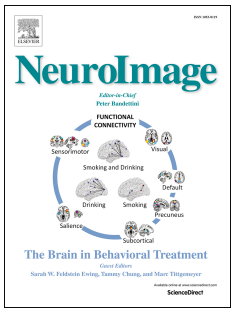
Accepted Manuscript



Noninvasive stimulation of the ventromedial prefrontal cortex modulates emotional face processing

Constantin Winker, Maimu A. Rehbein, Dean Sabatinelli, Mira Dohn, Julius Maitzen, Carsten H. Wolters, Volker Arolt, Markus Junghofer

PII: S1053-8119(18)30277-5

DOI: 10.1016/j.neuroimage.2018.03.067

Reference: YNIMG 14836

To appear in: *NeuroImage*

Received Date: 15 September 2017

Revised Date: 20 February 2018

Accepted Date: 28 March 2018

Please cite this article as: Winker, C., Rehbein, M.A., Sabatinelli, D., Dohn, M., Maitzen, J., Wolters, C.H., Arolt, V., Junghofer, M., Noninvasive stimulation of the ventromedial prefrontal cortex modulates emotional face processing, *NeuroImage* (2018), doi: 10.1016/j.neuroimage.2018.03.067.

Noninvasive stimulation of the ventromedial prefrontal cortex modulates emotional face processing

Constantin Winker[a,b], Maimu A. Rehbein[a,b], Dean Sabatinelli[c], Mira Dohn[a], Julius Maitzen[a], Carsten H. Wolters[a,b], Volker Arolt[b,d] & Markus Junghofer[a,b,*]

[a]Institute for Biomagnetism and Biosignalanalysis, University of Muenster, Germany

[b]Otto Creutzfeldt Center for Cognitive and Behavioral Neuroscience, University of Muenster, Germany

[c]Department of Psychology and BioImaging Research Center, University of Georgia, USA

[d]Department of Psychiatry, University of Muenster, Germany

*Corresponding author at: Institute for Biomagnetism and Biosignalanalysis, University of Muenster, Malmedyweg 15, 48149 Muenster, Germany.

Keywords: Emotion, Brain Stimulation, tDCS, MEG, EEG, Faces

Abstract:

The ventromedial prefrontal cortex (vmPFC) is associated with emotional states that can be characterized as positive affect. Moreover, a variety of psychiatric disorders that are associated with disturbed reactions toward reward- or safety-signaling stimuli reveal functional or structural anomalies within this area. Thus, neuromodulation of this region via transcranial direct current stimulation (tDCS) offers an attractive opportunity to noninvasively influence pleasant emotional and reward processing. Recent experiments revealed hemodynamic and electrophysiological evidence for valence specific modulations of emotional scene processing after excitatory and inhibitory tDCS of the vmPFC. Here, we identified that tDCS modulation of vmPFC during emotional face processing results in effects convergent with scene processing, in that excitatory tDCS increased neural reactivity during happy compared to fearful face perception, whereas inhibitory stimulation led to a converse effect. In addition, behavioral data (affect identification of ambiguous expressive faces) revealed a bias toward preferential processing of happy compared to fearful faces after excitatory compared to after inhibitory stimulation. These results further support the vmPFC as an appropriate target for noninvasive neuromodulation of an appetitive processing network in patients suffering from disturbed cognition of reward- and safety-signaling stimuli. It should however be noted that electrophysiological pre-tDCS differences at earlier time intervals of emotional face and scene processing appeared amplified by tDCS, which remains to be investigated.

1. Introduction

Perception and evaluation of emotional stimuli from our environment are vital to ensure survival and avoid hazardous situations. Beyond this fundamental drive to survive, healthy individuals possess an inherent, natural striving to maximize rewarding situations while minimizing personal risks and discomfort. Therefore, it is important to the perceiver to not only recognize salient stimuli with underlying emotional relevance, but also to categorize such stimuli as containing a pleasant or unpleasant valence. Only a few brain regions are considered consistently valence specific. As one of those regions, the ventromedial prefrontal cortex (vmPFC) has been shown to be particularly sensitive to changes in pleasant valence, and many studies point to a general effect independent of stimulus type, revealing enhanced vmPFC activation for, e.g., rewarding stimuli (Hajcak Proudfit, 2015; Knutson et al., 2003), pleasant visual stimuli (Junghofer et al., 2017; Sabatinelli et al., 2007), or imagery of pleasant events (Costa et al., 2010). In a similar but somewhat broader sense, increasing vmPFC activation during reduction of pain perception (Atlas et al., 2010) or during perception of safety signals in the course of fear extinction (Milad et al., 2007) can also be viewed as pleasant valence processing. However, there are also indications for vmPFC effects that appear in direct opposition to a specificity for pleasant valence. In their review, Myers-Schulz and Koenigs (2012) explain this apparent discordance by suggesting certain sub-areas of the vmPFC, only one of which might be connected to pleasant valence. This subarea, the anterior vmPFC/perigenual anterior cingulate region, depicts increased activation specifically during pleasant valence processing and shows growing activity with successful treatment of major depressive disorder (MDD). A subarea in the posterior vmPFC/subgenual cingulate cortex on the contrary is found to gain stronger activation in response to unpleasant stimuli and reveals relatively increased activity in MDD patients (see Myers-Schulz & Koenigs, 2012).

In this context, neuromodulation like subthreshold transcranial direct current stimulation (tDCS) offers an attractive opportunity to probe vmPFC function in general emotional and pleasant stimulus processing. As tDCS allows a noninvasive cortical excitation or inhibition of neuron populations (see Paulus, 2014), it should be possible to directly modulate vmPFC activation and to assess vmPFC modulation effects on emotional processing. In fact, there are studies reporting effects of tDCS of the vmPFC on emotional processing in the predicted direction, i.e., consistent with a specificity for pleasant valence: Chib et al. (2013) for instance showed that excitatory vmPFC stimulation led to an increase in face attractiveness ratings as well as stronger ventral midbrain activation. Two further studies revealed stimulation effects on fear extinction processes: Mungee et al. (2014) reported increased skin conductance responses (SCR) after inhibitory stimulation of the vmPFC, reflecting enhanced fear or reduced safety learning, while opposite effects on SCRs were shown for an excitatory

stimulation paradigm (Van't Wout et al., 2016)[1]. Taken together, the frequently reported valence specificity of the vmPFC and successful stimulation effects as shown by different research groups suggest that an excitation of this region by noninvasive brain stimulation, especially tDCS, could enhance neural processing of pleasant relative to unpleasant stimuli. An initial set of experiments in our lab further supports this hypothesis (Junghofer et al., 2017). In two independent experiments we investigated emotional scene processing after excitatory (anodal) and inhibitory (cathodal) tDCS of the anterior vmPFC, using functional magnetic resonance imaging (fMRI) and magnetoencephalography (MEG). Excitatory stimulation led to increases of neural activation specifically for pleasant scenes compared to unpleasant scenes in a distributed brain network including regions previously linked to emotional scene perception (Sabatinelli et al., 2011). Moreover, the opposite hemodynamic and electrophysiological activation patterns were shown after inhibitory stimulation, such that greater activation was evoked during unpleasant in comparison to pleasant scene perception. Magnetoencephalographic correlates of these valence specific brain stimulation effects covered early (<100 ms), mid-latency (100-300 ms) and late (300-600 ms) processing stages and were found at occipital, temporal, and occipito-temporal areas of the ventral visual processing stream as well as prefrontal cortex (PFC) regions. Timing and location of these clusters converged with models of salience detection in human PFC modulating scene processing in visual cortex regions (e.g., Freese & Amaral, 2006; Pessoa & Adolphs, 2010; Vuilleumier, 2005). In fact, salient stimuli which activate the appetitive or defensive system for preparation of approach or avoidance behavior, receive enhanced attention compared to irrelevant stimuli, and this leads to amplified perceptual processing (Lang et al., 1997). Electrophysiological (EEG/MEG) correlates at early, mid-latency, and late time intervals reflecting enhanced sensory processing of emotional visual stimuli are rather similar across different visual stimulus types, such as emotional scenes (e.g., Schupp et al., 2006), emotional faces (e.g., Morel at al., 2009; Rellecke et al., 2013; Schupp et al., 2004), emotional words (e.g., Kissler et al., 2007), or emotional gestures (Flaisch et al., 2009).

Based on the hypothesis that activity in the anterior vmPFC is associated with emotion-related states characterized as pleasant (Myers-Schulz & Koenigs, 2012), it could be assumed that excitatory tDCS of the vmPFC might evoke a positivity bias for emotional stimuli in general. If this is the case, the same modulations of valence biases as found for emotional scene processing after vmPFC-tDCS (Junghofer et al., 2017) should generalize to other stimulus categories such as emotional faces, words, or gestures. For a first approach

[1] It should be noted that these studies did not apply quasi reference free stimulation paradigms. Mungee et al. (2014) and Chib et al. (2013) placed the reference electrode above the dorsolateral PFC und thus simultaneously and inversely stimulated a main hub of top-down emotion regulation (e.g., Ochsner & Gross, 2005) while Van't Wout et al. (2016) used the right mastoid as reference, which is in direct vicinity to visual (occipito-temporal) brain regions. Hence, this makes interpretation of the underlying effects more complex due to two simultaneously and inversely stimulated regions.

to test a generalization of vmPFC-tDCS effects we here employed emotionally expressive faces, as these have been investigated in a wide range of emotional neuroscience studies (Adolphs, 2002; Sabatinelli et al., 2011; Vuilleumier & Pourtois, 2007). Happy and fearful faces generally predict reward or punishment, safety or danger, and are often associated with symptomatology of emotional disorders (e.g., Foti et al., 2010; Moser et al., 2008; Opel et al., 2017). Our initial experiments testing the modulatory impact of tDCS on vmPFC (Junghofer et al., 2017) focused on neural effects of modulation and employed an experimental design and evocative scene stimuli that are less amenable to the detection of behavioral modulation effects. To assess potential behavioral effects of vmPFC stimulation, we here applied a task in which we measured categorization of ambiguous face stimuli morphed between happy and fearful in expression (face-morph task). Furthermore, to investigate behavioral attention modulation effects by vmPFC-tDCS, we conducted a dot-probe task using prototypical happy and fearful faces as cues.

In the face-morph task, we expected faces with ambiguous expressions to be categorized more often as happy than fearful (positivity bias) after excitatory vmPFC-tDCS and to be categorized more often as fearful than happy (negativity bias) after inhibitory stimulation. With regard to the dot-probe task, we predicted an increased orienting toward happy faces and away from fearful faces (positivity bias) after excitatory vmPFC-tDCS and increased orienting toward fearful faces and away from happy faces (negativity bias) after inhibitory vmPFC-tDCS. For the magnetoencephalographic correlates, we expected the above described activation patterns in early, mid-latency, and late time intervals within frontal and occipital, temporal, and occipito-temporal regions, in the same direction as those reported for emotional scenes (Junghofer et al., 2017), with excitatory vmPFC-tDCS leading to increased activation for happy compared to fearful faces and inhibitory vmPFC-tDCS leading to reduced activation for happy compared to fearful faces. An additional conjoint analysis across the MEG data from the current study (tDCS-vmPFC effects on emotional face perception) and the data from the previous study (tDCS-vmPFC effects on emotional scene perception) was performed to statistically validate generalization across these two commonly used emotional media and to identify potential media specific tDCS-vmPFC effects.

2. Methods

2.1. Participants

Forty healthy students (20 female) of the University of Muenster gave written informed consent and participated in the study, which had been approved by the University's Human Subjects Review Board. As compensation every participant received 50 €. Before the

experimental session, participants completed the Beck Depression Inventory II (BDI-II; Beck et al., 1996) and the State Trait Anxiety Inventory (STAI; Laux et al., 1981) as well as the Symptom Check List (SCL-90-S; Franke, 2014) at the end of session (see Table 1).

2.2. tDCS

A DC Stimulator Plus (NeuroConn) applied a constant current of 1.5 mA for 10 min (0.9 C overall charge) through a pair of electrodes covered in saline-soaked sponges during each of two stimulation runs. Finite element method (FEM) based forward modeling of tDCS currents (Wagner et al., 2014) with a target of maximal unipolar stimulation of the anterior vmPFC and minimal stimulation of other brain areas resulted in a forehead-chin montage with a 3 x 3 cm forehead electrode located at mid-distance between 10-20 electrode positions Fz and Fp serving as stimulating component and a 5 x 5 cm electrode under the chin as an extracephalic reference (Fig. 1A; Junghofer et al., 2017). This kind of stimulation circumvents the inherent reference problem of tDCS (i.e., simultaneous inhibitory neuronal stimulation under cathodal and excitatory stimulation under anodal electrode) and can thus be termed ‘quasi’ reference free. Stimulation parameters (strength, duration, electrode size) were the same as in preceding studies to foster comparability. During excitatory or inhibitory stimulation, the forehead electrode was used as anode or cathode, respectively. The order of stimulation (excitatory-inhibitory or inhibitory-excitatory) was balanced across participants, who were blind to the stimulation conditions.

2.3. Stimuli

Stimuli consisted of photographs of human faces of different individuals with happy, fearful, and neutral expressions, taken from the Karolinska Directed Emotional Faces Database (Lundqvist et al., 1998), NimStim Set of Facial Expressions (Tottenham et al., 2009), and Radboud Faces Database (Langner et al., 2010). Stimuli were transformed into grayscale and overlaid on a gray background using Photoshop (Adobe Systems).

For the MEG measurement and each behavioral task, different subsets of faces were used. The presentation during MEG recording featured 64 stimuli, with 32 happy faces and 32 fearful faces of 64 different individuals. The face-morph task was assessed using four stimulus pairs featuring a happy and a fearful expression per each of four individuals. In the dot-probe task, face pairs of 20 individuals were used, showing each individual with happy and neutral or fearful and neutral expression per trial (60 different stimuli overall). Across all subsets of stimuli, face stimulus gender was equally balanced. All stimuli had a resolution of 1024 x 768 pixels and did not differ significantly in luminance and contrast between conditions. More detailed descriptions about the respective stimuli can be found in the sections regarding the conducted tasks.

2.4. Procedure

Each participant's head shape was first registered using a 3Space Fastrak (Polhemus), after which they were seated in the MEG chamber for an acclimation series, in which all stimuli were presented twice. Immediately following, participants were instructed to remain still during the event-related MEG measurement, in which the stimuli were presented again three times. For all MEG runs, order of stimulus presentation was pseudorandomized with an average duration time of 7 min per run. Subsequent to this measurement, the first of two tDCS runs was prepared and conducted. Following this, the first post-tDCS MEG measurement was assessed. Then participants accomplished the face-morph (8 min duration) and dot-probe tasks (15 min duration), followed by a break (60 min duration), resulting in an interval of approximately 90 min between the first and second tDCS session to allow for dissipation of modulatory effects (Nitsche et al., 2005). After the second tDCS session, participants were seated again in the MEG scanner for the second post-tDCS MEG measurement, followed by both behavioral tasks (see Fig. 1B for an overview of the procedure). Order of the behavioral tasks was balanced across participants with half of the participants starting both behavioral test sessions with the dot-probe task and the other half with the face-morph task.

2.5. Behavioral tests

Both behavioral tests were conducted in computerized form. A DirectIN PCB keyboard (Empirisoft) was used to keep temporal jittering by the reaction time input device at a minimum (<1 ms). Stimulus presentation was accomplished with an 85 Hz G90fB CRT monitor (ViewSonic).

2.5.1. Face-morph task

We conducted the face-morph task (adapted from McMahon & Leopold, 2012) to investigate modulation of emotional categorization of ambiguous facial expressions induced by tDCS of the vmPFC. Ambiguous facial expressions were created by morphing images that depicted the same individual with happy or fearful expressions using PsychoMorph software (users.aber.ac.uk/bpt/jpsychomorph; Tiddeman & Perrett, 2002). Morphing resulted in a face expression continuum ranging from a purely happy (100% happy, 0% fearful) to a purely fearful expression (0% happy, 100% fearful) in 101 steps. From this continuum, participants viewed the morph step with maximal ambiguity of categorization – also referred to as perceptual midpoint (PM) – and six neighboring morph steps corresponding to PM ±8%, PM ±16% and PM ±24% (i.e., seven total morph stimuli for each of four pairs; Fig. 2A), and were asked to categorize each face stimulus as happy or fearful by button press. The PM of each stimulus pair was assessed in a pre-test with eight participants not included in the MEG

study group. The PM, as estimated in the pre-test, corresponded to the point in which morphs were judged 50% of the time as fearful and 50% as happy, such that the six neighboring steps would allow for an optimal Weibull function fit of individual reactions.

After two preparation blocks to familiarize the participant with the procedure, the task block began. During the task block, morphed stimuli were presented in 112 trials with repetition of all seven morph steps 16 times. Each morph step would be presented once, before being repeated within a new random order. Participants were asked to determine by button press as quickly and as accurately as possible whether they perceived the presented face as either happy or fearful. A single trial consisted of the presentation of a fixation cross for 500 ms followed by a morph stimulus for up to 2000 ms duration, terminated by the button press. Trials with no button press during time of presentation counted as 'miss'. The procedure as described above (two preparation blocks, one task block) was repeated for all four face pairs. The morphing task was written and executed with the Psychophysics Toolbox software package (Brainard, 1997) in MATLAB (The MathWorks).

Data were used to fit the Weibull cumulative distribution function ($F(x) = 1 - e^{-(\alpha x)^{\beta}}$) with parameters α (scale parameter) and β (shape parameter). This fit was calculated across all face pairs within every participant. In addition, a parameter describing the goodness of fit of the function to the data was calculated. Analysis comprised *t*-tests for the variables Modulated PM (mPM), the shape parameter β, and the goodness of fit between both post-stimulation runs. Preceding the statistical analysis, all error trials as well as all trials with reaction times < 100 ms or > 3000 ms were excluded. This interval was chosen to allow for a later comparison with clinical samples which may show deficits in psychomotor speed. In succession, mean reaction times were calculated for each participant individually to exclude outliers, i.e., trials with reaction times > 3 x standard deviation. Identified by QQ-plots and box plots as outlier, one participant had to be excluded. Further, for the dependent variable Reaction Time an rmANOVA with factors Morph Step (H 24%, H 16%, H 8%, 0% (mPM), F 8%, F 16%, F 24%; H: Happy, F: Fearful) and Stimulation (Excitatory, Inhibitory) was run. Participants' choice of category (Happy, Fearful) was not included as a factor in the rmANOVA due to too many missing cases, as most participants categorized faces with higher amounts of happiness persistently as happy and vice versa for faces with higher amounts of fear.

2.5.2. Dot-probe task

The dot-probe task (adapted from MacLeod et al., 1986) was conducted to investigate tDCS induced changes in attentional orienting toward or away from emotional facial stimuli. In this task, participants were asked to identify the location of a dot, which was presented on the left or right side of a monitor and was preceded by face cue pairs with an emotional face

cue presented either at the dot-location (congruent) or at the opposite location (incongruent). Accelarated or slowed reaction times are inferred as representing attention to the chosen stimulus of focus. This relation can be simplified by the formula of the attentional bias (MacLeod & Mathews, 1988), which describes the difference between reaction times after incongruent and congruent cues. Using face pairs with emotional and neutral expressions as cues, it is possible to analyze the impact of emotional relative to neutral cues at dot-congruent and dot-incongruent positions.

In preparation for this task, participants were informed that they would see a centered white fixation cross followed by a white dot either on the left or on the right side of the monitor. Both index fingers rested on a left and right button of the keyboard and participants were asked to press the button corresponding to the correct side as quickly as possible when a dot appeared. They were also told that before the appearance of the dot, two photographs would be presented for a short duration. These photographs contained an emotional (happy or fearful) and a neutral face of the same individual and were presented simultaneously at the location where the dot would later appear.

A trial consisted of a presentation of the fixation cross for 1000 ms, followed by the stimulus pair for 1000 ms. Subsequent to the stimulus pair, the dot was presented until a button press was recorded. Each participant completed 160 trials with 80 pairs of happy and neutral, and 80 pairs of fearful and neutral facial expressions. During half of the trials (40 per group) the dot appeared at the position of a previously located emotional stimulus (congruent), while during the remaining trials the dot appeared on the opposing side (incongruent). Side of appearance for congruent and incongruent trials was balanced across trials. The dot-probe task-script was written and run with Presentation software (Neurobehavioral Systems).

Exclusion of errors and outliers was similar to the face-morph task leading to a drop out of one participant. The attentional bias was calculated as follows: $\frac{(RdLe - RdRe) + (LdRe - LdLe)}{2}$ with R = right, L = left, d = dot, e = emotional face. A positive value thus indicates a faster reaction for congruent trials relative to incongruent trials, whereas a negative value indicates faster response for incongruent trials relative to congruent trials. An rmANOVA with dependent variable Attentional Bias and factors Valence (Happy, Fearful) x Stimulation (Excitatory, Inhibitory) was then calculated. Statistical analysis of all behavioral data was conducted with SPSS (IBM).

2.6. MEG measurement and analysis

Event-related magnetic fields were measured during 192 trials including two repetitions of each of the 64 face stimuli. A trial consisted of a face presentation for 600 ms followed by a

jittered interstimulus interval (ISI) of 1000 – 2000 ms in which a gray background with a white fixation cross was displayed. Stimuli were presented in pseudorandomized order (controlled transitions of emotional categories) and did not repeat until the complete set of 64 individual face stimuli had been presented. For each measurement block (pre-tDCS, post-tDCS I, post-tDCS II), a different pseudorandomized order of stimulus presentation was used. Visual angle of presentation was 8.4° vertically and varied horizontally due to individual stimulus head shapes.

MEG measurement was conducted with a 275 whole-head sensor system (CTF Systems) with first-order axial gradiometers. To measure head movement during MEG recording, landmark coils were positioned on the nasion and in each earlobe. Head movement was defined as maximal deviation from the head's position at the beginning of an MEG run. Head movements of participants did not exceed 5 mm per run. MEG data was recorded continuously with a sampling rate of 600 Hz spanning a frequency range of 0 – 150 Hz to guarantee anti-aliasing filtering. Afterwards, data was sampled down to 300 Hz, high-pass filtered using a zero-phase Butterworth second-order filter of 0.1 Hz and low-pass filtered applying a fourth-order 48 Hz cutoff.

Sampled data was then split into single epochs of 800 ms length, ranging from -200 ms before to 600 ms after stimulus onset. Each epoch was baseline-adjusted by subtracting the mean activity of the 150 ms period prior to stimulus onset. All epochs per condition were then averaged. Underlying neural sources were estimated using the L2-Minimum-Norm estimation (L2-MNE; Hämäläinen & Ilmoniemi, 1994). A spherical shell with evenly distributed 2 x 350 dipoles served as a source model. Topographies of the L2-MNE were established with a Tikhonov regularization parameter of $k = 0.1$.

rmANOVAs with within-factors Stimulation (Excitatory, Inhibitory) and Valence (Happy, Fearful) were calculated for each estimated neural source and each time point. We searched for clusters showing a significant interaction effect of Stimulation x Valence. A nonparametric statistical testing procedure similar to the cluster mass-test used for analysis of fMRI data was applied that included correction for multiple comparisons (see Maris & Oostenveld, 2007). Corresponding to the intervals of analysis used by Junghofer et al. (2017) and described above, we defined early (0-100 ms), early to mid-latency (100-200 ms), mid-latency to late latency (200-300 ms) and late latency (300-600 ms) time intervals for separate statistical analyses.

Within each time interval, only estimated sources were considered for analysis that showed a significant interaction of Stimulation x Valence surpassing p-value < .05 (sensor-level criterion). Resulting F-values of the underlying sources forming a spatio-temporal cluster were then summed to constitute the cluster mass. Cluster masses were evaluated

against a distribution of 1,000 random permutations of the same data set. When the cluster mass was higher than the critical cluster mass of this distribution corresponding to a *p*-value = .05 (cluster-level criterion), the cluster was considered significant. In the case of clusters reaching an interval border of the predefined time intervals, analysis was repeated for an extended interval in steps of 50 ms, and collapsed across the original and preceding or subsequent interval, respectively. By this means it was possible to assess if the found cluster actually began or ended with the interval or if it was temporally extended. In case of new clusters appearing due to the extension of an interval, these findings were not considered.

Due to our focus on the hypothesized interaction of pleasant vs unpleasant valence modulated by excitatory and inhibitory tDCS in opposite ways, only post-stimulation runs were analyzed, initially. To further test for consistencies and differences of this relevant interaction across different stimulus types (pleasant and unpleasant scenes (Junghofer et al., 2017) and pleasant/happy and unpleasant/fearful faces), an additional three-way mixed-model ANOVA with within-factors Stimulation (Excitatory, Inhibitory) and Valence (Pleasant, Unpleasant) and a between-factor Stimulus Type (Faces, Scenes) was conducted across both study samples. Age and gender distribution of both subject groups were comparable (see Table 1) and relevant characteristics of the passive viewing task (ISI, number of trials, trial duration) were identical (see Supplementary Table 1).

Preprocessing and analysis of MEG data was conducted using the MATLAB based software EMEGS (emegs.org; Peyk et al., 2011).

3. Results

3.1. Behavioral tests

3.1.1. Face-morph task

3.1.1.1. *Modulated perceptual midpoint (mPM)*

A two-tailed *t*-test revealed a significant difference in mPM location after excitatory versus after inhibitory stimulation ($t(38) = 2.05$, $p = .047$) with the mPM displaced toward the fearful pole after excitatory stimulation (Fig. 2B). Thus, after excitatory compared to inhibitory stimulation, perception of ambiguous faces showed a positivity bias, as a lesser amount of happiness in the faces was necessary to lead to a categorization as happy, and/or a higher percentage of fearful expression in a morph stimulus was necessary to lead to a categorization as fearful.

3.1.1.2. *Shape* parameter *β and goodness of fit*

As the data for the shape parameter *β* and for the goodness of fit-parameter significantly deviated from normality for both conditions, Excitatory and Inhibitory, a nonparametric Wilcoxon-test was calculated to test for differences. For both parameters no differences were found yielding nonsignificant results for *β* (Z = -0.31, p = .757) and the goodness of fit (Z = -1.14, p = .253). Furthermore, the number of misses in this task were low in both stimulation conditions (Excitatory: *Mean*: 0.54%, *SD*: 0.78, *range*: 0 – 3.35%; Inhibitory: *Mean*: 0.3%, *SD*: 0.34, *range*: 0 – 1.34%).

3.1.1.3. *Reaction time*

For the analysis of Reaction Time a significant main effect for the factor Morph Step (F(6,234) = 51.24, p < .001) was observed (Fig. 2C). Within-subjects contrasts revealed the expected inverse quadratic contrast of decreasing reaction times with decreasing valence ambiguity, and longest reaction times at the mPM (quadratic: F(1,39) = 104.05, p < .001) but also a linear contrast (F(1,39) = 15.24, p < .001) due to generally faster reactions evoked by fearful compared to happy faces as well as higher order effects (cubic: F(1,39) = 11.68, p = .001; quartic: F(1,39) = 15.15, p < .001).

Although within-subjects effects for Stimulation and the interaction of Stimulation x Morph Step were nonsignificant (F(1,39) = 0.04, p = .849; F(6,234) = 1.35, p = .237), a trend-level cubic effect for the interaction Stimulation x Morph Step occurred (F(1,39) = 3.92, p = .055) (see Fig. 2C). Visual inspection of the data led to the post hoc hypothesis that the tDCS might have had a stronger impact on ambiguous stimuli while effects on quite distinct stimuli were, presumably due to floor effects, reduced. Testing this possibility, another rmANOVA was conducted without the most distinct morph steps (H 24%, F 24%), which were rated as happy or fearful respectively without any variance in more than half of all participants. In this analysis, the cubic within-subjects contrast for interaction Stimulation x Morph Step reached significance (F(1,39) = 5.39, p = .026) qualified by relatively faster responses for happier faces and slower responses for more fearful faces after excitatory stimulation and an inverted pattern after inhibitory stimulation.

3.1.2. Dot-probe task

Analysis of the dot-probe task data showed no significant effects for Stimulation (F(1,38) = 2.41, p = .129), Valence (F(1,38) = 0.38, p = .544), or the interaction of Stimulation x Valence (F(1,38) = 0.42, p = .521). Average errors were 1.34% (*SD*: 1.7, *range*: 0 – 8.13%) and 1.44% (*SD*: 2.16, *range*: 0 – 11.25%) for conditions Excitatory and Inhibitory, respectively.

3.2. MEG

3.2.1. Interaction Stimulation x Valence: Faces

In two of the predefined time intervals, spatio-temporal clusters for the predicted two-way interaction Stimulation x Valence were significant. A widely distributed and sustained cluster (357-507 ms, Cluster Mass (CM): 5252.9, Critical Cluster Mass (CCM): 1507.5) appeared in a late time interval spanning from occipital cortex over parietal, temporal, and frontal areas covering lateral dorsal and ventral regions revealing a significant interaction consistent with our hypothesis (Fig. 3A). Specifically, happy faces evoked increased activation in comparison to fearful faces after excitatory stimulation, whereas fearful faces evoked more activation compared to happy faces after inhibitory stimulation.

A second, more focal and short-lived cluster (193-240 ms, CM: 802.9, CCM: 674) in the early to mid-latency time interval occurred over right temporal regions and revealed relatively increased processing of fearful compared to happy faces after excitatory stimulation, while inhibitory stimulation led to an opposite activation pattern (Fig. 3B).

3.2.2. Interaction Stimulation x Valence: Faces and Scenes

The cluster permutation calculated for the combined studies (happy, fearful faces and pleasant, unpleasant scenes) again revealed a widely distributed and sustained spatio-temporal cluster for the interaction Stimulation x Valence (330-583 ms, CM: 7364, CCM: 1704) covering right occipito-temporal as well as frontal areas (Fig. 4A). Post hoc two-way ANOVAs of this cluster with factors Stimulation and Valence split up for Stimulus Type revealed significantly increased processing of both happy faces ($F(1,38) = 21.44$, $p < .001$) and pleasant scenes ($F(1,28) = 26.99$, $p < .001$) after excitatory compared to after inhibitory stimulation (Fig. 4B). There were no clusters with the opposite activation pattern.

Three clusters revealed significant three-way interactions (Stimulation x Valence x Stimulus Type): A first short-lived cluster appeared at a very early latency (37-77 ms, CM: 436, CCM: 270.5) at a medial prefrontal region with an activation pattern consistent with our hypotheses (i.e., positivity bias after excitatory tDCS and negativity bias after inhibitory tDCS) for facial stimuli and a reversed pattern (i.e., negativity bias after excitatory tDCS and positivity bias after inhibitory tDCS) for emotional scenes (Fig. 5A).

A second cluster was found in a sustained interval (167-320 ms, CM: 2244.6, CCM: 1154.5) at right inferior temporal regions (Fig. 5B). Convergent to the right temporal cluster found for Faces only (Fig. 3B), this temporally and spatially overlapping cluster showed an inverted stimulation pattern for emotional faces while neural activity evoked by emotional scenes revealed the predicted direction of stimulation effects.

Finally, a third cluster appeared during mid-latency (210-287 ms, CM: 654.9, CCM: 652), at right occipital cortex revealing a stimulation pattern for Scenes consistent with our hypotheses, while for Faces no differences between categories could be observed (Fig. 5C).

In this third and the second cluster, the direction of effects was inverted with respect to the first cluster (i.e., positivity bias for faces but negativity bias for scenes). For all three-way ANOVAs post hoc two-way ANOVAs were conducted split up for factor Stimulus Type (Supplementary Table 2).

3.2.3. Interaction Valence x Stimulus Type: Pre-tDCS

Based on the partly diverging stimulation effects on emotional faces and scenes, we analyzed post hoc whether there were any significant differences in processing of pleasant and unpleasant stimuli between both studies independent of tDCS. Therefore, we searched for clusters with a significant two-way interaction of Valence x Stimulus Type for the pre-tDCS MEG runs. As such we found an extended and sustained cluster (130-460 ms, CM: 21581.4, CCM: 2582.5) with an increased activation for pleasant compared to unpleasant scenes but lower activation for happy in comparison to fearful faces (Fig. 6). This result indicated that emotional faces and scenes show divergent modulation regarding valence during mid-latency and beginning late latency.

3.2.4. Interaction with factor Hemisphere

All clusters, which were situated in one hemisphere, were checked for lateralized effects post hoc. For this reason, a region of interest (ROI) containing the dipole positions contralateral to the original cluster was defined, and the additional factor Hemisphere (Left, Right) was included in the analysis run now across the original and the contralateral cluster. For clusters with slight hemispheric overlap the overlapping dipole positions were excluded beforehand. This procedure was applied to all clusters described above, except for the bilateral cluster at prefrontal cortex (Fig. 5A) and the cluster showing a large prefrontal, bilateral overlap during pre-tDCS assessment (Fig. 6).

The analysis of the event-related data with faces only yielded significant effects for the interaction Stimulation x Valence x Hemisphere during the mid-latency (193-240 ms; $F(1,38) = 7.64$, $p = .009$) and the late latency cluster (357-507 ms; $F(1,38) = 7.14$, $p = .011$). For the right temporal cluster, which had shown a significant three-way interaction of Stimulation x Valence x Stimulus Type during the mid-latency time window (167-320 ms), a significant four-way interaction of Stimulation x Valence x Stimulus Type x Hemisphere emerged as well ($F(1,66) = 8.51$, $p = .005$), whereas for the right occipital cluster (210-287 ms) the four-way interaction did not reach significance ($F(1,66) = 1.34$, $p = .251$). The post hoc-analysis of the cluster during late latency (330-583 ms) showing a significant effect for the interaction Stimulation x Valence across both stimulus types revealed no significant three-way interaction Stimulation x Valence x Hemisphere ($F(1,66) = 0.64$, $p = .428$). In summary, a right lateralization can be assumed for the effect of Stimulation x Valence in temporal cortex

(193-240 ms) during mid-latency and occipital, temporal, parietal, and frontal areas during late latency (357-507 ms) for the processing of emotional faces. Furthermore, for interaction Stimulation x Valence x Stimulus Type the effect within right temporal regions was hemisphere specific, whereas the right occipital cluster did not show a right lateralization for the same interaction. Analysis of lateralization did not reach significance for the conjoint analysis across studies of type Stimulation x Valence in right occipital, temporal, parietal, and frontal areas.

4. Discussion

The aim of this study was to investigate if modulation of emotional scene perception induced by tDCS of vmPFC (Junghofer et al., 2017) would be also demonstrable in neural and behavioral measures of sociocommunicative face perception. Thus, we presented happy and fearful faces in a passive viewing task during MEG measurement and assessed behavioral data testing for expected shifts of attentional and interpretational biases during perception of happy and fearful faces as well as ambiguous stimuli containing both emotional expressions.

In the MEG analysis of estimated sources, we found a long-lasting and widely distributed spatio-temporal cluster within the late time interval (357-507 ms) spanning across right lateralized occipital, temporal, parietal, and frontal areas with an interaction for Valence x Stimulation (Fig. 3A), with increased activation during happy faces compared to fearful faces after excitatory stimulation and the reverse effect after inhibitory stimulation. This cluster corresponds in time and location with recent findings during emotional scene perception (Junghofer et al., 2017). The combined analysis across both stimulus types (Faces, Scenes; Fig. 4) revealed the same cluster interaction across an extended interval (330-583 ms), further supporting our hypothesis of a valence specific, but stimulus general area within vmPFC that is modulated by tDCS. Affected regions point to visual emotional processing, as ventral and dorsal pathways originating in occipital cortex and travelling along parietal to frontal and temporal cortices are involved (Schupp et al., 2006; Vuilleumier, 2005). In addition, the relative timing of activation in both clusters indicate temporal activation within the interval of the Late Positive Potential component (LPP), which increases during processing of emotional material and usually starts around 300 ms and typically lasts until the end of stimulus presentation[2] (Cuthbert et al., 2000; Schupp et al., 2006).

Results of the behavioral face-morph task align with the MEG findings within late time intervals and further support the hypothesis that valence specific vmPFC regions can be modulated by tDCS. After excitatory relative to inhibitory stimulation, participants required a

[2] With the fixed stimulus presentation time of 600 ms, stimulus offset-expectancy processes might account for the breakdown of significance slightly before stimulus offset.

higher degree of fearful expression in an essentially ambiguous face to categorize it as fearful, and/or a lesser amount of happiness to categorize it as happy (Fig. 2B). Various studies in decision making also identified the vmPFC as core region for coding the overall reward value of stimuli (e.g., Kahnt et al., 2011). This may be interpreted such that the reward value of happy faces increased and the value of fearful faces decreased after excitatory vmPFC stimulation eventually leading to the observed biased valence decisions. This behavioral evidence for modulation of emotional stimulus evaluation and eventual decision making after a single tDCS session is remarkable considering that the behavioral tests invariably followed the MEG session and thus occurred within a period of reduced modulatory effects (Nitsche et al., 2008). Furthermore, analysis of reaction times within the face-morph task showed a cubic trend for the interaction Stimulation x Morph Step (Fig. 2C). After exclusion of the most distinct morph stimuli with almost perfect identification, the interaction of Stimulation x Morph Step revealed a significant cubic effect with relatively decreased reaction times after excitatory compared to inhibitory stimulation for ambiguous faces with a greater degree of happiness. The reverse effect was observed for ambiguous faces with higher amounts of fear. This systematic influence on response speed indicates a bias towards a facilitated identification of happiness after excitatory, but fear after inhibitory vmPFC stimulation. This bias was only present for the more ambiguous faces, consistent with the modest modulation parameters and available flexibility in face appraisal.

In addition to the electrophysiological and behavioral evidence supporting our hypotheses, the MEG analysis also identified an inferior temporal cluster, which, although more focal and phasic (193-240 ms) than the primary effects, showed enhanced activation in response to fearful faces compared to happy faces after excitatory compared to after inhibitory stimulation (Fig. 3B). In this time interval, no cluster with convergent effects across both stimulus types (scenes, faces) was found, indicating that this mid-latency inverse effect may be face specific. In fact, a three-way interaction for Stimulation x Valence x Stimulus Type revealed a significant cluster within this region between 167 and 320 ms and a second right occipital cluster in a similar time interval (210-287 ms), where Stimulus x Valence interactions for emotional scenes led to the predicted outcome, whereas opposing effects were found for facial stimuli (Fig. 5B & C; Supplementary Table 2). Moreover, a mixed-model ANOVA for interaction Valence x Stimulus Type of pre-tDCS activity also revealed a temporally overlapping cluster (130-460 ms) covering the right temporal cortex as well as large areas of right parietal cortex and bilateral prefrontal cortex (Fig. 6). While processing of pleasant scenes evoked increased neural activation in comparison to unpleasant scenes (positivity bias) in this cluster, fearful faces evoked greater activation relative to happy faces (negativity bias). Interestingly, when comparing this pre-tDCS pattern with the post-excitatory tDCS activation (blue bars in Fig. 5B & C), a similar direction of interaction was observed. In

contrast, after inhibitory stimulation, there was a drop in activation particularly for pleasant scenes and fearful faces (orange bars in Fig. 5B & C). One interpretation of this effect is that, in addition to hedonic stimulus valence, the amount of emotional arousal evoked by the stimulus might have contributed to the MEG signal amplitude. In fact, emotional arousal is the main factor for the modulation of affect-related ERP components like the Early Posterior Negativity (EPN) and LPP (e.g., Junghofer et al., 2001; Keil et al., 2002; Schupp et al., 2006). For the large pre-tDCS effect (Fig. 6), it is hard to disentangle effects of hedonic valence and emotional arousal. However, as subjective stimulus arousal was not controlled in this experiment, it is possible that there was a systematic difference such that pleasant scenes evoked greater arousal than unpleasant scenes, while fearful faces evoked higher arousal than happy faces. Therefore, under these conditions, the reported three-way interaction could indicate that after excitatory tDCS of vmPFC, no change had been induced, or a priori arousal differences have been amplified, respectively, while after inhibitory tDCS, stimuli with potentially higher arousal potential had been attenuated. Although these possible stimulus type specific effects on tDCS modulated arousal appear smaller, more focal and less sustained than the observed effects on hedonic valence, future studies should investigate possible co-modulations further.

Finally, a third cluster in a very early time window (37-77 ms) at medial prefrontal areas (Fig. 5A) revealed a significant three-way interaction with an activation pattern inverted to the mid-latency three-way interactions (Fig. 5B & C). This quite early effect did not show a priori pre-tDCS differences of valence processing between stimulus categories. However, a meta-analysis of neuroimaging studies on emotional face and scene perception (Sabatinelli et al., 2011) revealed stimulus specific sub areas in medial prefrontal cortex regions. Thus, this early three-way interaction might reflect stimulus type specific effects, which however awaits confirmation by future studies.

Results of the post hoc-analyses of hemispheric dominance indicate stimulus processing with partially stronger right lateralization. Alhough the interaction effect of Stimulation x Valence across stimulus types did not reach significance, inspection of the MEG findings for emotional scenes only (Junghofer et al., 2017) show effects in a high number of right situated regions across time. In context of bottom-up attention processes this right hemispheric dominance conforms with the strongly right lateralized ventral stream of bottom-up attention processes (ventral attention network or task-negative network) which can be found amongst others in right temporal parietal junction and right ventral frontal cortex, and of which the vmPFC is a major hub (Corbetta & Shulman, 2002; Fox et al., 2006).

Taken together, we find strong tDCS modulation effects consistent with our hypotheses and across stimulus types first appearing at 330 ms and lasting until 583 ms after stimulus

onset. It might be possible that the vmPFC-tDCS modulation shows full effect not before late time intervals. For example, Sabatinelli et al. (2013) report significant correlations between LPP and BOLD activity within ventral striatum, a subcortical hub of the dopaminergic reward circuit, of which the vmPFC is the major cortical interconnection. Findings at earlier time intervals, which have been inconsistent across both stimulus types may be driven by more stimulus specific factors, for example face processing, which is reflected in the face-sensitive N170 component and shows clear differences between faces and scenes (Thom et al., 2014). Another possible influence may have come from additional modulation of stimulus arousal. Hence, it would be interesting if this late consistent modulation effect can be generalized further for other valence specific stimulus types, e.g., rewarding stimuli vs stimuli indicating loss, or conditioned aversive stimuli vs safety signals to confirm valence modulation independent of other factors.

It should be mentioned that – in the whole head analysis - there were not any modulatory effects of tDCS on emotional face processing at the local site of stimulation, which conforms to our previous studies on emotional scene processing where local stimulation effects were verifiable neither in the fMRI nor in the MEG study. This finding does not surprise as remote neurostimulation network effects without effects on the stimulated area itself have also frequently been reported by other researchers (Chib et al., 2013; Kimbrell et al., 2002; Notzon et al., 2017). It is noteworthy though that a post hoc region of interest (ROI) analysis of the vmPFC in fact revealed a significant interaction effect of Stimulation x Valence consistent with our hypothesis (Supplementary Fig. 1). Moreover, a ROI analysis of vmPFC activation before stimulation revealed increased activation for pleasant compared to unpleasant stimuli (Supplementary Table 3, Supplementary Fig. 2), further supporting the specificity of the vmPFC for positive valence and its modulation capability by tDCS.

4.1. Limitations

We did not identify any significant vmPFC-tDCS effects in the dot-probe task. Possible reasons for this might lie in our sample of healthy participants. Schmukle (2005) for instance showed a very low retest reliability for the dot-probe task in two independent healthy samples. Hence, it might be possible that the given task sensitivity is sufficient to detect differences within clinical samples only (Bar-Haim et al., 2007; Peckham et al., 2010). In addition, exposure times of the relevant cues may be a critical factor. In their meta-analysis, Bar-Haim et al. (2007) report significant effects for healthy participants only during subliminal exposure times of dot-probe cues.

The outer morph steps in the face-morph task proved to be highly distinct and difficult to modulate, which led to missing cells for the forced choice task data. In terms of the hypothesis that tDCS of vmPFC might have its strongest impact on the processing of

ambiguous stimuli, for future investigations, it might be useful to use morphs within a narrower range around the PM, perhaps optimally chosen by use of an individual adaptive test procedure.

The current study did control for arousal of pleasant and unpleasant face stimuli. Three-way interactions with reversed valence effects in dependency of stimulus type, however, may be a result of face and/or scene intensity as additional factor of influence. Though, it should be noted that the above described interpretations regarding the mid-latency reversed pattern in the light of possible arousal differences is quite speculative and should be investigated further.

The current flow induced by tDCS is rather coarse and does thus not allow a precise stimulation of specific brain regions with clearly defined borders. However, a computer simulation of the current flows based on a FEM conductor model (Wagner et al., 2014) revealed a predominant stimulation of anterior vmPFC which is in line with the regions of 'positive affect' as described by Myers-Schulz and Koenigs (2012) and a relatively weak stimulation of other regions, especially posterior parts of the vmPFC. An increase in focality via realistic head modelling for each participant and the application of multi-electrode current delivery might further improve the effects of anterior vmPFC stimulation. Because of physical laws, tDCS will always stimulate superficial regions to a higher extent than deeper structures (Wagner et al., 2016). Thus, in contrast to the anterior more superficial parts, a quite specific noninvasive stimulation of posterior vmPFC regions is impossible. Thus, with respect to capabilities of tDCS it appears as a fortunate circumstance that regions revealing a positivity bias are in the range of transcranial stimulation.

To reduce variance, we here opted for a within study design with direct comparison of excitatory and inhibitory stimulation. This study does thus not provide any inferences on the effect of excitatory or inhibitory brain stimulation compared to a session without any stimulation[3], which could have been assessed with a third so called sham or placebo stimulation. However, while subjects cannot differentiate anodal from cathodal stimulation they can typically clearly differentiate between real and sham stimulation. Future studies with between-subject designs and additional sham stimulation are thus necessary to resolve the specific impact of anodal and cathodal stimulation with respect to baseline on emotional processing.

5. Conclusion and outlook

[3] A comparison with the pre-tDCS session would not be valid as these sessions always preceded the post-tDCS sessions (i.e., differentiation from order and habituation effects is not possible).

With this study we were able to replicate previous findings (Junghofer et al., 2017) supporting the role of the vmPFC as a valence driven region which can be succesfully modulated by excitatory and inhibitory tDCS. Here, we showed that this modulation also occurs for the processing of pleasant and unpleasant, namely happy and fearful, facial expressions. Moreover, we could show that these modulatory effects were strong enough to shift face perception toward a preferential processing of happy compared to fearful expressions and eventually modulate affect related decisions. These first results supporting a generalization of positive vmPFC-tDCS effects should stimulate convergent studies with other kinds of visual emotional material such as printed emotional words but also for other domains such as auditory (e.g., liked/disliked music), somatosenory (e.g., stroke/pain), or olfactory (e.g., rose/hydrosulfide) sensations.

In spite of the strong convergence of effects on emotional scene and face processing, we also observed differences which might be derived in part from baseline differences of valence processing and emotional arousal as well as specific characteristics of both stimulus types. Follow up studies with other stimulus material as mentioned above should help to further differentiate generalized and stimulus type specific effects.

The convergent findings across emotional scene and face processing indicate a promising application of tDCS of vmPFC in clinical settings. Medial prefrontal areas show abnormalities in a variety of disorders that are associated with disturbed reactions toward reward- or safety-signaling stimuli, e.g., posttraumatic stress disorder (Milad et al., 2009), obsessive compulsive disorder (Apergis-Schoute et al., 2017), MDD and dysphoria (Grimm et al., 2009; Sabatinelli et al., 2015), generalized anxiety disorder (Greenberg et al., 2013), and obesity (Opel et al., 2015). Therefore, our approach may prove to be an inexpensive, safe, and user-friendly add-on to standard therapeutic interventions in the future.

Funding

This work was funded by the Interdisciplinary Center for Clinical Research of the University of Muenster (Ju2/024/15)

Conflict of interest

The authors declare no competing financial interests.

Acknowledgments

We thank Karin Wilken, Hildegard Deitermann, and Ute Trompeter for their help with the MEG data collection, and Andreas Wollbrink for technical assistance.

References

Adolphs, R. (2002). Neural systems for recognizing emotion. *Current Opinion in Neurobiology*, *12*(2), 169–177. http://doi.org/10.1016/S0959-4388(02)00301-X

Apergis-Schoute, A.M., Gillan, C.M., Fineberg, N.A., Fernandez-Egea, E., Sahakian, B.J., & Robbins, T.W. (2017). Neural basis of impaired safety signaling in obsessive compulsive disorder. *Proceedings of the National Academy of Sciences*, *114*(12), 3216–3221. http://doi.org/10.1073/pnas.1609194114

Atlas L.Y., Bolger N., Lindquist M.A., Wager T.D. (2010). Brain mediators of predictive cue effects on perceived pain. *J Neurosci*; *30*(39): 12964-12977. https://doi.org/10.1523/jneurosci.0057-10.2010

Bar-Haim, Y., Lamy, D., Pergamin, L., Bakermans-Kranenburg, M.J., & van IJzendoorn, M.H. (2007). Threat-related attentional bias in anxious and nonanxious individuals: a meta-analytic study. *Psychological Bulletin*, *133*(1), 1–24. http://doi.org/10.1037/0033-2909.133.1.1

Beck, A.T., Steer, R.A., & Brown, G.K. (1996). Manual for the Beck Depression Inventory-II, *Psychological Corporation*, *San Antonio*. https://doi.org/10.1037/t00742-000

Brainard, D.H. (1997). The Psychophysics Toolbox. *Spatial Vision*, *10*, 433–436. https://doi.org/10.1163/156856897x00357

Chib, V.S., Yun, K., Takahashi, H., & Shimojo, S. (2013). Noninvasive remote activation of the ventral midbrain by transcranial direct current stimulation of prefrontal cortex. *Translational Psychiatry*, *3*(6), e268. http://doi.org/10.1038/tp.2013.44

Costa, V.D., Lang, P.J., Sabatinelli, D., Versace, F., & Bradley, M.M. (2010). Emotional imagery: assessing pleasure and arousal in the brain's reward circuitry. *Human Brain Mapping*, *31*(9), 1446–1457. http://doi.org/10.1002/hbm.20948

Corbetta, M., & Shulman, G.L. (2002). Control of goal-directed and stimulus-driven attention in the brain. *Nature Reviews Neuroscience*, *3*(3), 201–215. http://doi.org/10.1038/nrn755

Cuthbert, B.N., Schupp, H.T., Bradley, M.M., Birbaumer, N., & Lang, P.J. (2000). Brain potentials in affective picture processing: covariation with autonomic arousal and affective report. *Biological Psychology*, *52*(2), 95–111. http://doi.org/10.1016/S0301-0511(99)00044-7

Flaisch, T., Schupp, H.T., Renner, B., & Junghofer, M. (2009). Neural systems of visual attention responding to emotional gestures. *NeuroImage*, *45*(4), 1339–1346. http://doi.org/10.1016/j.neuroimage.2008.12.073

Foti, D., Olvet, D.M., Klein, D.N., & Hajcak, G. (2010). Reduced electrocortical response to threatening faces in major depressive disorder. *Depression and Anxiety*, *27*(9), 813–820. http://doi.org/10.1002/da.20712

Fox, M.D., Corbetta, M., Snyder, A.Z., Vincent, J.L., & Raichle, M.E. (2006). Spontaneous neuronal activity distinguishes human dorsal and ventral attention systems. *Proceedings of the National Academy of Sciences*, *103*(26), 10046–10051. http://doi.org/10.1073/pnas.0604187103

Franke, G.H. (2014). *Symptomcheckliste-90-S*, *Hogrefe*, *Göttingen*.

Freese, J.L., & Amaral, D.G. (2006). Synaptic organization of projections from the amygdala to visual cortical areas TE and V1 in the macaque monkey. *J Comp Neurol*, *496*(5), 655–667. http://doi.org/10.1002/cne

Greenberg, T., Carlson, J.M., Cha, J., Hajcak, G., & Mujica-Parodi, L.R. (2013). Ventromedial prefrontal cortex reactivity is altered in generalized anxiety disorder during fear generalization. *Depression and Anxiety*, *30*(November 2012), 242–250. http://doi.org/10.1002/da.22016

Grimm, S., Boesiger, P., Beck, J., Schuepbach, D., Bermpohl, F., Walter, M., … Northoff, G. (2009). Altered negative BOLD responses in the default-mode network during emotion processing in depressed subjects. *Neuropsychopharmacology*, *34*(4), 932–943. http://doi.org/10.1038/npp.2008.81

Hajcak Proudfit, G. (2015). The reward positivity: From basic research on reward to a biomarker for depression. *Psychophysiology*, *52*(2014). http://doi.org/10.1111/psyp.12370

Hämäläinen, M.S., & Ilmoniemi, R.J. (1994). Interpreting magnetic fields of the brain: minimum norm estimates. *Medical and Biological Engineering and Computing*, *32*, 35–42. https://doi.org/10.1007/BF02512476

Junghofer, M., Bradley, M.M., Elbert, T.R., & Lang, P.J. (2001). Fleeting images: a new look at early emotion discrimination. *Psychophysiology*, *38*, 175–178. http://doi.org/10.1111/1469-8986.3820175

Junghofer, M., Winker, C., Rehbein, M.A., & Sabatinelli, D. (2017). Noninvasive stimulation of the ventromedial prefrontal cortex enhances pleasant scene processing. *Cerebral Cortex*, *27*(6), 3449–3456. http://doi.org/10.1093/cercor/bhx073

Kahnt T., Heinzle J., Park S.Q., Haynes J.D. (2011). Decoding different roles for vmPFC and dlPFC in multi-attribute decision making. *NeuroImage*, *56*(2):709-15. https://doi.org/10.1016/j.neuroimage.2010.05.058

Keil, A., Bradley, M.M., Hauk, O., Rockstroh, B., Elbert, T., & Lang, P.J. (2002). Large-scale neural correlates of affective picture processing. *Psychophysiology*, *39*(5), 641–9. http://doi.org/10.1017.S0048577202394162

Kimbrell, T.A., Dunn, R.T., George, M.S., Danielson, A.L., Willis, M.W., Repella, J.D., … Wassermann, E.M. (2002). Left prefrontal-repetitive transcranial magnetic stimulation (rTMS) and regional cerebral glucose metabolism in normal volunteers. *Psychiatry Research: Neuroimaging*, *115*(3), 101–113. http://doi.org/10.1016/S0925-4927(02)00041-0

Kissler, J., Herbert, C., Peyk, P., & Junghofer, M. (2007). Buzzwords: Early cortical responses to emotional words during reading. *Psychological Science*, *18*(6), 475–480. https://doi.org/10.1111/j.1467-9280.2007.01924.x

Knutson, B., Fong, G.W., Bennett, S.M., Adams, C.M., & Hommer, D. (2003). A region of mesial prefrontal cortex tracks monetarily rewarding outcomes: characterization with rapid event-related fMRI. *NeuroImage*, *18*(2), 263–272. http://doi.org/10.1016/S1053-8119(02)00057-5

Lang, P.J., Bradley, M.M., & Cuthbert, B.N. (1997). Motivated attention: Affect, activation, and action, in: Lang, P.J., Simons, R.F., & Balaban, M. (Eds.), Attention and orienting: sensory and motivational processes, Psychology Press, New York, pp. 97-135. https://doi.org/10.4324/9780203726457

Langner, O., Dotsch, R., Bijlstra, G., Wigboldus, D.H.J., Hawk, S.T., & van Knippenberg, A. (2010). Presentation and validation of the Radboud Faces Database. *Cognition & Emotion*, *24*(8), 1377–1388. http://doi.org/10.1080/02699930903485076

Laux, L., Glanzmann, P., Schaffner, P., & Spielberger, C.D. (1981). Das State-Trait-Angstinventar (STAI) [The State-Trait Anxiety Inventory], *Hogrefe*, *Göttingen/Bern*.

Lundqvist, D., Flykt, A., & Öhman, A. (1998). The Karolinska Directed Emotional Faces – KDEF, CD ROM from Department of Clinical Neuroscience, Psychology section, Karolinska Institutet, ISBN 91-630-7164-9. http://dx.doi.org/10.1037/t27732-000

MacLeod, C., & Mathews, A. (1988). Anxiety and the allocation of attention to threat. *The Quarterly Journal of Experimental Psychology*, *40*(4), 653–670. http://doi.org/10.1080/14640748808402292

MacLeod, C., Mathews, A., & Tata, P. (1986). Attentional bias in emotional disorders. *Journal of Abnormal Psychology*, *95*(1), 15–20. http://doi.org/http://dx.doi.org/10.1037/0021-843X.95.1.15

Maris, E., & Oostenveld, R. (2007). Nonparametric statistical testing of EEG- and MEG-data. *Journal of Neuroscience Methods*, *164*(1), 177–90. http://doi.org/10.1016/j.jneumeth.2007.03.024

McMahon, D.B.T., & Leopold, D.A. (2012). Stimulus timing-dependent plasticity in high-level vision. *Current Biology*, *22*(4), 332–337. http://doi.org/10.1016/j.cub.2012.01.003

Milad, M.R., Wright, C.I., Orr, S.P., Pitman, R.K., Quirk, G.J., & Rauch, S.L. (2007). Recall of fear extinction in humans activates the ventromedial prefrontal cortex and hippocampus in concert. *Biological Psychiatry*, *62*(5), 446–454. http://doi.org/10.1016/j.biopsych.2006.10.011

Milad, M.R., Pitman, R.K., Ellis, C.B., Gold, A.L., Shin, L.M., Lasko, N.B., … Rauch, S.L. (2009). Neurobiological basis of failure to recall extinction memory in posttraumatic stress disorder. *Biological Psychiatry*, *66*(12), 1075–1082. http://doi.org/10.1016/j.biopsych.2009.06.026

Morel, S., Ponz, A., Mercier, M., Vuilleumier, P., & George, N. (2009). EEG-MEG evidence for early differential repetition effects for fearful, happy and neutral faces. *Brain Research*, *1254*, 84–98. http://doi.org/10.1016/j.brainres.2008.11.079

Moser, J.S., Huppert, J.D., Duval, E., & Simons, R.F. (2008). Face processing biases in social anxiety: an electrophysiological study. *Biological Psychology*, *78*(1), 93–103. http://doi.org/10.1016/j.biopsycho.2008.01.005

Mungee, A., Kazzer, P., Feeser, M., Nitsche, M.A., Schiller, D., & Bajbouj, M. (2014). Transcranial direct current stimulation of the prefrontal cortex: a means to modulate fear memories. *NeuroReport*, *25*(7), 480–484. http://doi.org/10.1097/WNR.0000000000000119

Myers-Schulz, B., & Koenigs, M. (2012). Functional anatomy of ventromedial prefrontal cortex: implications for mood and anxiety disorders. *Molecular Psychiatry*, *17*(2), 132–141. http://doi.org/10.1038/mp.2011.88

Nitsche, M.A., Cohen, L.G., Wassermann, E.M., Priori, A., Lang, N., Antal, A., … Pascual-Leone, A. (2008). Transcranial direct current stimulation: state of the art 2008. *Brain Stimulation*, *1*, 206–223. http://doi.org/10.1016/j.brs.2008.06.004

Nitsche, M.A., Seeber, A., Frommann, K., Klein, C.C., Rochford, C., Nitsche, M.S., … Tergau, F. (2005). Modulating parameters of excitability during and after transcranial direct current stimulation of the human motor cortex. *The Journal of Physiology*, *568*(1), 291–303. http://doi.org/10.1113/jphysiol.2005.092429

Notzon, S., Steinberg, C., Zwanzger, P., & Junghofer, M. (2017). Modulating emotion perception: opposing effects of inhibitory and excitatory prefrontal cortex stimulation. Biological Psychiatry: *Cognitive Neuroscience and Neuroimaging*, 1–8. http://doi.org/10.1016/j.bpsc.2017.12.007

Ochsner, K.N., & Gross, J.J. (2005). The cognitive control of emotion. *Trends in Cognitive Sciences*, *9*(5), 242–249. http://doi.org/10.1016/j.tics.2005.03.010

Opel, N., Redlich, R., Grotegerd, D., Dohm, K., Heindel, W., Kugel, H., … Dannlowski, U. (2015). Obesity and major depression: body-mass index (BMI) is associated with a severe course of disease and specific neurostructural alterations. *Psychoneuroendocrinology*, *51*, 219–226. http://doi.org/10.1016/j.psyneuen.2014.10.001

Opel, N., Redlich, R., Grotegerd, D., Dohm, K., Zaremba, D., Meinert, S., … Dannlowski, U. (2017). Prefrontal brain responsiveness to negative stimuli distinguishes familial risk for major depression from acute disorder. *Journal of Psychiatry & Neuroscience*, *42*(5), 1–10. http://doi.org/10.1503/jpn.160198

Pessoa, L., & Adolphs, R. (2010). Emotion processing and the amygdala: from a “low road” to “many roads” of evaluating biological significance. *Nature Reviews Neuroscience*, *11*(11), 773–783. http://doi.org/10.1038/nrn2920

Paulus, W. (2014). Transcranial brain stimulation : potential and limitations. *E-Neuroforum*, 29–36. http://doi.org/10.1007/s13295-014-0056-6

Peckham, A.D., McHugh, R.K., & Otto, M.W. (2010). A meta-analysis of the magnitude of biased attention in depression. *Depression and Anxiety*, *27*(12), 1135–1142. http://doi.org/10.1002/da.20755

Peyk, P., De Cesarei, A., & Junghofer, M. (2011). ElectroMagnetoEncephalography software: overview and integration with other EEG/MEG toolboxes. *Computational Intelligence and Neuroscience*, *2011*. http://doi.org/10.1155/2011/861705

Rellecke, J., Sommer, W., & Schacht, A. (2013). Emotion effects on the N170: a question of reference? *Brain Topography*, *26*(1), 62–71. http://doi.org/10.1007/s10548-012-0261-y

Sabatinelli, D., Bradley, M.M., Lang, P.J., Costa, V.D., & Versace, F. (2007). Pleasure rather than salience activates human nucleus accumbens and medial prefrontal cortex. *Journal of Neurophysiology*, *98*(3), 1374–9. http://doi.org/10.1152/jn.00230.2007

Sabatinelli, D., Fortune, E.E., Li, Q., Siddiqui, A., Krafft, C., Oliver, W.T., … Jeffries, J. (2011). Emotional perception: meta-analyses of face and natural scene processing. *NeuroImage*, *54*(3), 2524–2533. http://doi.org/10.1016/j.neuroimage.2010.10.011

Sabatinelli, D., Keil, A., Frank, D.W., & Lang, P.J. (2013). Emotional perception: correspondence of early and late event-related potentials with cortical and subcortical functional MRI. *Biological Psychology*, *92*(3), 513–519. http://doi.org/10.1016/j.biopsycho.2012.04.005

Sabatinelli, D., McTeague, L.M., Dhamala, M., Frank, D.W., Wanger, T.J., & Adhikari, B.M. (2015). Reduced medial prefrontal-subcortical connectivity in dysphoria: Granger causality analyses of rapid functional magnetic resonance imaging. *Brain Connectivity*, *5*(1), 1–9. http://doi.org/10.1089/brain.2013.0186

Schmukle, S.C. (2005). Unreliability of the dot probe task. *European Journal of Personality*, *19*(7), 595–605. http://doi.org/10.1002/per.554

Schupp, H.T., Öhman, A., Junghofer, M., Weike, A.I., Stockburger, J., & Hamm, A.O. (2004). The facilitated processing of threatening faces: an ERP analysis. *Emotion*, *4*(2), 189–200. http://doi.org/10.1037/1528-3542.4.2.189

Schupp, H.T., Flaisch, T., Stockburger, J., & Junghofer, M. (2006). Emotion and attention: event-related brain potential studies. *Progress in Brain Research*, *156*, 31–51. https://doi.org/10.1016/s0079-6123(06)56002-9

Thom, N., Knight, J., Dishman, R., Sabatinelli, D., Johnson, D.C., & Clementz, B. (2014). Emotional scenes elicit more pronounced self-reported emotional experience and greater EPN and LPP modulation when compared to emotional faces. *Cognitive, Affective & Behavioral Neuroscience*, *14*(2), 849–60. http://doi.org/10.3758/s13415-013-0225-z

Tiddeman, B., & Perrett, D. (2002). Transformation of dynamic facial image sequences using static 2D prototypes. *The Visual Computer*, *18*(4), 218–225. http://doi.org/10.1007/s003710100142

Tottenham, N., Tanaka, J.W., Leon, A.C., McCarry, T., Nurse, M., Hare, T.A., … Nelson, C. (2009). The NimStim set of facial expressions: judgments from untrained research participants. *Psychiatry Research*, *168*(3), 242–9. http://doi.org/10.1016/j.psychres.2008.05.006

Van't Wout, M., Mariano, T.Y., Garnaat, S.L., Reddy, M.K., Rasmussen, S.A., & Greenberg, B.D. (2016). Can transcranial direct current stimulation augment extinction of conditioned fear? *Brain Stimulation*, *9*(4), 529–536. http://doi.org/10.1016/j.brs.2016.03.004

Vuilleumier, P. (2005). How brains beware: neural mechanisms of emotional attention. *Trends in Cognitive Sciences*, *9*(12), 585–94. http://doi.org/10.1016/j.tics.2005.10.011

Vuilleumier, P., & Pourtois, G. (2007). Distributed and interactive brain mechanisms during emotion face perception: evidence from functional neuroimaging. *Neuropsychologia*, *45*(1), 174–94. http://doi.org/10.1016/j.neuropsychologia.2006.06.003

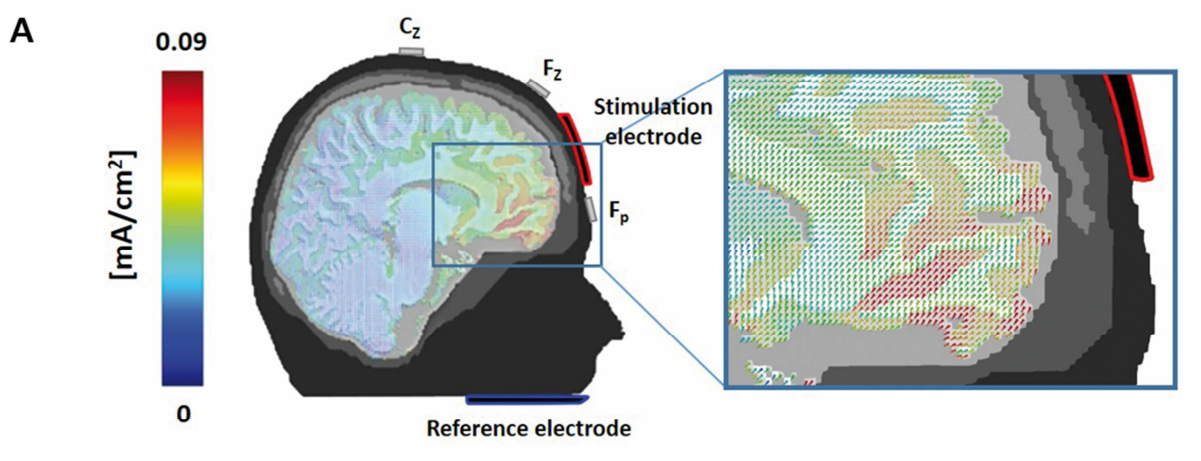
Wagner, S., Rampersad, S.M., Aydin, Ü., Vorwerk, J., Oostendorp, T.F., Neuling, T., … Wolters, C.H. (2014). Investigation of tDCS volume conduction effects in a highly realistic head model. *Journal of Neural Engineering*, *11*(1), 16002. http://doi.org/10.1088/1741-2560/11/1/016002

Wagner, S., Lucka, F., Vorwerk, J., Herrmann, C.S., Nolte, G., Burger, M., & Wolters, C.H. (2016). Using reciprocity for relating the simulation of transcranial current stimulation to the EEG forward problem. *NeuroImage*, *140*, 163–173. http://doi.org/10.1016/j.neuroimage.2016.04.005

	Faces	**Scenes**
	***N* = 40**	***N* = 33**
	***M* (*SD*)**	***M* (*SD*)**
Age	24.6 (2.9)	23.58 (4.18)
STAI		
State	30.15 (5.02)	33.18 (5.4)
Trait	30.37 (6.58)	32.15 (8.3)
Trait *T*-value	44.90 (8.83)	46.97 (9.6)
BDI-I		2.78 (2.59)[†]
BDI-II	2.2 (2.57)	
SCL-90-S		
GSI	40 unremarkable	
PSDI	40 unremarkable	
PST	10.1 (6.79)	

Table 1

To ensure the assessment of healthy samples, clinical questionnaires with regard to symptoms of major depressive disorder (BDI-II) as well as general psychiatric symptoms (SCL-90-S) were assessed in the current study (Faces). To check for conspicuities regarding symptoms of trait-and state-dependent anxiety, the STAI was assessed. Due to conjoint analyses across independent samples the assessed data from the previous study (Scenes) is reported here in addition with questionnaires BDI-I and STAI. There were no indications for conspicuities in the assessed samples. STAI: State Trait Anxiety Inventory; BDI: Becks Depression Inventory; SCL: Symptom Check List; GSI: Global Severity Index; PSDI: Positive Symptom Distress Index; PST: Positive Symptom Total. [†]*N* = 32 due to missing data.



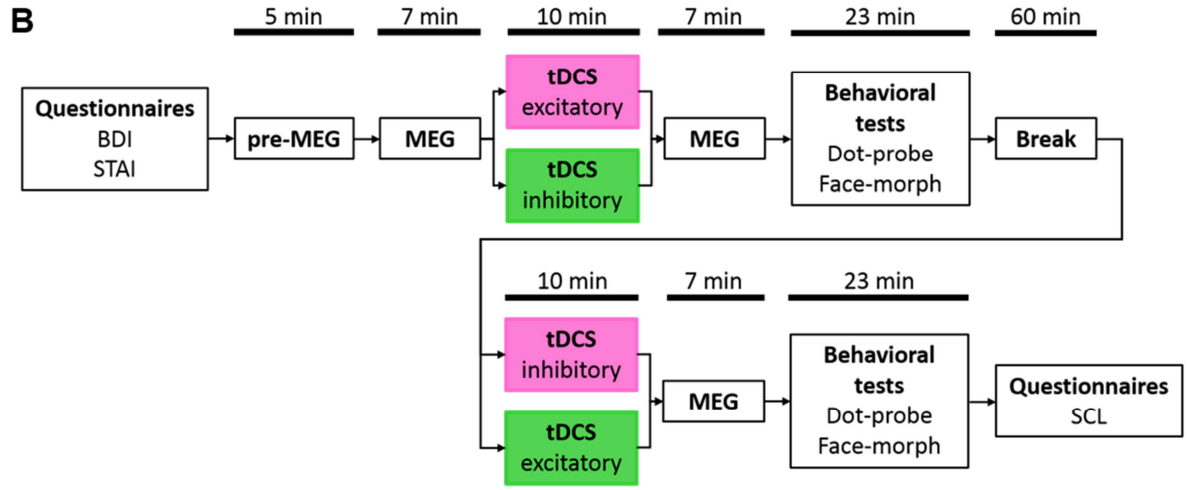
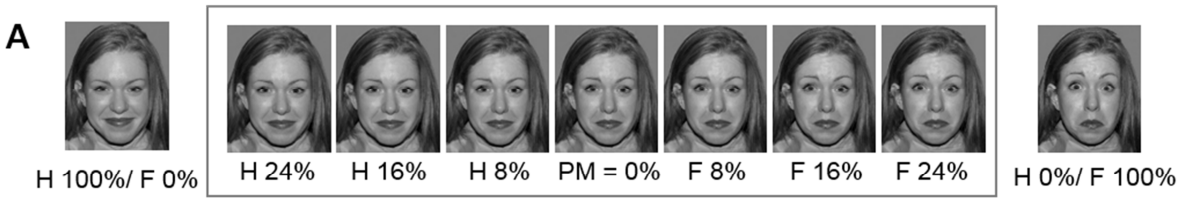
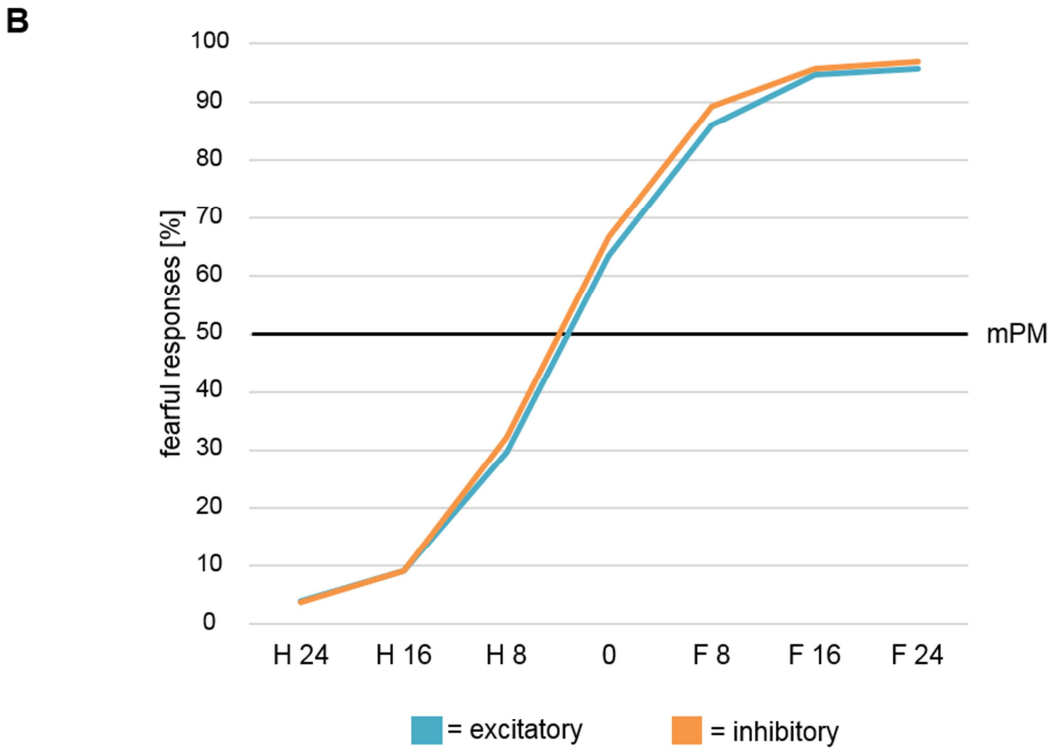


Figure 1

(A) tDCS setup. By means of a FEM based forward model of the current flow, it was possible to position the stimulation electrode (above forehead) and reference electrode (below chin) for precise targeting of the vmPFC. Colored cones within the volume conductor model indicate strength and direction of the current. Current strength reaches highest values in anterior vmPFC areas, while surrounding areas receive little to no current. **(B)** Study timetable. Order of stimulation was balanced with half the participants receiving excitatory tDCS first and inhibitory tDCS second (pink box) and vice versa for the other half (green box). MEG and behavioral test durations are presented as average values, respectively, due to jittered inter-trial intervals in the passive viewing task (MEG) and individual differences in the behavioral tasks.


A
H 100%/ F 0%
H 24%
H 16%
H 8%
PM = 0%
F 8%
F 16%
F 24%
H 0%/ F 100%


B
fearful responses [%]
100
90
80
70
60
50
40
30
20
10
0
mPM
H 24
H 16
H 8
0
F 8
F 16
F 24
= excitatory
= inhibitory

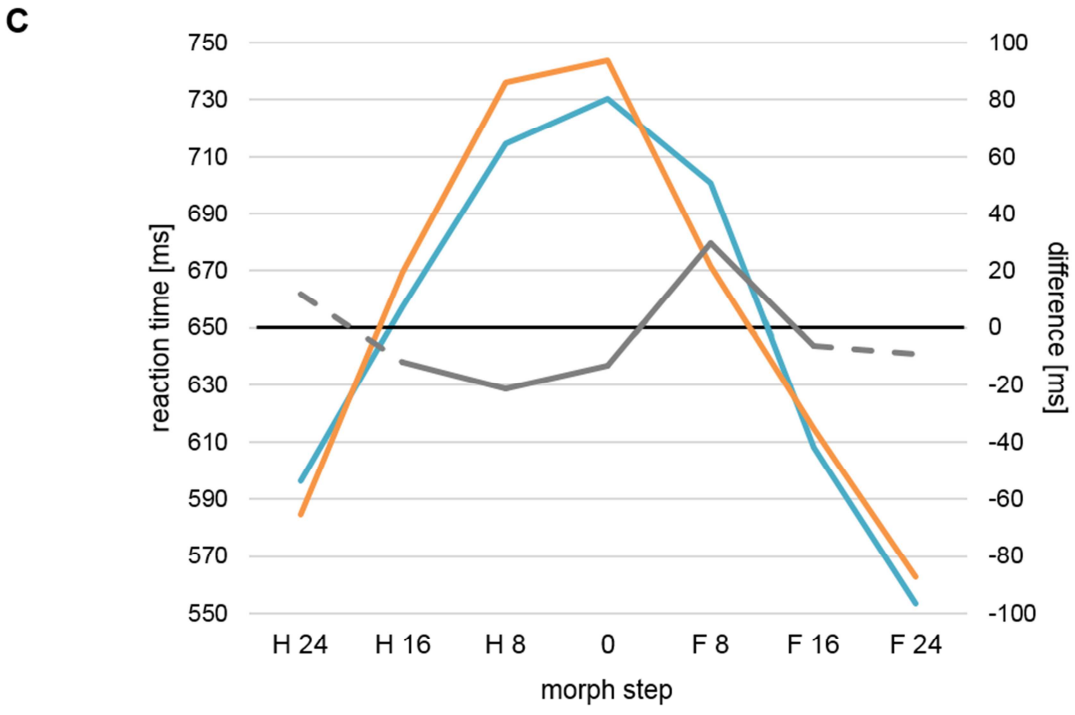
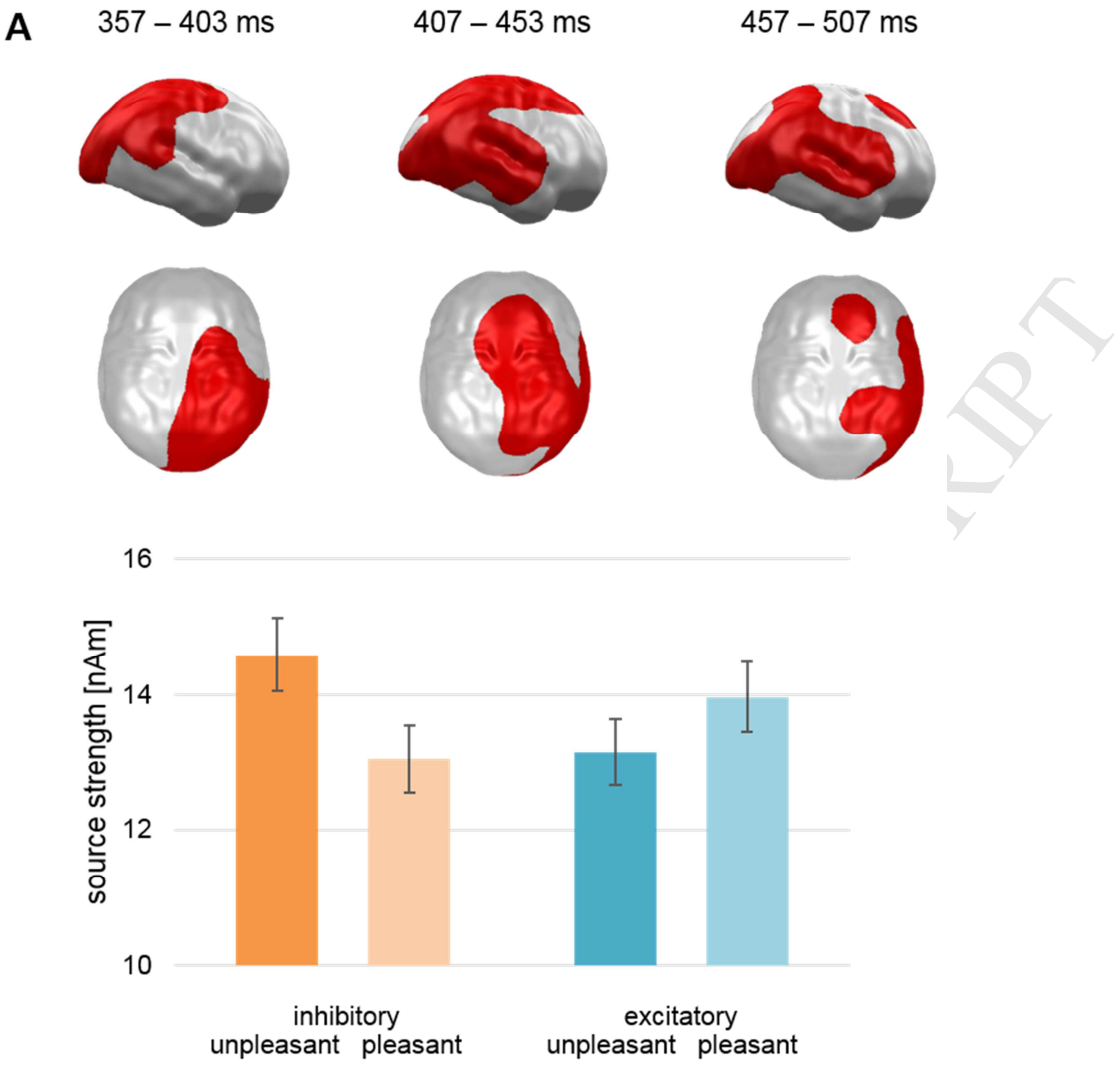

C
reaction time [ms]
750
730
710
690
670
650
630
610
590
570
550
difference [ms]
100
80
60
40
20
0
-20
-40
-60
-80
-100
H 24
H 16
H 8
0
F 8
F 16
F 24
morph step

Figure 2

(A) Face-morph task. Morphed facial stimuli from left to right reveal increasing fearful and decreasing happy expression. The pre-assessed perceptual midpoint (PM) is centrally positioned. Six morph steps around PM were chosen for optimal response function coverage. **(B)** Shift of modulated perceptual midpoint (mPM). A significant difference for the Weibull function modeled mPM between both post-stimulation conditions was observed. After excitatory stimulation, relatively less amount of happiness in a face was necessary to categorize an ambiguous face as happy and vice versa after inhibitory stimulation. **(C)** Reaction time analysis. After exclusion of the most distinct morph steps (H 24%, F 24%), a significant cubic effect occurred for the interaction Stimulation x Morph Step. The difference (Excitatory minus Inhibitory; solid grey line) reveals relatively faster reactions for happy faces after excitatory stimulation and relatively faster reactions for fearful faces after inhibitory stimulation.


A
357 – 403 ms
407 – 453 ms
457 – 507 ms
16
14
12
10
source strength [nAm]
inhibitory
unpleasant
pleasant
excitatory
unpleasant
pleasant

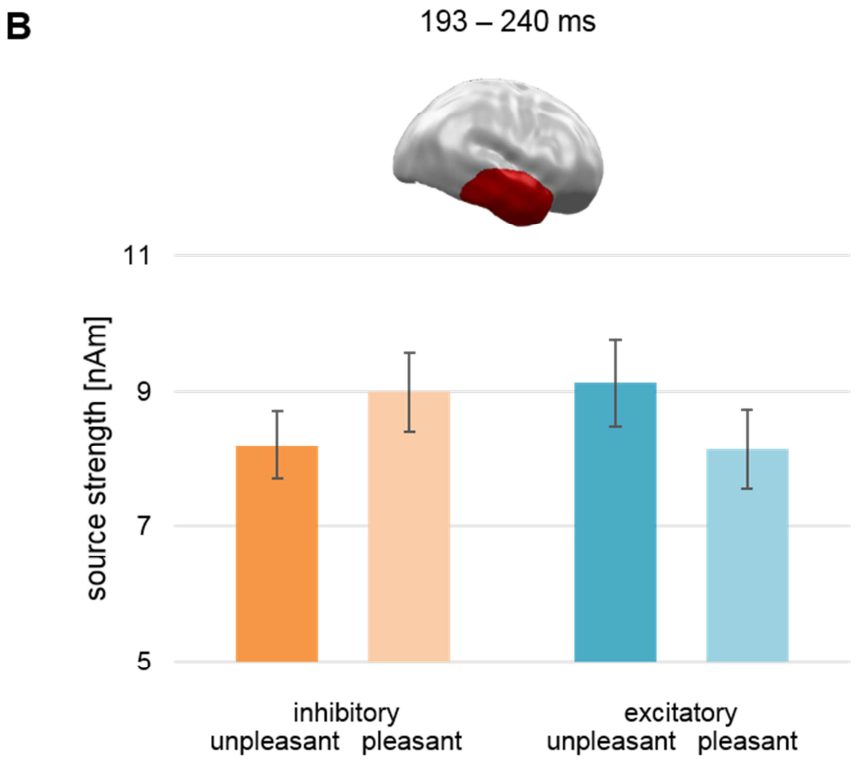

B
193 – 240 ms
11
9
7
5
source strength [nAm]
inhibitory
unpleasant
pleasant
excitatory
unpleasant
pleasant

Figure 3

(A) Late latency cluster for the interaction of Stimulation x Valence. A widely distributed and sustained spatio-temporal cluster was found spanning across right occipital, parietal, temporal as well as frontal regions in the late latency time interval. The activation pattern within this cluster is consistent with our hypothesis with higher activation for happy faces in comparison to fearful faces after excitatory stimulation and vice versa after inhibitory stimulation. **(B)** Mid-latency cluster for interaction Stimulation x Valence. A more short-lived and more focal cluster with an activation pattern in reversed direction to our hypothesis was observed in the right temporal cortex. Bar graph insets indicate standard error of the mean (±SEM).

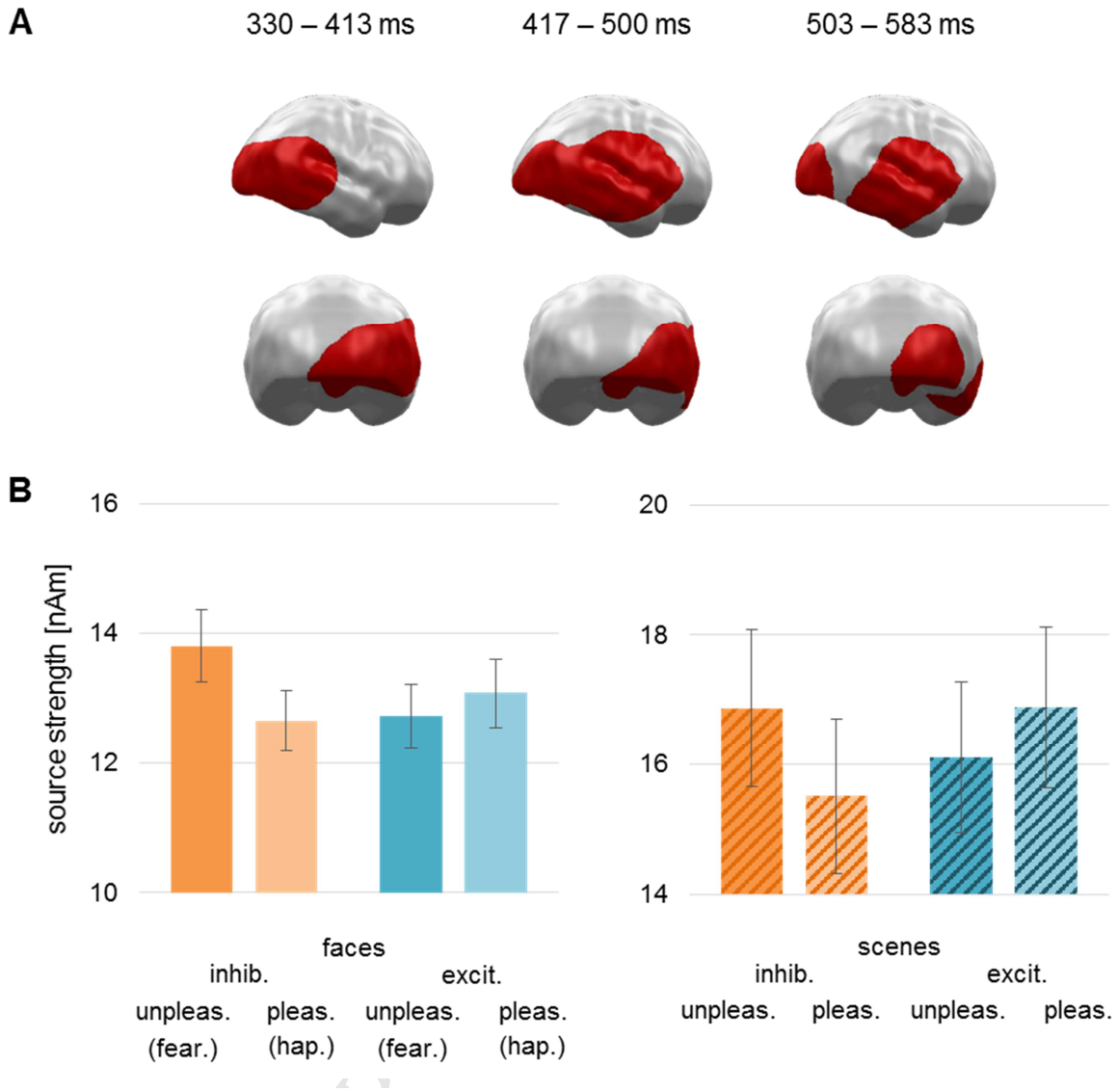
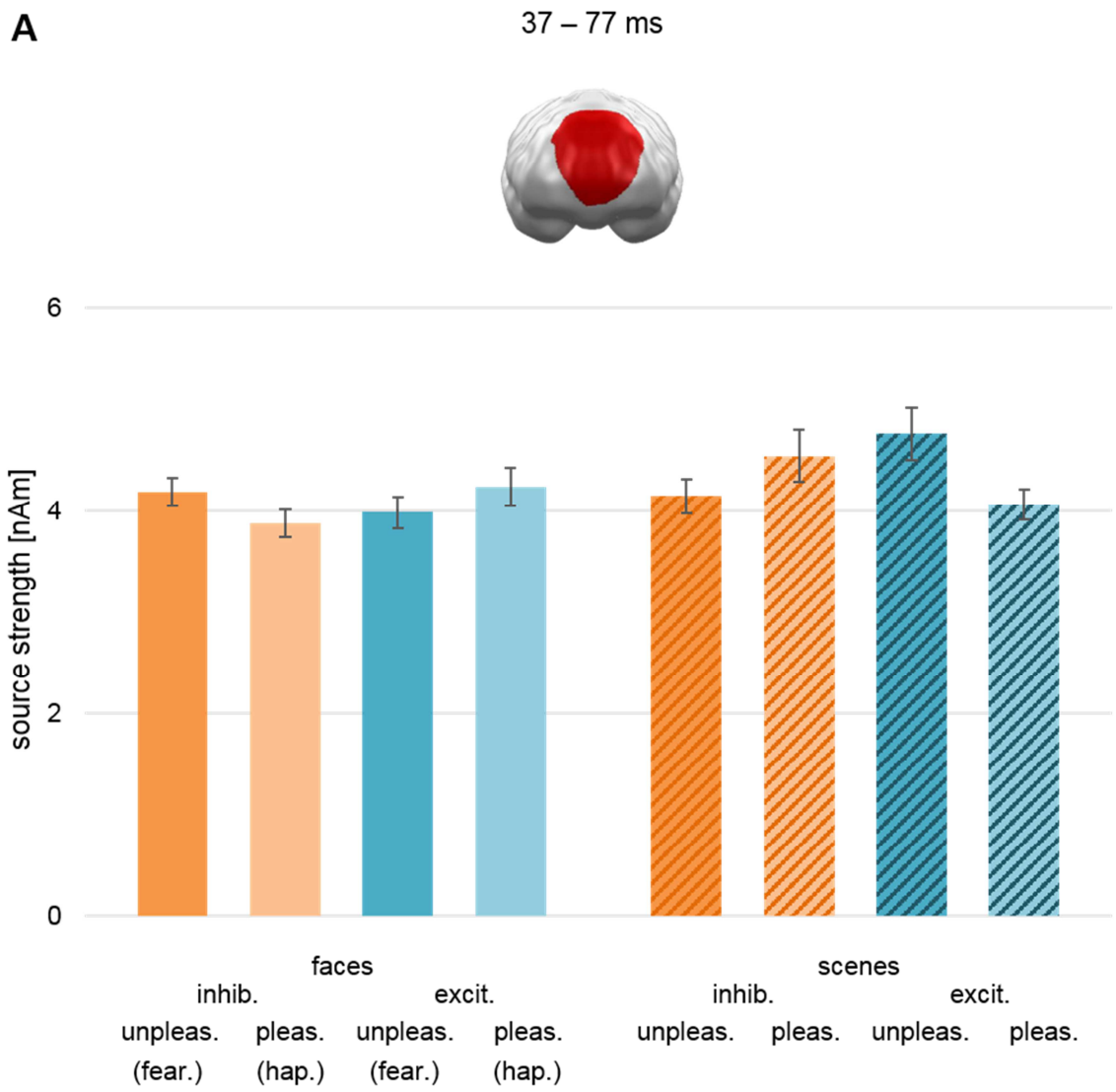
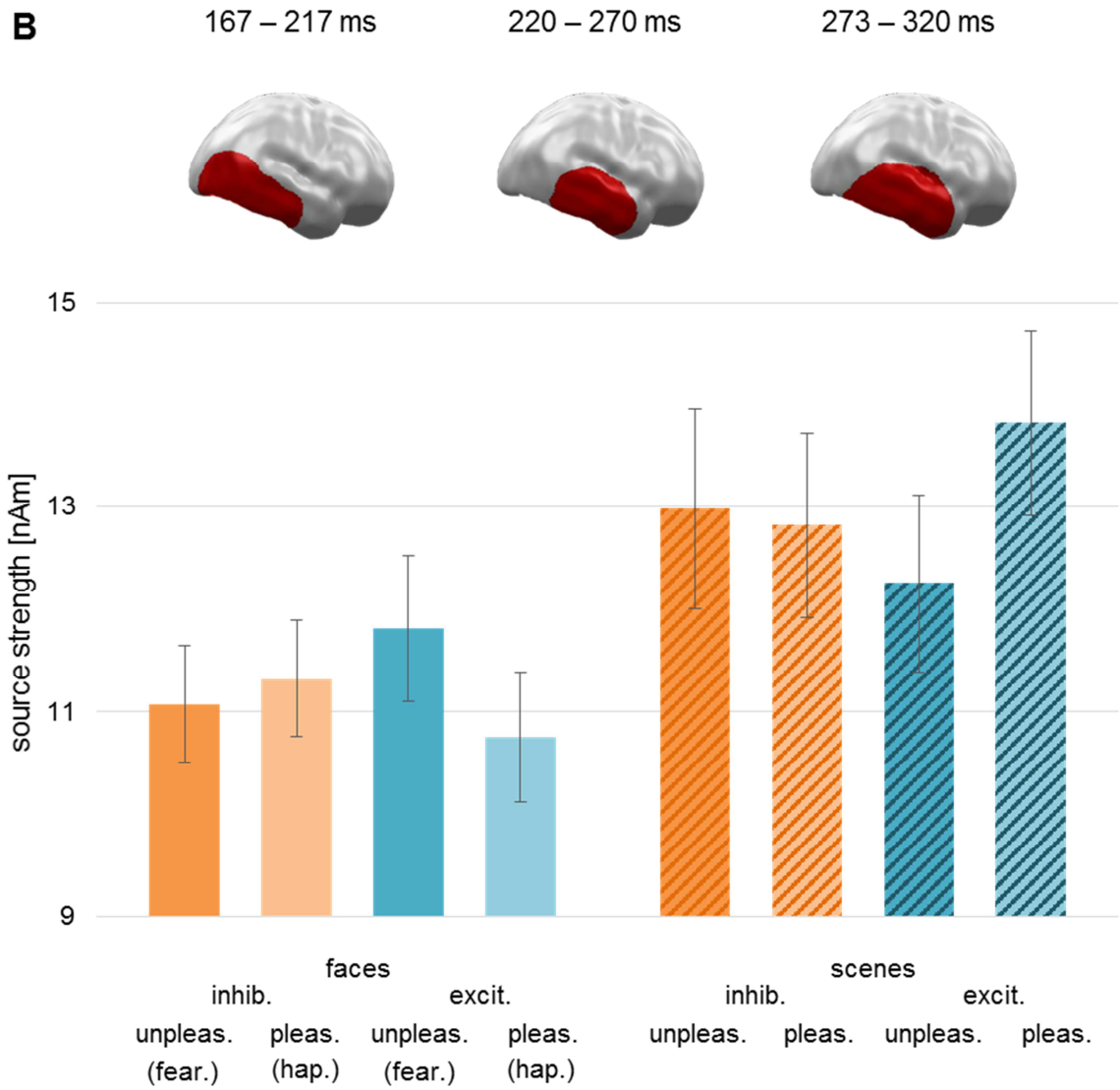


Figure 4

(A) Conjoint ANOVA across faces and scenes. The conjoint ANOVA across both the assessed sample with presented emotional faces and the sample with presented emotional scenes from our previous study (Junghofer et al., 2017) revealed a significant spatio-temporal cluster for the interaction Stimulation x Valence with a pattern convergent to the results for Faces only (Figure 3A). **(B)** Separate post hoc ANOVAs split up for factor Stimulus Type. Post hoc analyses revealed highly significant Stimulation x Valence interactions for both stimulus categories. Bar graph insets indicate standard error of the mean (±SEM).


A
37 – 77 ms
6
4
2
0
source strength [nAm]
faces
scenes
inhib.
excit.
inhib.
excit.
unpleas. (fear.)
pleas. (hap.)
unpleas. (fear.)
pleas. (hap.)
unpleas.
pleas.
unpleas.
pleas.


B
167 – 217 ms
220 – 270 ms
273 – 320 ms
15
13
11
9
source strength [nAm]
faces
inhib.
excit.
unpleas. (fear.)
pleas. (hap.)
unpleas. (fear.)
pleas. (hap.)
scenes
inhib.
excit.
unpleas.
pleas.
unpleas.
pleas.

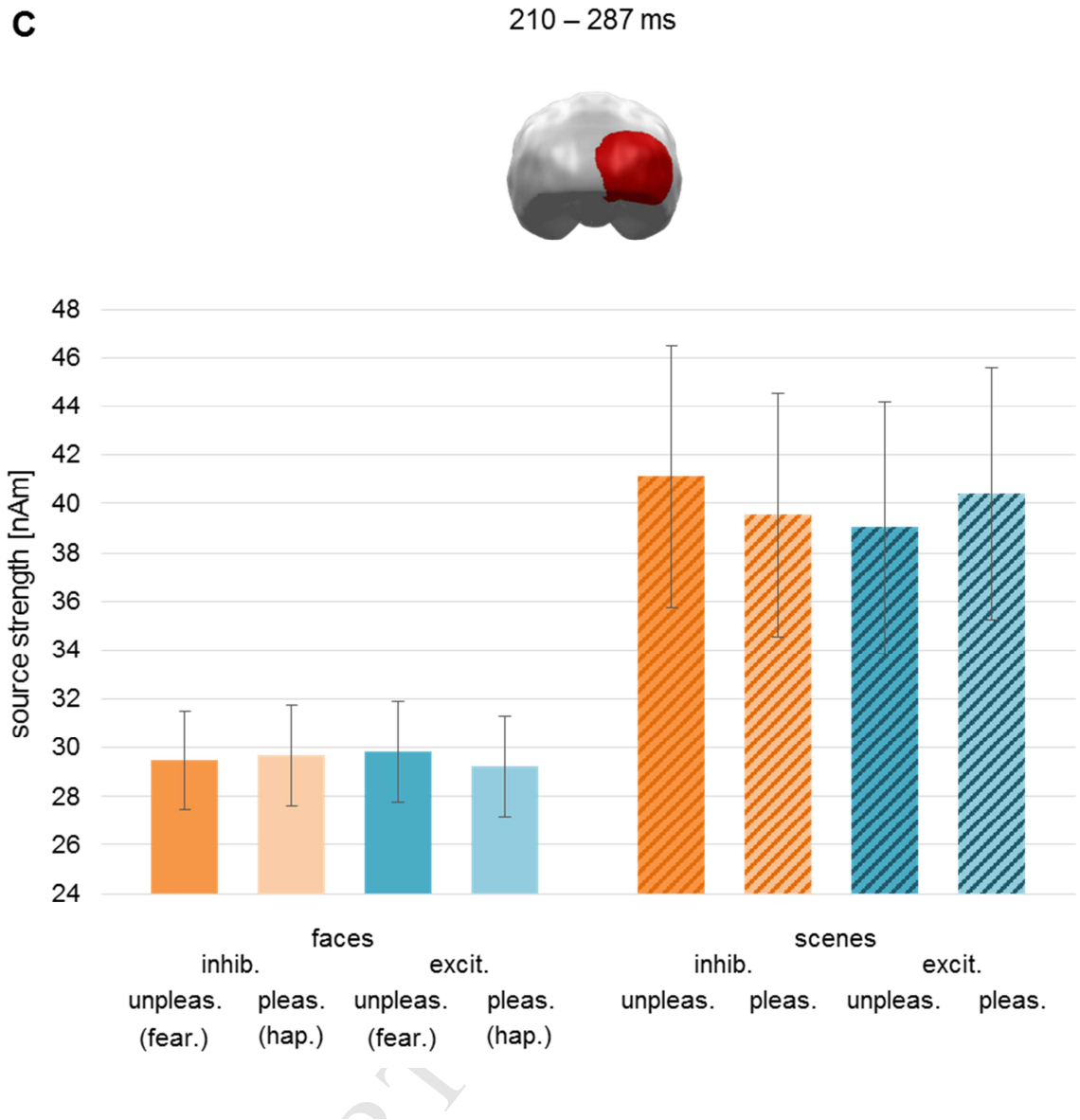


Figure 5

(A) Early prefrontal cluster for the three-way interaction Stimulation x Valence x Stimulus Type. A bilateral prefrontal cluster in a very early time interval (<100 ms) revealed an activation pattern consistent with our hypothesis for emotional faces and a reversed activation pattern for emotional scenes. **(B)** At right temporal cortex a significant three-way interaction reflected previous findings of our separate analyses within both studies with a reversed activation pattern for emotional faces (see Fig. 3B) and an activation consistent with our hypothesis for emotional scenes (see Junghofer et al., 2017). **(C)** Right occipital mid-latency cluster for the three-way interaction. The direction of this interaction shows an activation pattern consistent with our hypothesis for emotional scenes, while for emotional faces almost no difference between conditions can be observed. Bar graph insets indicate standard error of the mean (±SEM).

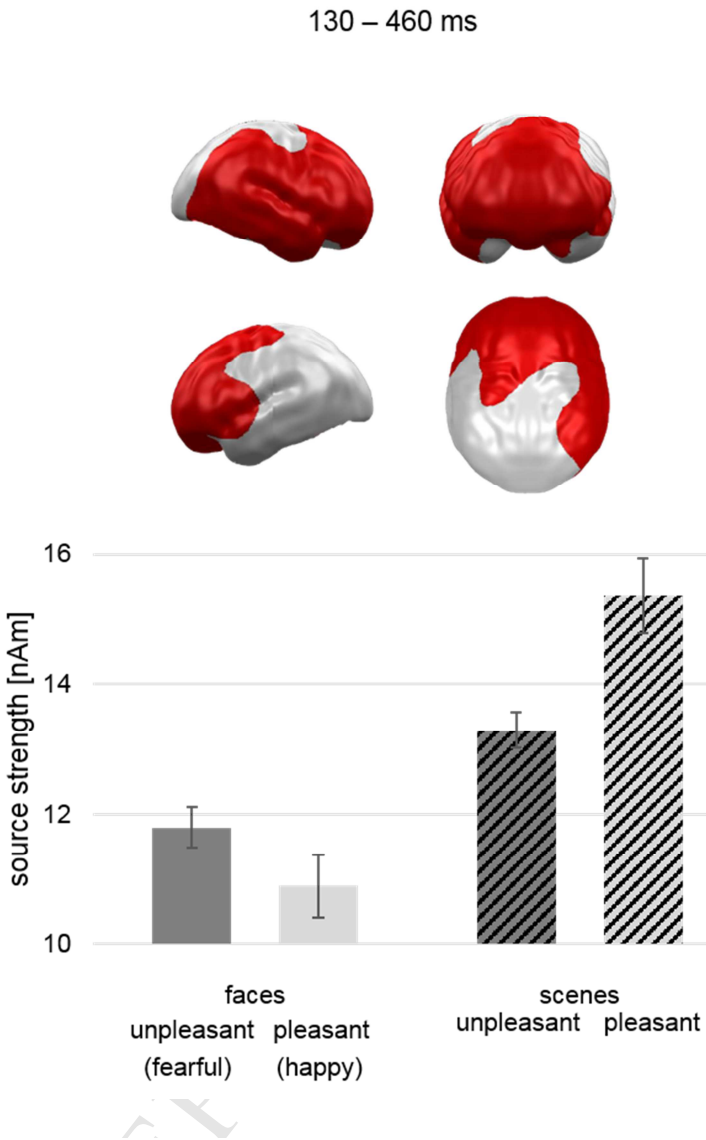


Figure 6

A conjoint ANOVA across the assessed sample with presented emotional faces and the sample with presented emotional scenes both measured before tDCS (i.e. pre-tDCS) revealed a widely distributed and sustained spatio-temporal cluster for the Stimulus Type x Valence interaction qualified by a relative negativity bias (Fearful>Happy) for emotional faces but a relative positivity bias (Pleasant>Unpleasant) for emotional scenes. This effect might be driven by relative differences in perceived arousal within the stimulus categories. Bar graph insets indicate standard error of the mean (±SEM).