Low-frequency alternating current stimulation rhythmically suppresses gamma-band oscillations and impairs perceptual performance

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ARTICLE INFO

Keywords:
TACS-MEG
Visual cortex
Gamma
Alpha
Phase-amplitude coupling (PAC)
Visual detection

ABSTRACT

Low frequency oscillations such as alpha (8–12 Hz) are hypothesized to rhythmically gate sensory processing, reflected by 40–100 Hz gamma band activity, via the mechanism of pulsed inhibition. We applied transcranial alternating current stimulation (TACS) at individual alpha frequency (IAF) and flanking frequencies (IAF-4 Hz, IAF+4 Hz) to the occipital cortex of healthy human volunteers during concurrent magnetoencephalography (MEG), while participants performed a visual detection task inducing strong gamma-band responses. Occipital (but not retinal) TACS phasically suppressed stimulus-induced gamma oscillations in the visual cortex and impaired target detection, with stronger phase-to-amplitude coupling predicting behavioral impairments. Retinal control TACS ruled out retino-thalamo-cortical entrainment resulting from (subthreshold) retinal stimulation. All TACS frequencies tested were effective, suggesting that visual gamma-band responses can be modulated by a range of low frequency oscillations. We propose that TACS-induced membrane potential modulations mimic the rhythmic change in cortical excitability by which spontaneous low frequency oscillations may eventually exert their impact when gating sensory processing via pulsed inhibition.

1. Introduction

Cortical oscillations and their cross-frequency interaction are likely to constitute important mechanisms supporting the organization of neuronal processing. Alpha-band oscillations (8–12 Hz) are hypothesized to rhythmically gate information flow in the brain via pulsed inhibition of sensory processing, whereas gamma-band oscillations (40–100 Hz) are thought to support local processing (Jensen and Mazaheri, 2010; Klimesch et al., 2007; Tallon-Baudry and Bertrand, 1999). We aimed to test the hypothesis that stimulus-induced increase in gamma-band power in the visual cortex, associated with bottom-up visual processing (Bastos et al., 2015; Fries, 2015), can be actively modulated by the phase of slower oscillations, particularly in the alpha band. While correlational data from MEG studies in humans (Jensen and Mazaheri, 2010; Klimesch et al., 2007; Tallon-Baudry and Bertrand, 1999) and intralaminar recordings in monkeys (Spaak et al., 2012) has revealed coupling between alpha phase and gamma amplitude, the causal role of alpha oscillations in modulating gamma-band power remains unresolved. We therefore applied transcranial alternating current stimulation (TACS) at individual alpha frequency (IAF) to the visual cortex (Oz-Cz montage) in human volunteers performing a visual detection task to mimic the impact of alpha phase-related cortical excitability fluctuations on endogenous gamma activity during visual stimulus processing. A second goal of this study was to test (i) whether TACS is capable of modulating behaviorally relevant neuronal activity in the human brain, an assumption recently challenged by current estimates from modelling studies (Huang et al., 2017) and intracranial recordings (Lafon et al., 2017; Opitz et al., 2016) reporting intensities at the lower limit of effectiveness in humans, as well as cadaver studies (Voroslakos et al., 2018) (but see Opitz et al., 2017), and (ii) whether its effect can be attributed to transcranial as opposed to retinal stimulation (Kar and Krekelberg, 2012; Schutter, 2016). While simultaneous TACS-MEG recordings (Helfrich et al., 2014b, 2016) are limited by the spatial interference of stimulation and recording electrodes, both affixed to
the scalp, the combination of TDCS/TACS and MEG (Marshall et al., 2016; Neuling et al., 2015; Soekadar et al., 2013; Witkowski et al., 2016) allowed us to transcranially impose oscillating currents on the visual cortex without sacrificing any MEG sensors, which facilitates the assessment of stimulus-induced gamma power modulation in the visual cortex directly underlying the TACS electrodes (Oz-Cz montage). Using a combination of spatial filtering and TACS artifact suppression techniques, we extracted gamma-band oscillatory signals from the visual cortex during TACS and estimated cross-frequency coupling between TACS phase and gamma amplitude. To control for the potential impact of electrical stimulation of the retina and resulting retino-thalamo-cortical entrainment (Kar and Krekelberg, 2012; Schutter, 2016), we also applied TACS with a retinal stimulation montage (Fpz-Cz). To further assess the frequency-specificity of coupling between TACS phase and gamma amplitude, we applied TACS at two flanking frequencies of IAF -4 Hz and IAF +4 Hz. We hypothesized that occipital TACS (but not retinal control TACS) at alpha frequency should impose rhythmic excitability fluctuations in the visual cortex, mimicking the effect of spontaneous oscillations. Occipital TACS was therefore expected to cause a general decrease and, more specifically, a rhythmic suppression of visual stimulus-induced gamma power, and consequently a reduction of bottom-up visual stimulus processing and associated perceptual detection performance.

2. Material and methods

2.1. Participants

All participants were recruited from a database of the Radboud University Nijmegen. In total, 17 participants (5 males, 12 females, age: 24.3 ± 0.7 (mean ± SEM) with normal, or corrected-to-normal vision by contact lenses only, were included in the study. Sample size is in accordance with MEG studies successfully inducing gamma-band oscillations in visual cortex with the same stimulus in N = 14 (Hoogenboom et al., 2006) or applying simultaneous TCS in N = 5 (Soekadar et al., 2013), N = 14 (Garcia-Cossio et al., 2016), N = 16 (Hanley et al., 2016) or N = 17 (Neuling et al., 2015) subjects. All participants conformed to standard inclusion criteria for MRI, MEG, and TACS. Written informed consent was obtained prior to start of the experiment according to the Declaration of Helsinki. The study was approved by the local ethics committee. Participants were financially compensated at 10 Euros per hour. Data are reported from 15 participants; two participants had to be excluded since no visual stimulus-induced gamma-band response could be detected even during Sham periods.

2.2. Procedure

To gather a sufficient amount of trials, participants took part in two

Fig. 1. Experimental paradigm and setup. (A) Timeline of a single trial. Participants fixated a small white dot in the center of the screen and were allowed to blink, until 1500 ms later the white dot turned grey to indicate the end of the blink period. At 2500 ms an inward-moving grating appeared around the fixation dot, which contained a slowly rotating asterisk in its center. Participants had to report the direction of rotation, by button-press, as soon as the visual stimulus disappeared at 5900 ms and before the next trial started at 6900 ms. TACS was turned on 500 ms into the blink period and turned off 2400 ms after visual stimulus onset and 1000 ms before visual stimulus offset, thus lasting for 5400 ms each trial. (B) TACS electrode montage. TACS was applied via three 5 × 5 cm rubber electrodes attached in a dual-montage setup: an occipital montage with electrodes located at Oz and Cz, and a retinal montage with electrodes at Fpz and Cz, with electrode Cz used in both montages. The cables connected to the electrodes were twisted at the shortest possible distance and lead left-ward away from the head towards the shoulder and out of the MEG helmet. (C) Experimental design matrix. Seven different trial conditions were pseudorandomly intermingled: 2 montages (retinal, occipital) x 3 frequencies (IAF, IAF -4 Hz, IAF + 4 Hz plus one stimulation-free condition), with ~100 trials per condition, i.e. ~700 trials in total. Due to limitations in total stimulation duration per day (defined by the local ethics committee) the experiment was split into two sessions of ~350 trials each, separated by at least 1 day. (D) Group average topography of stimulus-induced gamma-band power in source space (DICS frequency domain beamforming). Virtual channels were extracted (LCMV time domain beamforming) from the 16 gridpoints in visual cortex showing the highest relative increase in gamma-band power from baseline.
Experimental sessions on two separate days, but with all experimental conditions tested in each session. A structural MRI was obtained on a separate day. In the beginning of the first session, participants were familiarized with the experimental task. Otherwise, both experimental sessions followed the same procedures: After TACS and ECG electrodes were applied, participants were familiarized with the stimulation, and individual stimulation intensity was determined. Then, participants were seated in the MEG, and four minutes of resting state data were collected (two minutes eyes-open, two minutes eyes-closed) before they performed a rotation-detection task in blocks of 10 min with short breaks in between. Total MEG time was 90 min per session.

2.3. Rotation detection task

Participants performed a rotation detection task in which they had to indicate by button-press the rotation direction of an asterisk in the center of an inward moving high-contrast grating (Fig. 1A). Participants were instructed to fixate a white dot on grey background in the center of the screen. Each trial started with a 1.5 s period in which participants were allowed to blink. Participants were asked to refrain from blinking as soon as the fixation dot turned grey. A ‘baseline’ period of two seconds followed after which an inward-moving (0.8°/second) black-and-white high contrast grating with concentric circles (2.5 cycles/degree) appeared on screen covering 8 degrees of visual angle (adapted from Hoogenboom et al., 2006) for 3.4 s. In the center of the inward moving grating an asterisk was present that slowly rotated either clock-wise or counter-clock wise. The rotation rate was continuously updated after each trial using an adaptive-staircase procedure (Watson and Pelli, 1983) so that participants were roughly 80% correct in detecting rotation direction. The goal of the task was to keep the participants fixated and assure a stable level of attention throughout the experiment, as well as to assess TACS effects on detection accuracy.

2.4. TACS

TACS was applied using a battery-driven NeuroConn DC+ stimulator (NeuroConn GmbH, Ilmenau, Germany) connected to three 5 × 5 cm conductive, non-ferromagnetic rubber electrodes that were attached to

Fig. 2. Occipital TACS rhythmically suppressed stimulus-induced gamma oscillations. (A) Time-frequency representations (TFR) of oscillatory power time-locked to visual stimulus onset for IAF TACS (TACS) vs. TACS-free trials (Sham) (see Fig. S2 for separate analyses of different TACS frequencies). Occipital TACS caused a stronger suppression of the gamma response than retinal TACS (p < 0.001), and only occipital but not retinal TACS decreased gamma power compared to Sham trials (all p < 0.001; Table S2). (B) TFRs as in A, but only for segmented and averaged timelocked to TACS peaks and with the x-axis normalized to phase-angles in radians (see Fig. S4 for separate analyses of all frequencies). Inserted curves represent the TACS cycle. Occipital TACS phasically suppressed stimulus-induced gamma-band activity relative to Sham TACS (orange bars underneath TFR indicate significant difference of the individual peak frequency gamma power from zero, pFDR < 0.05), whereas retinal TACS did not show any PAC relative to Sham. (C) Phase-amplitude coupling (PAC, as indexed by the ‘modulation index’, MI; Tort et al., 2010) between the phase of TACS and the amplitude of the stimulus-induced gamma power during visual stimulus presentation (after subtraction of PAC during TACS in pre-visual-stimulus baseline and surrogate PAC to control for the potential impact of residual TACS artifacts on PAC). PAC was larger for occipital TACS (blue) than for retinal TACS (orange) and for surrogate data for all TACS frequencies (all p < 0.005), whereas retinal TACS did not differ from surrogates (p > 0.3). See Fig. S4 for periods before and during visual stimulation. (D) Accuracy on the rotation-detection task was reduced for occipital compared to retinal TACS (p < 0.05), irrespective of TACS frequency. (E) The more the individual TACS-phase-gamma-amplitude-coupling differed between occipital and retinal TACS, the stronger was the individual performance decrease in rotation detection for occipital relative to retinal TACS (r13 = −0.58; p < 0.05).
the scalp following the international 10–20 system, creating an occipital montage (Oz-Cz) and a retinal montage (Fpz-Cz), which shared electrode Cz (Fig. 2B). The surface area of the scalp was thoroughly cleaned using alcohol and Nuprep skin preparation gel (Weaver and Company, Aurora, CO, USA). Then electrodes were attached using conductive Ten20 paste (Weaver and Company, Aurora, CO, USA) ensuring no paste was applied outside of the contact area of the electrodes (Marshall et al., 2016). Impedances were kept below 5 Kohm. Due to the stickiness of the paste no further mounting aids were required. Electrode cables were connected in a manner that minimized the size of the current loop formed on the scalp. Cables for the Oz-Cz montage were connected to the anterior side of electrode Oz, and to the posterior side of electrode Cz. Cables for the Fpz-Cz montage were connected to the posterior side of electrode Fpz, and the anterior side of electrode Cz. Switching between montages was handled by galvanically isolated switchbox that was controlled by the stimulation PC.

Several steps were taken to minimize the artifacts produced by the presence of additional material in the magnetically shielded room (MSR). First, all electrodes and cables, including connectors, were checked for ferromagnetic properties by moving the items inside the helmet while inspecting the effects on the MEG signal (several rubber electrodes had to be dismissed as they produced artifacts in the MEG signal). Second, a CAT6 electronically shielded cable connected the electrodes to the stimulator, which was located outside the MSR. The cable was fixed to the chair in the MSR to minimize movement. Third, the cables attached to the scalp were twisted for each montage to keep the resulting current loop as small as possible. The cables attached to the scalp ran left of the subject, fixed to the shoulder, downwards towards the chair, and away from the helmet.

Stimulation frequency was adjusted per participant based on the individual alpha frequency. To this end, the individual alpha frequency (IAF) was determined, with a resolution of 0.2 Hz, at the beginning of each session as the peak frequency of the difference in power spectra between an eyes-closed and eyes-opened resting state session (10.31 ± 0.41 Hz, mean ± SD across subjects). During the experiment, stimulation frequency was set for each trial to either IAF -4 Hz, IAF, or IAF +4 Hz (Fig. 2C).

Stimulation intensity was titrated at the beginning of each session to 90% of the individual retinal phosphene threshold, separately per montage (peak-to-peak TACS amplitude, no DC offset, for Oz-Cz: 963 ± 319 μA and Fpz-Cz: 231 ± 114 μA; mean ± SD, range for Oz-Cz: 450–1750 μA, for Fpz-Cz: 50–550 μA). Retinal phosphene threshold was determined by increasing the current strength of TACS at IAF from 100 mA in steps of 100 mA until the participants started to perceive retinal phosphene. Note that stimulation intensity was adjusted per montage, since the goal of the retinal montage was to control for retinal stimulation effects by increasing the proximity to the retina, while decreasing the proximity to the occipital cortex (in which case higher stimulation intensities would have been required). The choice of subthreshold intensity for both montages ensured that no visual phosphene perception interfered with (i) the transcranial stimulation effects, (ii) the visual stimulus-induced gamma responses, and (iii) the detection task performance.

During each trial (except for Sham trials), TACS was applied for ~5.4 s at one of the three frequencies (IAF -4 Hz, IAF, or IAF +4 Hz) and via one of the two montages (occipital Oz-Cz or retinal Fpz-Cz) (Fig. 2C). Stimulation started 1 s before the baseline period to allow for a build-up of potential entrainment effects and continued throughout the 2 s baseline period into the visual stimulus presentation period. The stimulation was turned off 2.4 s into the visual stimulation period after completing a full number of cycles at the particular stimulation frequency. In total, 700 trials (600 TACS trials + 100 Sham trials) were acquired per subject, distributed over two sessions (resulting in a total stimulation time of 27 min per session.), and pseudo-randomized in order (i.e., all seven experimental conditions randomly applied within each consecutive block of seven trials) to ensure that an equal amount of trials of each condition was completed after every session and that there would be no more than two direct repetitions of the same TACS condition.

2.5. MEG data acquisition

Whole-head MEG was recorded using a 275-channel axial gradiometer CTF system (CTF MEG systems, VSM MedTech Ltd.) sampling at 1200 Hz, with a hardware low pass filter at 300 Hz. Head localization coils were placed on the nasion, and in the left- and right-ear canals. The position of the head was recorded at the beginning of the experiment and was monitored, and adjusted during breaks using online head-position tracking if head motion exceeded 3 mm (Stolk et al., 2013). Eye-tracking was conducted throughout the experiment using an EyeLink 1000 eyetracker (SR Research Ltd, Ottawa, Canada), sampling at 2 kHz. Electrocardiogram (ECG) was recorded using two electrodes in a bipolar montage placed on the left collarbone, and right hip.

2.6. Data analysis

2.6.1. Preprocessing

Data analyses was conducted using the FieldTrip toolbox (Oostenveld et al., 2011) and custom Matlab scripts for Matlab 2014b (Mathworks, Natick, USA). First, trials that included blinks during the baseline- or visual stimulation period were detected by bandpass filtering the horizontal, and vertical motion eye-tracker channels between 1 and 15 Hz (4th order, two-pass, Butterworth). Trials that exceeded a z-score of 5 were rejected. Second, trials that included SQUID-jumps were detected by first high-pass filtering the data at 30 Hz (4th order, two-pass, Butterworth) to attenuate the stimulation artifact. Trials of which the first-order temporal derivative exceeded a z-score of 25 were rejected. This resulted in an average of 9% ± 8% (Mean ± SD) rejected trials per subject. The data were down-sampled to 600 Hz after epoching into trials from ~3.4–3.6 s after onset of the gamma-inducing visual stimulus.

2.6.2. DICS beamforming

To identify voxels in visual cortex for which visual stimuli induced a clear gamma band increase (and for which thus neuronal processing of the incoming stimulus can be assumed), we employed a Dynamic Imaging of Coherent Sources (DICS) beamformer (Gross et al., 2001) approach, which allows to locate the spatial origin of an oscillatory signal. Based on the DICS beamforming results, voxels of interest were then selected for further analysis. This two-step procedure was computationally less expensive than extracting virtual channels by LCMV beamformer from all possible grid points and performing power analyses for each one of them to select those voxels responding to visual stimulation. A single-shell head model (Nolte, 2003) was created from the individual MRIs. Next, an equally-spaced grid with 0.5 mm3 based on a standard MN1 template MRI with 1 mm3 resolution was created. This template grid was warped to each subject’s individual anatomy to easily average and compare gridpoints across subjects. Then, a spatial filter was designed to maximize the sensitivity to the expected gamma-band response produced by the visual stimulation. To this end, we selected the trials without stimulation and epoched them into a baseline period of ~2.0 to ~0.001 s and an activation period from 0.4 to 2.399 s after visual stimulus onset, thus creating two epochs of exactly 2.0 s length. After removing linear trends, we calculated the cross-spectral density (CSD) matrix for both baseline and activation epochs, as well as for both epochs combined, at 60 Hz with 15 Hz frequency smoothing using a multi-taper approach, thus resulting in an analyzed frequency band of 45–75 Hz. A common spatial filter was calculated using a DICS beamformer on the CSD of the combined baseline and activation data using 5% regularization. Note that the spatial filter was calculated on and applied to TACS-free Sham trials only to identify voxels showing a gamma response to visual stimulation. The choice of regularization parameter and the
rationale to use TACS-free data was motivated by the objective to achieve maximal sensitivity for the visually induced gamma band activity of interest. Including TACS trials in calculating the covariance matrix and adjusting the regularization parameter accordingly (e.g. 0%, or 5% of a tACS-free covariance matrix) resulted in suppression of all gamma band activity (see section FFT interpolation) and was thus abandoned. The resulting spatial filter was then applied to the activation and baseline data separately. To find the gridpoints showing the maximal increase in gamma-band power in response to visual stimulation, the relative gamma-power change from baseline was calculated by dividing for each voxel in source space the absolute change from baseline by the baseline gamma power. While omitting TACS trials during DICS beamformer calculation resulted in a filter less effective in removing TACS artifacts, it ensured good sensitivity to visually induced gamma oscillations, which was the key dependent variable of this study. Additional band-stop filtering, FFT interpolation and baseline subtraction were used instead for TACS artifact removal (see below).

2.6.3. LCMV virtual channels
To enhance sensitivity to the visually induced gamma-band response, we used Linear Constrained Minimum Variance (LCMV) beamforming (Van Veen et al., 1997) to extract virtual channel time courses from those gridpoints that showed the strongest gamma-band power increase from baseline (as previously identified by DICS beamforming) in each participant (Fig. 1D) and were located inside the visual cortex mask of the AAL atlas (Tzourio-Mazoyer et al., 2002), including all striate and extra-striate regions (method adapted from Marshall et al., 2016). From these data, we created a new grid with 10 gridpoints. After bandpass-filtering (40–70 Hz) to maximize spatial filter sensitivity to the gamma-band, the covariance matrix was calculated for epochs from −2.3–2.3 s relative to visual stimulus onset on Sham trials, and spatial filters were calculated using 5% regularization. The raw sensor-level data was multiplied by the resulting spatial filters to obtain virtual channel time courses for each of the 10 gridpoints. While omitting TACS trials during LCMV beamformer calculation resulted in a filter less effective in removing TACS artifacts, it ensured good sensitivity to visually induced gamma oscillations, which was the key dependent variable of this study. Additional band-stop filtering, FFT interpolation and baseline subtraction were used instead for TACS artifact removal (see below). For each subsequent analysis, we first analyzed each of the 10 time courses separately before averaging the results per subject.

2.6.4. FFT interpolation
The main focus of the study was the effect of low frequency TACS on visually-induced gamma-band oscillations. Therefore, our original strategy was to ignore the artifact-loaded signal at the stimulation frequency itself and only analyze the gamma-band power modulation with respect to the known TACS phase. However, while TACS was applied at lower frequencies (range: 5–16 Hz) and thus well outside the stimulus-induced gamma-band of interest (i.e., 45–75 Hz), the magnitude of the TACS artifact in the MEG signal was orders of magnitude larger than the magnetic fields produced by the brain, and the higher harmonics of the TACS frequency still affected frequency bins within the gamma-band of interest (Supplementary Fig. S1B-C). TACS artifacts and their harmonics could not be sufficiently suppressed by bandstop filters alone. LCMV spatial filters have previously been used to extract the brain signal of interest while attenuating the TACS artifact due to the suppression of correlated sources (e.g. Marshall et al., 2016; Neuling et al., 2015; Neuling et al., 2017), although a full suppression is mathematically impossible (Mikulecky et al., 2017). Unfortunately, we could not follow this approach, since LCMV spatial filter calculation based on TACS trials did not only suppress TACS artifacts, but also the stimulus-induced gamma-band response of interest: In fact, with that procedure, only 3 out of 17 participants still showed a clear gamma-band response during Sham trials, although the gamma-response is known to be very reliable (Hogenboom et al., 2006; Scheerings et al., 2009, 2011). In contrast, with our approach, only 2 out of 17 subjects did not show a visible gamma-band response. We therefore calculated LCMV spatial filters on Sham trials only, with the priority of preserving gamma-band responses in the visual cortex, while still attenuating TACS artifacts, though to a lesser degree. In addition, we employed an FFT interpolation approach (Fig. S1B,C) previously used to attenuate line-noise in EMG recordings (Mewett et al., 2001) to effectively suppress the TACS frequency and its harmonics in all conditions (using the MATLAB code for FFT interpolation that has been provided by Calvin Eiber and Alexander Pietersen under https://www.mathworks.com/matlabcentral/fileexchange/54228-remove-line-noise). Data were transformed to the frequency domain using a Fast Fourier transform (FFT) with 0.2 Hz frequency resolution (after zero-padding each trial to 5 s). Then, for each TACS trial the magnitude spectrum was interpolated from −1 to 1 Hz around the TACS frequency, and each of its harmonics up until the Nyquist frequency, while the phase spectrum remained intact. The narrowness of the interpolated bins ensured that the comparatively broad visually induced gamma-band activity remained intact. The interpolated frequency domain data was then transformed back into the time domain using an inverse FFT. As the effect of overall magnitude attenuation of the signal depended on the TACS frequency, we generated appropriate control conditions by applying the same FFT interpolation approach to copies of the Sham trials. The resulting frequency-specific Sham control conditions thus ensured fair comparisons even if harmonics inside the gamma frequency-band were interpolated. Importantly, we do not claim that our or any other approach is able to perfectly remove all TACS-related artifacts. The FFT interpolation does not account for potential side-band artifacts related to heartbeat and respiration (Nouy et al., 2016). However, unless considerable stimulus-induced variations in respiration and heart rate can be expected (e.g., for emotional stimuli), it is a reasonable assumption that residual TACS artifacts have comparable effects on the baseline and the period during visual stimulation, and consequently, baseline subtraction largely removes TACS-related residual artifacts (Kasten et al., 2018; Neuling et al., 2017; Nouy and Siegel, 2018). Therefore, baseline subtraction was performed for both time-frequency and PAC analyses.

2.6.5. Time-frequency analysis
To assess visual stimulus-induced gamma-band responses we calculated time-frequency representations (TFRs) of power by means of a sliding window FFT. We used the ’multitaper’ method as implemented in FieldTrip (ft.reanalysis), multiplying a series of orthogonal Slepian sequences (aka discrete prolate spheroidal sequence, DPSS) to achieve a well-controlled frequency smoothing of 10 Hz (Mitra and Pesaran, 1999). For each trial, these tapers were applied to a sliding time window of 500 ms that was moved in steps of 20 ms over the entire trial, estimating for each time step the power between 30 Hz and 100 Hz for times up to 1 Hz. While zero-padding of data segments to 10 s produced an artificial frequency resolution of 0.1 Hz, the true frequency resolution was at 2 Hz due to the 500 ms data window length. The mean and any linear trend were removed prior to calculating the FFT. The gamma-band response is usually best represented as relative change from baseline to account for its comparably low amplitude. During TACS trials, however, any residual noise in the baseline may thereby result in spuriously low ratios. We thus first subtracted the mean of the baseline (−1 to −0.2 s) from the activation period to remove residual frequency- and montage-specific TACS-related artifacts, as well as their heartbeat- and respiratory-related modulation (but see Neuling et al., 2017; Nouy et al., 2016), which per design were comparable in baseline and visual activation periods of the same condition. We then calculated the log-ratio with respect to a common baseline period derived from the average of all Sham trials and thus unaffected by TACS-artifacts (ensuring a fair comparison of the gamma-band response between conditions and relative to Sham trials). An unbiased estimate of individual gamma peak frequency was derived by first calculating, for all conditions, the relative change in gamma power from Sham baseline, fitting a 23rd order
To assess whether TACS phasically modulated the power of the visually-induced gamma-band responses, we evaluated the gamma-band power dynamics in TACS peak-locked TFRs. To this end, we first calculated TFRs of each trial as described in the previous paragraph, but decreased the size of the sliding time window to one cycle of the amplitude-providing frequency to be more sensitive to transient changes in the gamma-band across the TACS cycle. Also, data were normalized per trial to allow robust single-trial assessment of phasic gamma power modulation for subsequent TACS-phase-gamma-amplitude-coupling analyses. To take variations in signal-to-noise ratio into account (e.g., due to residual artifacts), TFRs were z-normalized trial-by-trial by subtracting the mean and dividing by the standard deviation of the trial’s baseline, resulting in an estimate of time-locked power that is relatively robust against noisy trials and extreme values (see Grandchamp and Delorme (2011)).

Next, we detected the peaks of the TACS cycle in the output copy of the TACS signal as provided by the stimulation device, using Matlab’s findpeaks’ function. Peaks were defined as the local maxima on the z-transformed stimulation signal with a minimum width of a quarter cycle of the stimulation signal, a minimum height of 1, and a minimum distance of 0.9 cycles. The data between 0.5 and 1.5 s after visual stimulus onset (thus excluding the initial evoked gamma response) were epoched into segments around each peak with a duration of 4 cycles of the respective TACS frequency. For each TACS frequency, a comparable segmentation was also applied to the Sham trials, using randomly chosen stimulation signals from the respective TACS trials. This resulted in a specific Sham control condition for each TACS frequency, effectively providing a surrogate distribution of gamma power values relative to the TACS cycle. Finally, averages were created for each of the TACS frequencies and respective Sham surrogates. As the number of epochs depends on the number of cycles (being higher for higher frequencies), we applied a random subsampling approach to create unbiased averages. We first determined the stimulation frequency with the smallest number of epochs and then averaged 500 randomly drawn subsamples of that size per condition. To allow direct comparison between TACS frequencies and averaging across subjects (with individualized IAF), we transformed the time-axis to radians by adjusting the step size during TFR calculation accordingly. Importantly, TACS peak-locked TFRs were calculated for both baseline and visual stimulation period. Since the baseline period does not contain any visually-induced gamma-band responses, but may contain residual TACS artifacts, it serves as an excellent control against TACS artifact-related spurious phase-amplitude coupling. Respective Sham TFRs were then subtracted from the TACS peak-locked TFRs, and individual gamma power values were compared for each phase angle (radians) with two-sided one-sample t-tests against zero using FDR correction for multiple comparisons (Fig. 2B and Fig. S3).

### Phase-amplitude coupling

To more formally quantify and compare the extent to which TACS phasically modulates the gamma-band response, we estimated phase-amplitude coupling (PAC) using Tort’s Modulation Index (MI) by calculating the normalized Kullback-Leibler (KL) divergence of the histogram of TACS phase-binned gamma amplitude to a uniform distribution (Tort et al., 2010). In case of significant PAC, the histogram diverges from a uniform distribution. To this end, the gamma-band amplitude was determined by convolving the virtual channel data between 0.5 and 1.5 s after visual stimulus onset with a 5-cycle moving time window multiplied with a Hanning taper for frequencies from 30 Hz to 100 Hz in steps of 1 Hz, while a 1 s time window was used for estimating the phase of the TACS signal similarly to the gamma-band magnitude (Jiang et al., 2015). The phase-difference between the gamma-band power envelope and TACS signal was subsequently calculated and compared to a uniform distribution using Tort’s Modulation Index (MI). As for TACS peak-locked TFRs, we randomly subsampled the data for each condition 500 times using a sample size equal to the lowest number of trials across conditions, to prevent any bias due to unequal trial numbers between conditions. MIs were calculated for each random subsample and then averaged. As a control, PAC was also estimated for surrogate data, for which the phase-providing TACS signal was randomly phase-shifted to create frequency-specific surrogate PAC values for each TACS condition. We used one-sample t-tests to test for significant PAC after subtracting PAC values at baseline before visual stimulus onset and respective visual-stimulation induced changes in surrogate PAC values. We used repeated-measures ANOVAs (no correction for non-sphericity was necessary) on the baseline- and TACS-corrected data, followed by paired-sample t-tests were appropriate, to compare PAC between TACS montage and frequency conditions.

### TACS artifact-to-brain-signal-ratio

To quantify TACS artifact size at sensor and source space and to exclude that a potential task-related modulation of TACS artifact size confounded our results, we performed a control analysis recently implemented by Kasten et al. (2018): We calculated the ratio of gamma power at baseline between TACS and TACS-free data both for the sensor level and source reconstructed data (prior to subtraction). At the sensor level, the ratio was 4058:1 for the occipital montage (for occipital sensors; MLO, MZO, and MRO) and 7:1 for the retinal montage (for retinal sensors). For the source reconstructed data the ratio between TACS and SHAM was reduced to 83 for the occipital montage and 2 for the retina control montage. Thus, expectably, residual artifacts were larger for the occipital montage at occipital sensors or occipital voxels. But notably, TACS artifacts were almost 50 times smaller at the source level compared to the sensor level. More importantly, to exclude that residual TACS artifacts differed during visual stimulation and baseline, we calculated the amplitude envelope of the MEG signal in the gamma range during visual stimulation and at baseline for occipital sensors, prior to source reconstruction and any artifact removal, and thus mainly reflecting the TACS artifact in the gamma range. Next, we calculated the amplitude ratio during visual stimulation relative to baseline. This ratio was averaged within each montage, over frequencies, and correlated with our main effects of interest: the montage effect on gamma power, PAC, and behavior. The average power ratios for both occipital as well as retinal control did not differ significantly from 1 (occipital $p > 0.8$; retinal $p > 0.7$). Furthermore, ratios for both montages did not differ significantly from each other ($p > 0.4$), and neither ratio correlated with any of our main effects of interest (all $p > 0.2$; see Results). The fact that the ratios did not differ significantly from 1 demonstrates that the TACS artifact amplitude did not change significantly during visual stimulus presentation, and that the difference observed after artifact correction is thus unlikely explained by a visual stimulus-related difference in TACS artifact amplitude.
3. Results

Participants performed a forced-choice visual discrimination task in which they had to report the rotation direction of a centrally presented asterisk inside an moving inward high-contrast grating (Fig. 1A), known to produce a pronounced gamma oscillatory response in early visual cortex (Hoogenboom et al., 2006) (Hoogenboom et al., 2006). During each trial, we applied TACS in either a visual (Oz-Cz) or a retinal (Fpz-Cz) montage and at either of three frequencies (i.e., IAF -4 Hz, IAF, +4 Hz; Fig. 1B and C) while recording ongoing brain oscillatory activity using whole-head magnetoencephalography (Fig. 1). Sham trials were intermingled as TACS-free reference epochs. Data from 15 of the 17 participants is reported here, as two showed no detectable gamma band response even in TACS-free Sham trials.

3.1. Occipital TACS suppressed average gamma power

Gamma power was extracted at individual’s gamma peak frequency (see Methods; see Fig. 1D for localization of group average gamma bands). Average gamma peak frequency was 57.6 Hz ± 8.5 Hz (mean ± SD). In all conditions, a significant increase in gamma-band power was observed during visual stimulus presentation (one-sample t-tests, all p < 0.001; Table S1). Importantly, visual stimulus-induced gamma responses differed between TACS montages as revealed by the main effect of a montage (occipital, retinal, sham) x frequency (IAF-4, IAF, IAF+4 Hz) rmANOVA (F1,14,0.9 = 26.08; p < 0.000001, η²p = 0.651), in that occipital TACS caused a stronger suppression of the gamma response than retinal TACS (paired-samples t-test averaged across TACS frequencies: t = 6.90, PBF < 0.0001, d = 1.781; see Fig. 2A, S2, and S3). In fact, only occipital TACS (t = 4.90, PBF < 0.001, d = 1.265) but not retinal control TACS (t = 0.26, PBF = 1) caused a decrease in gamma power compared to Sham trials, excluding a confound by retinal stimulation. There was no significant main effect for TACS frequency (F2,28 = 2.80, p = 0.08) but a significant interaction between montage and frequency (F1,56 = 4.38, p < 0.01, η²p = 0.238), driven by occipital TACS causing a larger suppression of gamma-band power for the IAF-4 Hz compared to IAF + 4 Hz condition (t = 4.18, p < 0.001, d = 1.080), whereas frequencies did not differ for retinal TACS or Sham (all p > 0.1).

3.2. Occipital TACS phase rhythmically modulated gamma power

To test whether the net suppression actually resulted from a phasic modulation, or more specifically, a rhythmic suppression of gamma-band power by TACS phase, we calculated TACS peak-locked TRFs (Fig. 2B; Fig. S3). Visually induced gamma-band power (at its dominant frequency of 57.6 Hz ± 8.5 Hz) decreased at particular points in the phase cycle (pDFDR < 0.05), but did not increase at any point. However, there were rhythmic increases around 40 Hz and 70–100 Hz for occipital, but not for retinal TACS between −π and 0 (pDFDR < 0.05). Although some activity in the 40 Hz range is also visible in the non-peak-locked TRFs (Fig. 2A), it is outside the range of the main visually induced gamma-band response, the modulation of which was in the focus of this study. To more formally quantify the strength of phasic gamma power modulation, we assessed phase-amplitude coupling (PAC) between TACS phase and gamma-band power in the visual cortex by calculating Tort’s Modulation Index (MI, Tort et al., 2010) both before and during visual stimulus presentation. We calculated the MI for each TACS condition at the individual peak-gamma frequency. For comparison, montage- and frequency-specific surrogate samples were created by phase-shifting the TACS signal from the respective TACS condition. PAC was significantly larger for occipital than for retinal TACS, as reflected by a significant main effect of montage (Fig. 2C) during visual stimulation (F1,14 = 64.53, p < 0.00001, η²p = 0.822), but neither a main effect of frequency (p > 0.6) nor an interaction (p > 0.6). Importantly, this effect remained significant (F1,14 = 17.06, p < 0.001, η²p = 0.549) after correcting for potentially spurious phase-amplitude coupling. To this end we subtracted PAC values calculated on the baseline period, which included TACS but lacked the visual stimulus-induced gamma response (pPACpTACS). Since any spurious PAC, that may potentially be introduced by residual TACS artifacts alone, is equally present in the baseline and in the visual stimulation period, this subtraction approach effectively protects against TACS artifact-related false positive PAC. In addition, we calculated the same difference in PAC values for the surrogate data (pPACpTACS(psurrogate−psurrogate)), the result of which was subtracted from the respective TACS difference ((pPACpTACS−pPACpTACS(psurrogate−psurrogate))). In fact, only occipital (t = 5.23, p < 0.001, d = 1.351) but not retinal control TACS (t = 0.168, p > 0.8) showed significant PAC during visual stimulus presentation relative to baseline and relative to the respective effect in the PAC surrogates (Fig. S4). The lack of a significant increase in PAC from baseline for retinal stimulation renders it unlikely that the effects observed during occipital stimulation are due to TACS artifacts, or an interaction between visually induced gamma oscillations and TACS. Thus, ruling out retinal entrainment and spurious PAC due to residual artifacts, occipital TACS did indeed produce a phasic modulation of stimulus-induced gamma power that was comparable in strength across stimulation frequencies.

3.3. Occipital TACS decreases rotation discrimination

Participants correctly identified the rotation direction in 78% of trials (SEM = 2%) with an average reaction time of 409 ms (SEM = 11 ms), for correct trials. There was a small, but significant decrease in hit rate for occipital (77.98% ± 2.66%) compared to retinal TACS (78.33% ± 2.26%) when taking into account the participant's baseline performance in Sham trials as a covariate in a 2 × 3 repeated-measures ANCOVA (montage x TACS frequency; see Fig. 2D). We observed a main effect of montage (F1,15 = 5.81, p = 0.029, η²p = 0.279) and an interaction between montage and baseline performance in the Sham trials (F1,15 = 11.69, p < 0.01, η²p = 0.438). The interaction indicates that subjects performing better in the rotation discrimination task during Sham also showed stronger TACS-related impairment than weakly performing subjects, a relationship that is also reflected by the correlation between performance during Sham trials and performance reduction during occipital TACS relative to retinal control TACS (r = 0.66, p < 0.007). Importantly, the effect of TACS montage on behavioral performance was correlated with the effect of TACS montage on PAC (r = −0.5753, p = 0.031) as validated with a robust regression analysis (r = −0.39, p = 0.03) that is less sensitive to outliers (Holland and Welsch, 1977) (Fig. 2E). Thus, subjects in which occipital TACS caused a stronger rhythmic suppression of stimulus-induced gamma-band activity (compared to retinal TACS) also suffered from stronger performance impairment in the rotation discrimination task for occipital TACS (compared to retinal TACS) (Fig. 2E).

4. Discussion

The aim of this study was to test a core prediction of the pulsed inhibition hypothesis (Jensen and Mazaheri, 2010; Klimesch et al., 2007), namely that the phase of slower oscillations, particularly in the alpha band, modulates the bottom-up processing of sensory information, which is reflected by stimulus-induced gamma-band oscillations (Bastos et al., 2015; Fries, 2015), and thereby affects perceptual performance. In line with this notion, we found that TACS to the occipital cortex (but not retinal control TACS) rhythmically suppressed visual stimulus-induced gamma-band power and that the degree of this suppression predicted the reduction in visual detection performance. The rhythmic fluctuations in visual cortex excitability imposed by TACS therefore seem to mimic the functional effects that are otherwise produced by the mechanism of pulsed inhibition during spontaneous alpha oscillations. However, since frequencies at IAF ± 4 Hz were equally effective, it is less likely that a specific entrainment of endogenous alpha oscillations mediated this
effect, and more likely that TACS caused a more direct modulation of cortical excitability by rhythmically shifting the neurons’ membrane potential.

4.1. TACS rhythmically suppresses visual-induced gamma power

The net suppression of visually induced gamma power by occipital TACS (Fig. 2A) ties in well with the observed phase-specific decrease (but not increase) in gamma power (Fig. 2B), suggesting that the corresponding excitability fluctuations imposed on the visual cortex by occipital TACS may be asymmetric, just as has been proposed for spontaneous alpha-band oscillations (Jensen and Mazaheri, 2010; Schalk, 2015). However, TACS at neighboring frequencies 4 Hz slower or faster than individual alpha frequency also caused a net suppression (Fig. 2A) and rhythmic modulation (Fig. 2C) of stimulus-induced gamma responses (with the lowest TACS frequency even causing the strongest net suppression). There are at least three potential explanations for the lack of frequency-specificity with respect to phase-amplitude coupling: Firstly, TACS at neighboring frequencies 4 Hz away from individual peak frequency may have still been able to entrain ongoing spontaneous alpha oscillations (Antal and Herrmann, 2016; Helfrich et al., 2014b). This would be in line with the targeted oscillatory network showing resonance properties reflecting an Arnold Tongue, according to which stimulation slightly off the endogenous frequency can still cause entrainment if applied at higher intensities (Ali et al., 2013), however, 4 Hz maybe too far off and intensities too low here. Secondly, TACS at neighboring frequencies may have entrained high theta band and low beta band oscillations instead, both of which phasically modulate gamma power in the visual cortex (Bastos et al., 2015; Fries, 2015). Thirdly, transcranial currents may have directly imposed excitability fluctuations at stimulation frequency on relevant visual cortex neurons, thus merely mimicking the functional outcome of endogenous low-frequency neuronal oscillations without the ‘entrainment’ of an already ongoing endogenously generated oscillation. While the current study cannot distinguish between these explanations, the latter explanation may be favorable as it requires the least assumptions. As shown in Fig. 2C, we observed maximal gamma power suppression around the falling flank of the TACS signal as derived from the stimulation output (i.e., the anodal-to-cathodal current transition in electrode Oz), whereas gamma power was most well preserved during the rising flank (i.e., the cathodal-to-anodal current transition in electrode Oz). Caution is advised when interpreting the phase of a TACS signal with respect to its phasic neuronal effects in a specific brain region, given the complex distribution of current direction across the heavily folded visual cortex (Neuling et al., 2012b; Rahman et al., 2013). Alpha TACS (or oscillatory TDC) work has often assumed the trough to be the less excitable phase (Notbohm and Herrmann, 2016; Sheldon and Mathewson, 2018), but even for spontaneous EEG alpha oscillations, the precise phase-excitability relationship has been inconclusive. While Mathewson et al. (2009) found reduced visual detection performance during the alpha trough of parietal electrodes, Dugue et al. (2011) found increased phosphene probability in response to transcranial magnetic stimulation (TMS) during the parietal alpha trough. When it comes to phase-amplitude coupling (PAC) between spontaneous oscillations of different frequencies, the power of the faster oscillation is not always maximal during the peak or trough of the slower oscillations, but can also be shifted towards its flanks e.g., commonly observed for slow sleep oscillation-spindle coupling (Mölle et al., 2011; Staresina et al., 2015) or theta-to-high-frequency-oscillations (Tort et al., 2013). Importantly, also for spontaneous alpha-gamma PAC in occipital cortex periods of maximal gamma power have been found to be shifted toward the descending phase of the alpha cycle in source level MEG data (Roux et al., 2013). Our findings are complemented by work from Helfrich et al. (2016), who reanalyzed an earlier TACS-EEG dataset (Helfrich et al., 2014a) and found increased cross-frequency coupling between spontaneous alpha oscillations (8–12 Hz band) and task-related gamma band activity during 10 Hz TACS compared to sham. They found gamma power during alpha troughs to be enhanced compared to alpha peaks, however, comparisons with rising and falling flanks were not reported. While these results support the idea that TACS at 10 Hz can facilitate alpha-band to gamma coupling, they did not resolve at that time whether gamma was entrained at the precise TACS frequency, and whether retinal entrainment may have been involved, two issues we explicitly addressed in this study.

4.2. TACS related gamma modulation is behaviorally relevant

Occipital TACS compared to retinal control TACS caused a small but significant impairment in visual detection performance, which can, importantly, not be attributed to retinal stimulation. Beyond that, subjects showing a stronger TACS related modulation of gamma power also showed stronger drops in visual accuracy. Thus, as predicted by the pulsed inhibition hypothesis, phasic suppression of stimulus-related gamma oscillations suppressed bottom-up neuronal processing and thus impaired perception. Importantly, the behavioral relevance of TACS-related neuronal activity modulations already shows that the observed neuronal effects cannot be attributed to residual artifacts.

4.3. TACS effects cannot be explained by subthreshold retinal entrainment or residual artifacts

One of the key challenges in studies using TACS is not only the current flow to unintended brain regions, but also to extracranial neuronal structures, such as the retina, which is particularly sensitive to stimulation; TACS easily excites the retina and induces retinal phosphenes, i.e., a sensation of flickering light at stimulation frequency (Kar and Krekelberg, 2012; Schutter, 2016). Importantly, even stimulation intensities below phosphene threshold may entrain visual cortex activity via the retino-thalamo-cortical pathway, explaining why even subconscious intermittent photic stimulation can produce cognitive effects (for a review see Schutter, 2016). To exclude this potential confound, we included a control montage (Fpz-Cz) with the frontal electrode even closer the eyes and matched the stimulation intensity on the retina for both montages by independently adjusting it to 80% of the subjects’ individual phosphene threshold. This ensured that (i) no retinal phosphenes were induced throughout the experiment, and (ii) the amount of effective current reaching the retina was comparable between montages, while only the occipital montage exerted a direct transcranial impact on the visual cortex. Indeed, we did not observe any effect of the retinal control TACS on either behavior, or on phase-amplitude coupling with gamma band power, or on the relationship between both. This demonstrates that the behaviorally relevant modulatory effects of occipital TACS observed in the current experiment are not explained by indirect retino-thalamo-cortical but rather by direct transcranial cortical stimulation.

A second challenge for TACS studies in combination with EEG or MEG are the stimulation artifacts that contaminate the recordings, particularly at the stimulation frequency but also at its harmonics, as well as at heartbeat- and respiration-related side bands (Noury et al., 2016; Noury and Siegel, 2018). While beamformer spatial filter techniques (like the one used in the current study) can considerably attenuate the artifact when correctly parameterized (Neuling et al., 2017), they may not be able to remove it completely due to its non-linearity (Noury et al., 2016) and mathematical constraints related to (un-)correlated sources (Mäkelä et al., 2017). In the current study, we deliberately circumvent most of these issues by focusing entirely on the effects of low-frequency TACS on high-frequency stimulus-induced gamma power. Importantly, we applied identical artifact removal procedures to all TACS montages, TACS frequencies, and created respective frequency-specific Sham conditions (see Methods for details). Moreover, TACS periods before and during visual stimulation (containing the same amount of residual TACS artifacts) were contrasted to determine the induced gamma response (see Methods for details), an approach for which exists consensus that it effectively
controls for residual TACS artifacts \cite{Neuling2017, Noury2018}. While we cannot exclude residual artifacts surviving beamforming, high-pass filtering, and FFT interpolation procedures, they would be present in the baseline and the period during visual stimulation. By subtracting the baseline from the visual stimulation period, we therefore largely corrected for residual TACS artifacts. Even if random dynamic fluctuations of artifacts due to e.g., heart-rate, breathing, and head movements may only allow an imperfect removal, they would mainly result in unystematic residual artifacts. While theoretically possible, we consider it very unlikely that the monotonously repeated visual stimuli in our task cause any systematic changes in respiration or heart-rate and thus in the side bands of the TACS frequency \cite{Neuling2016} that could explain our findings, especially since no effects on gamma power or behavior were observed for the retinal control montage. Moreover, a dedicated control analysis \cite{Kasten2018} did not reveal a visual stimulus-related modulation of TACS artifact amplitude in the gamma range nor a difference of this ratio between montages. However, for the subtraction approach to be successful, it may be important to first reduce TACS artifact amplitude considerably \cite{Kasten2018} (e.g., by spatial filtering band-stop filtering, and FFT interpolation) to prevent event-related oscillations to be masked by unystematic residual artifacts surviving the subtraction approach.

Another caveat deserves discussion: While matching the retinal control montage to achieve similar amounts of retinal stimulation effectively controlled for the possibility of subliminal retinal-thalamo-cortical entrainment, it inevitably resulted in larger stimulation intensities for the experimental than for the control montage \cite{Kasten2018}, and thus in larger residual TACS artifacts as well as stronger cutaneous stimulation. Unfortunately, an ineffective control montage that nonetheless produces identical TACS artifact amplitudes at the anatomical regions of interest as well as identical cutaneous stimulation is not possible to the best of our knowledge. But can our montage-specific findings be actually explained by larger residual TACS artifacts? We believe they cannot for the following reasons: (1) Beamformer solutions for all conditions were calculated on the same TACS-free sham trials only and did thus not bias signal extraction; (2) residual artifacts were first removed by absolute baseline subtraction, preventing baseline residual artifacts from biasing induced gamma power; (3) a roughly sinusoidal \cite{Bergmann2016} residual TACS artifact, which is neither asymmetric nor contains spikes at a particular phase, is unlikely to produce spurious phasic suppression of gamma power once per TACS cycle and confined to the frequency band of visually-induced gamma power; (4) occipital but not retinal control TACS showed significant PAC relative to respective montage- and frequency-specific phase-shifted surrogate data, controlling for unspecified effects of residual TACS artifacts on PAC; (5) if differences in stimulation intensity would explain our findings, its inter-individual variation should correlate with the amount of gamma suppression or PAC, which it does not \cite{Kasten2018}; (6) residual TACS artifacts cannot explain the observed correlation of PAC with behavior. The second question is whether our montage-specific findings can be explained by potentially stronger cutaneous stimulation and thus somatosensory input and tactile perception (and theoretically stronger distraction from the behavioral task)? Differences in somatosensory co-stimulation can never be ruled out entirely when comparing different electrode montages, as somatosensory stimulation not only depends on the stimulation voltage but also the distribution and sensitivity of receptors in the skin relative to the electrode locations \cite{Antal2016}. Unfortunately, the randomized intra-session design prevented a condition-specific assessment of perceived sensation without drawing their attention to the different trial types, but at least none of the participants did report any particularly unpleasant (and thus distracting) sensations in hindsight. In any case, a general performance decrease due to tactile distraction would not explain the observed phasic gamma power suppression or its correlation with behavior. Our control condition with reduced intensity can, however, not rule out entirely the possibility of a cross-modal entrainment of visually induced gamma power by (subliminal) somatosensory co-stimulation, which may (or may not) be stronger for the occipital montage. In summary, we are nonetheless confident that the reported findings cannot be better explained by residual TACS artifacts or sensory stimulation confounds.

4.4. General implications for TACS-MEG/EEG research

Given the increasing use of TACS and TDCS for the non-invasive modulation of neuronal activity and cognitive function on the one hand, and the recent criticism regarding replicability \cite{Horvath2014, Horvath2015}, effectiveness of commonly applied current intensities \cite{Lafon2017, Opitz2016, Voroslakos2018}, and confounds by peripheral \cite{Kar2012, Schutter2016} on the other hand, our findings have important implications reaching beyond the specific research question of the present study. Numerous studies \cite{Alekseichuk2016, Brittain2013, Cecere2015, Helfrich2014b, Joundi2012, Neuling2012a, Pogosyan2009} have provided compelling behavioral evidence that TACS does affect human brain function. However, due to the massive presence of TACS artifacts in EEG/MEG recordings \cite{Kasten2018}, direct assessment of a TACS-related phase-dependent modulation of neuronal activity at the stimulation frequency is inherently problematic \cite{Bergmann2016}. In contrast, by investigating TACS-phase-to-gamma-power-coupling, we could demonstrate that TACS is indeed able to rhythmically suppress visual stimulus-induced gamma oscillations in a behaviorally relevant manner, while ruling out retinal entrainment or residual artifacts as alternative explanation. This implicates that, if adequately controlled, TACS-MEG at common stimulation intensities is a powerful tool to study and manipulate oscillatory brain activity and behavioral performance non-invasively in humans.

Conflicts of interest

The authors declare no conflict of interests.

Acknowledgements

This work was supported by The Netherlands Organisation for Scientific Research, VICI Grant 453-09-002, ALW Open Competition Grant 822-02-011.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.neuroimage.2018.09.047.

References

Alekseichuk, I., Turi, Z., Amador de Lara, G., Antal, A., Paulus, W., 2016. Spatial working memory in humans depends on theta and high gamma synchronization in the prefrontal cortex. Curr. Biol. 26, 1513–1521.

Ali, M.M., Sellers, K.K., Frohlich, F., 2013. Transcranial alternating current stimulation modulates large-scale cortical network activity by network resonance. J. Neurosci. 33, 11262–11275.

Antal, A., Herrmann, C.S., 2016. Transcranial alternating current and random noise stimulation: possible mechanisms. Neural Plast. 2016, 36146907.

Bastos, A.M., Vezoli, J., Bosch, C.A., Schoffelen, J.M., Oostenveld, R., Dowdall, J.R., de Weerd, P., Kennedy, H., Fries, P., 2015. Visual areas exert feedforward and feedback influences through distinct frequency channels. Neuron 85, 390–401.

Bergmann, T.O., Karabanov, A., Hartwigsen, G., Thielsecker, A., Siebner, H.R., 2016. Combining non-invasive transcranial brain stimulation with neuroimaging and electrophysiology: current approaches and future perspectives. Neuroimage 140, 4–19.

Brittain, J.S., Probert-Smith, P., Aziz, T.Z., Brown, P., 2013. Tremor suppression by rhythmically transcranial current stimulation. Curr. Biol. 23, 436–440.

Cecere, R., Rees, G., Romei, V., 2015. Individual differences in alpha frequency drive crossmodal illusory perception. Curr. Biol. 25, 231–235.
