted Pearson correlation analysis using Z-scores. The mean HMOA value of the left and right SLF III was correlated with the rTMS effect over the percentage of seen targets (% seen targets post-rTMS for rTMS minus sham in the no-tone minus the tone condition), for both the SMA and the left IPL groups.

## Results
### Behavioral Results

*rTMS Over the SMA*

For the analysis of the mean RTs, we found a main effect of time, $F(1, 14) = 19.15$, mean squared error (MSE) = 3040,

$P = 0.001$, $\eta^2_p = 0.58$, and alerting, $F(1, 14) = 35.15$, MSE = 3451, $P < 0.001$, $\eta^2_p = 0.72$. Participants' RTs were shorter in the post-rTMS condition than in the pre-rTMS condition, and in the tone condition than in the no-tone condition. No other main effects or interactions were found (all $P$'s > 0.210).

For the percentage of seen targets, we found a main effect of TMS, $F(1, 14) = 5.65$, MSE = 355.4, $P = 0.032$, $\eta^2_p = 0.29$, and alerting, $F(1, 14) = 30.60$, MSE = 152.8, $P < 0.001$, $\eta^2_p = 0.69$. More targets were perceived in the tone compared with the no-tone condition, and in the rTMS compared with the sham condition. The interaction between time and alerting, $F(1, 14) = 4.94$, MSE = 20, $P = 0.043$, $\eta^2_p = 0.26$ demonstrated that the alerting effect was reduced in the post-rTMS as compared with pre-rTMS condition. Although the interaction between time, TMS, and alerting was not significant, $F(1, 14) = 2.20$, MSE = 30, $P = 0.160$, $\eta^2_p = 0.14$, we had a clear *a priori* hypothesis about a TMS by alerting interaction after rTMS was applied. We therefore performed a 2 × 2 ANOVA with the factors of TMS and alerting in the pre-rTMS and post-rTMS conditions. This analysis demonstrated that the interaction between TMS and alerting was far from significance pre-rTMS ($F < 1$), whereas it was significant post-rTMS, $F(1, 14) = 8.72$, MSE = 22.3, $P = 0.010$, $\eta^2_p = 0.38$. Planned comparisons revealed that the percentage of seen targets was significantly larger for the rTMS compared with the sham condition in the no-tone condition, $F(1, 14) = 6.08$, $P = 0.027$, but not in the tone condition, $F(1, 14) = 2.80$, $P = 0.116$. To explore the *a priori* hypothesis of a time by alerting interaction in the rTMS group, we conducted a 2 × 2 ANOVA with the factors of time and alerting for the sham and rTMS conditions. It demonstrated that the interaction between time and alerting was far from significance for the sham condition ($F < 1$), whereas it was significant for the rTMS condition, $F(1, 14) = 6.90$, MSE = 23.7, $P = 0.020$, $\eta^2_p = 0.33$. Although planned comparisons revealed that the difference for the percentage of seen targets between the pre- and the post-rTMS conditions was not significant either for the tone or the no-tone conditions ($F > 1$ and $F(1, 14) = 1.14$, $P = 0.303$, respectively), the difference between pre- and post-rTMS conditions was greater for the no-tone condition. These results demonstrated that after rTMS was applied over the SMA, the percentage of seen targets was increased compared with the sham condition for the no-tone condition, leading to a reduction of the alerting effect (see Fig. 2). No other main effects or interactions were found (all $P$'s > 0.100).

For a', we observed a main effect of alerting, $F(1, 14) = 6.54$, MSE = 0.002, $P = 0.023$, $\eta^2_p = 0.32$ and an interaction between TMS and alerting, $F(1, 14) = 4.85$, MSE = 0.0003, $P = 0.045$, $\eta^2_p = 0.26$. Perceptual sensitivity was overall larger for the tone than the no-tone condition, and this alerting effect was larger for the sham than the rTMS condition. Although the interaction between time, TMS, and alerting was not significant, $F(1, 14) = 1.41$, MSE = 0.001, $P = 0.255$, $\eta^2_p = 0.09$, we had a clear *a priori* hypothesis about a TMS by alerting interaction after rTMS was applied. A 2 × 2 ANOVA in the post-rTMS condition with the factors of TMS and alerting revealed a significant interaction between TMS and alerting, $F(1, 14) = 6.71$, MSE = 0.0004, $P = 0.021$, $\eta^2_p = 0.32$, whereas in the pre-rTMS condition, no interaction was found ($F < 1$). Planned comparisons demonstrated that the perceptual sensitivity was significantly greater for the rTMS than the sham condition in the no-tone condition, $F(1, 14) = 5.40$, $P = 0.036$, but not in the tone condition, $F(1, 14) = 1.87$, $P = 0.19$. To explore the *a priori* hypothesis of a time by alerting interaction in the rTMS group, we conducted a 2×2 ANOVA with the factors of time and alerting for the sham and rTMS conditions. It demonstrated that for the sham condition, there

was a significant interaction between time and alerting, $F(1, 14) = 3.39$, MSE = 0.0003, $P = 0.087$, whereas for the rTMS condition, the interaction was not significant ($F < 1$). This suggests that the rTMS over the SMA seems to increase the perceptual sensitivity compared with the sham condition in the no-tone condition, preventing for the increase of the alerting effect over the perceptual sensitivity that is observed in the sham condition in the post-rTMS task. No other main effects or interactions were found (all $P$'s > 0.150).

For beta", we found a main effect of alerting, $F(1, 14) = 5.01$, MSE = 0.065, $P = 0.042$, $\eta^2_p = 0.26$, and an interaction between time and alerting, $F(1, 14) = 5.16$, MSE = 0.023, $P = 0.039$, $\eta^2_p = 0.27$. Response criterion was more conservative in the no-tone condition than in the tone condition, and this difference was reduced in the second session as compared with the first session both for the rTMS and the sham conditions. The interaction between time, TMS, and alerting was far from significance ($F < 1$) and therefore it was not further explored. No other main effects or interactions were found (all $P$'s > 0.116).

Finally, for the confidence ratings, we found a main effect of alerting, $F(1, 14) = 20.65$, MSE = 0.05, $P < 0.001$, $\eta^2_p = 0.60$, with higher confidence ratings in the tone condition than in the no-tone condition. No other main effects or interactions were significant (all $P$'s > 0.086).

### rTMS Over the Left IPL

For the analysis of the mean RTs, we found a main effect of time, $F(1, 15) = 15.53$, MSE = 4712, $P = 0.001$, $\eta^2_p = 0.51$, and alerting, $F(1, 15) = 32.06$, MSE = 4182, $P < 0.001$, $\eta^2_p = 0.68$. Participants' RTs were shorter in the post-rTMS condition than in the pre-rTMS condition, and in the tone condition than in the no-tone condition.

For the percentage of seen targets, a main effect of alerting was found, $F(1, 15) = 40.60$, MSE = 128.1, $P < 0.001$, $\eta^2_p = 0.73$, with more targets perceived in the tone condition than in the no-tone condition. However, no interactions were significant (all $P$'s > 0.455).

The signal detection theory analyses revealed a main effect of alerting for a', $F(1, 15) = 17.19$, MSE = 0.003, $P = 0.001$, $\eta^2_p = 0.53$, with increased perceptual sensitivity in the tone condition than in the no-tone condition, whereas no main effects or interactions were found for beta". Finally, for the confidence ratings, a main effect of alerting was found, $F(1, 15) = 26.74$, MSE = 0.032, $P < 0.001$, $\eta^2_p = 0.64$, demonstrating higher confidence ratings in the tone condition than in the no-tone condition. No other main effects or interactions were found (all $P$'s > 0.185 for all analyses).

In sum, results demonstrate that the tone was effective in producing an alerting effect, with more targets perceived, higher perceptual sensitivity, and shorter responses in the tone condition than in the no-tone condition. Phasic alertness also affected metacognitive aspects of consciousness, with higher confidence ratings in the tone condition as compared with the no-tone condition. However, alerting effects over response criterion seem to be less replicable, as modulations in response criterion were only found for the SMA group. When rTMS was applied over the SMA, the alerting effect observed for the percentage of seen targets was reduced post-rTMS as compared with the pre-rTMS task and this effect was driven by an increase of seen targets when the alerting tone was absent. Signal detection theory analyses indicated that the rTMS modulation over the alerting effect is related to changes in perceptual sensitivity rather than to changes in response criterion.
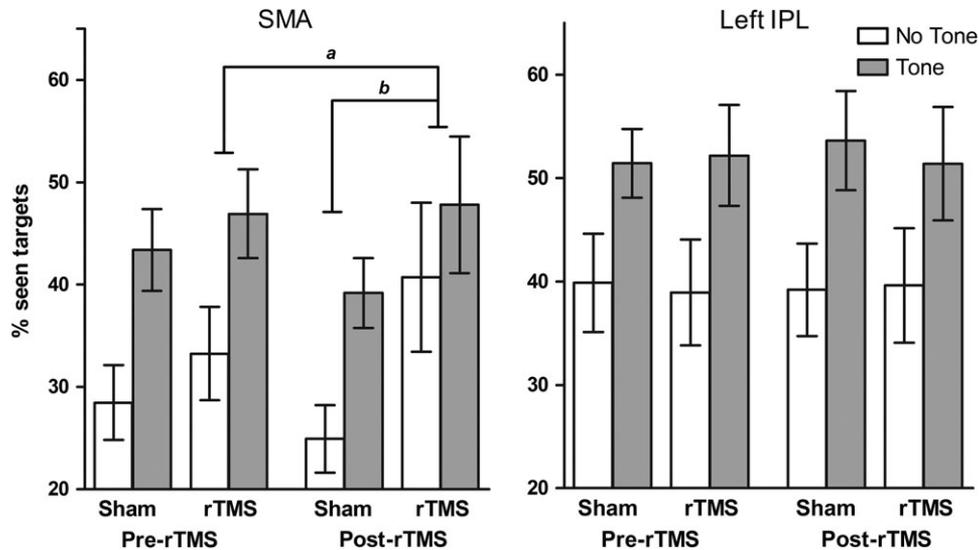
**Figure 2.** Percentage of seen targets for tone and no-tone conditions before and after rTMS for the sham and rTMS conditions in the SMA and the left IPL groups. (*a*) The 2 × 2 ANOVA significant interaction between time and alerting for the rTMS condition [$F(1, 14) = 6.90$, MSE = 23.7, $P = 0.020$, $\eta^2_p = 0.33$] and (*b*) the 2 × 2 ANOVA significant interaction between TMS and alerting for the post-rTMS task [$F(1, 14) = 8.72$, MSE = 22.3, $P = 0.010$, $\eta^2_p = 0.38$]. Errors bars represent standard errors of the mean.

Alerting effects on RTs, response criterion, or confidence ratings were not significantly modulated by rTMS over the SMA. Moreover, rTMS over the left IPL did not modulate the alerting effect (for any of the dependent variables analyzed), demonstrating that the rTMS effect over the SMA was region-specific.

### Tractography Analysis

As previously reported, a statistically significant rightward asymmetry was found for the HMOA index of the SLF III, with greater HMOA values on the right (mean HMOA = 0.104) than the left hemisphere (mean HMOA = 0.091), $t(30) = 8.830$, $P < 0.001$.

Results demonstrated a negative correlation between the rTMS effect in the SMA group for the percentage of seen targets with the integrity of the right SLF III ($r = -0.628$, $P = 0.012$), whereas the correlation with the integrity of the left SLF III did not reach significance ($r = 0.176$, $P = 0.531$). The more increased the integrity of the right SLF III, the more reduced the rTMS effect for no-tone as compared with tone trials (Fig. 3).

When exploring the data in the 2 different alerting conditions independently, we observed that the integrity of the right SLF III was significantly correlated with the rTMS effect in the no-tone condition ($r = -0.582$, $P = 0.023$), while the effect did not reach significance in the tone condition ($r = -0.428$, $P = 0.112$).

A similar analysis in the left IPL group demonstrated no significant correlations either for the right SFL III ($r = -0.083$, $P = 0.760$) or the left SLF III ($r = -0.431$, $P = 0.095$).

### Discussion

The present study was designed to explore the contribution of the SMA to the interaction between phasic alertness and conscious perception, by means of rTMS as a causal perturbation method. We used an experimental paradigm that allows manipulating phasic alertness and measuring its effects over the conscious perception of near-threshold stimuli. A second aim of the study was to explore the influence of the white matter microstructure of the SLF III over the rTMS effects found.

Consistent with previous results (Kusnir et al. 2011; Botta et al. 2014; Chica et al. 2016; Petersen et al. 2017), the alerting tone was effective in modulating the conscious perception of peripheral visual stimuli. The sensory quality of the stimuli was improved, as shown by the increase in the percentage of seen targets and the perceptual sensitivity in the tone as compared with the no-tone condition. In addition, phasic alertness sped up motor responses, as reaction times were shorter in the tone than in the no-tone condition. In this study, the alerting tone was presented after a variable fixation interval in order to exclude factors related to the temporal orienting attention (Coull and Nobre 2008; Yanaka et al. 2010; Rohenkohl et al. 2011). In our paradigm, the alerting effect over response criterion was less replicable, since the modulation observed in the SMA group was not replicated in the IPL group. Therefore, alerting effects over the percentage of seen targets seem to be more related to changes in the perceptual sensitivity than response criterion. The inclusion of a metacognitive measure (confidence response) provided new data about the relationship between conscious perception and the subjective report of the experience. Phasic alertness affected metacognitive aspects of consciousness, with higher confidence ratings in the tone condition as compared with the no-tone condition.

It has been proposed that phasic alertness could improve conscious perception by the activation of fronto-parietal networks through the ascending thalamic–brain stem projections (Robertson et al. 1998; Kusnir et al. 2011). According to Lamme (2003), the activation of the fronto-parietal network, previous to the presentation of the target, would facilitate conscious access. However, these factors seem to be necessary but not sufficient for conscious perception, as the alerting potentiation effect does not inevitably lead to conscious perception. In line with this hypothesis, a phasic alerting tone helps improving perceptual spatial neglect in patients with right hemisphere damage (Robertson et al. 1998). The thalamic–brain stem projections recruited by phasic alertness should remain intact in such
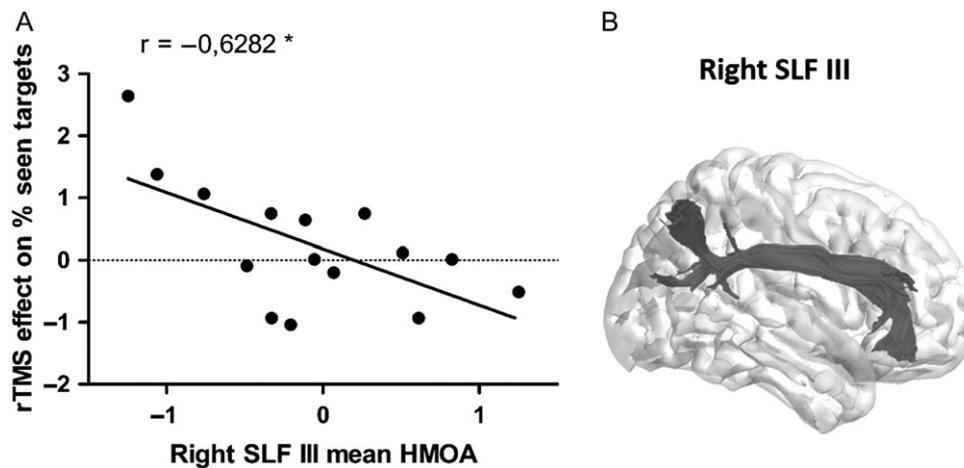
**Figure 3.** (A) Pearson correlation between the rTMS effect over the SMA on the percentage of seen targets and the mean HMOA of the right SLF III. Line of best fit ($y = -0.91x + 0.18$; $r^2 = 0.40$). (B) Virtual *in vivo* dissection of the right SLF III using deterministic tractography.

patients and may activate the right fronto-parietal regions that are not damaged, compensating for their perceptual spatial deficits (Robertson et al. 1998).

As expected based on our previous work (Chica et al. 2016), rTMS over the SMA produced a modulation of the interaction between phasic alertness and conscious perception, demonstrating its causal implication. The facilitation effect of phasic alertness over conscious perception was reduced after rTMS over SMA, as compared with the pre-rTMS condition and the sham stimulation condition. However, this reduction of the facilitation effect was led by an increase on the percentage of seen targets in the no-tone condition (see Fig. 2). This result was unexpected because the 1 Hz rTMS protocol is assumed to have an inhibitory effect based on the evidence of studies in which TMS is applied over motor areas, where a behavioral effect can be observed (Chen et al. 1997; Maeda et al. 2000; Muellbacher et al. 2000; Hilgetag et al. 2001; Wagner et al. 2007). However, we cannot ensure an inhibitory effect over other non motor areas, such as the SMA. The increase in the percentage of seen targets in the no-tone condition after the rTMS may be due to an excitatory effect of the stimulation. The activation of the SMA (and probably its connection with the ACC) would lead to an enhancement on conscious perception, which was more evident in the no-tone condition (as in the tone condition, the performance could not be enhanced over the threshold of consciousness).

Another plausible explanation may be the role of the SMA and neighboring areas on action inhibition (Sumner et al. 2007). The rTMS disruption of this function could lead to an action facilitation that would be more evident in the no-tone condition. According to that possibility, one would expect rTMS effects also over RTs, which are not observed. However, in this experiment, participants were instructed to response accurately to the detection task, priming precision rather than speed. As a result, mean RTs are longer (~700 ms, see Table 1) than in a classical location task (~390 ms, Gabay et al. 2012). Although previous studies have demonstrated the implication of the pre-SMA in motor selection and preparation, demonstrating effects on RTs (Yanaka et al. 2010), in a localization task like the one used here, the specific motor response cannot be prepared before target appearance, preventing for response preparation. This may also explain why rTMS over the SMA did not modulate RTs. These factors may prevent from observing

rTMS effects over RTs, and therefore, it cannot be considered as strong evidence against neither the action inhibition account nor the possibility of an excitatory rather than inhibitory effect of the rTMS protocol applied. In any case, rTMS over the SMA modulated the alertness and consciousness interaction, demonstrating a causal role of this area in the relationship between these 2 processes.

The confidence response was not modulated by rTMS, suggesting a dissociation in the role of the SMA between conscious access and the subjective experience of consciousness. This would be in accordance with data showing that metacognitive processes are more related with structures such as dorsolateral or rostrolateral prefrontal cortices (Fleming and Dolan 2012).

The rTMS over the left IPL did not lead to any significant modulation of the behavioral measures. Based on our previous study (Chica et al. 2016), we did not expect any interactions between phasic alertness and conscious perception associated with this region, constituting an active control to the effects found in the SMA. We may have expected a modulation of the overall conscious perception following its stimulation. Nevertheless, the results are in agreement with the theories proposing that conscious perception does not rely in a specific region but depends on a distributed fronto-parietal network (Dehaene and Changeux 2011). If that were the case, the inhibition of one particular region would not be enough to impair conscious perception.

Finally, we explored the structural microstructure of the ventral branch of the SLF. A right hemispheric lateralization of this tract was observed in the mean HMOA, as previously found with other measures (Thiebaut de Schotten et al. 2011). We found a negative correlation between the integrity of the right SLF III and the rTMS effect for no-tone as compared with tone trials. A good integrity of this fasciculus, connecting inferior parietal and frontal regions, could protect from the rTMS effects, especially in the no-tone condition. These results are in agreement with models based on fMRI data postulating the important role of the ventral network in saliency detection (Corbetta et al. 2008; Uddin 2014) and highlight the importance of taking into account white matter microstructure to better understand cognitive functions. In the present study, we show that rTMS modulations of the alertness and consciousness interactions depend on the integrity of the right SLF III. In a previous study by Chica et al. (2017), the integrity of the left SLF III was correlated with the neural interaction between alertness

and consciousness observed in the ACC. Although both results imply different SLF III hemispheres, the different nature of the measures correlated with the integrity of the tract may be on the base of the dissimilarities found.

Research linking white matter microstructure and behavioral or neurofunctional data is still scarce, and more investigation will be necessary to disentangle its particularities. However, this result could have important implications in the study of attention and consciousness deficits after brain damage. For example, patients suffering from neglect present important deficits in orienting attention and consciousness, and these deficits can be improved with phasic alerting (Robertson et al. 1998). Integrity of the right SLF III might be a good predictor of patients' response to phasic alertness treatments.

In conclusion, rTMS of the SMA modulated the interaction between phasic alertness and conscious perception, highlighting a causal role of this region, may be through its connection with the ACC (Chica et al. 2016). Additionally, the white matter microstructure of the ventral branch of the SLF was correlated with the rTMS modulation of the alerting effect. These results provided new data for the understanding of how different attentional networks interact with conscious perception and about the neural mechanisms underlying these interactions, hopefully lending to new clinical tools in the rehabilitation of consciousness impairments.

## Funding

## Notes

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