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Exploring the effect of transcranial direct current stimulation during sleep on fear extinction consolidation -

Aims

Fear conditioning is a widely used experimental model for studying fear learning, extinction, and the dysfunctional responses seen in anxiety disorders (Lonsdorf et al., 2017). This approach examines various learning phases, such as acquisition, extinction, and retrieval. In a typical protocol, a danger-related stimulus (CS+) is paired with an aversive stimulus (US), while a safety-related stimulus (CS-) is not, allowing assessment of both threat and safety responses.

REM sleep is crucial for consolidating extinction memory. Studies in rodents show that REM sleep deprivation impairs both fear extinction and recall (Jung & Noh, 2021) and disrupting the infralimbic (IL) cortex during REM sleep negatively affects extinction memory consolidation (Hong et al., 2024). In humans, REM sleep increases vmPFC activation, enhancing extinction recall (Menz et al., 2016). REM sleep activation in regions such as the amygdala, hippocampus, ACC, and vmPFC overlaps with areas involved in fear learning and extinction, underscoring REM sleep's importance (Pace-Schott et al., 2015).

Transcranial direct current stimulation (tDCS) is an emerging tool for brain modulation, applying mild electrical currents to enhance or reduce cortical excitability based on polarity—anodal stimulation increases excitability, while cathodal decreases it (Nitsche & Paulus, 2000). Evidence suggests tDCS can boost memory consolidation during sleep, with studies showing anodal tDCS during REM sleep enhancing motor memory (Nitsche et al., 2010) and slow-oscillatory tDCS during non-REM sleep aids declarative memory (Cellini et al., 2019).

This study seeks to test whether prefrontal cortex (PFC) modulation via tDCS during sleep can enhance extinction learning, as well as explore associated neural mechanisms.

Protocol and procedure

Following a pilot investigation phase involving 18 participants for refining procedures to maximize data quality, address potential confounds and ensure that the experimental conditions are robust enough to capture the effects of interest accurately, the results of two experiments are presented to address the above-mentioned aims.

Experiment 1

Participants

32 subjects (age 24.16 ± 4.1 years, 18 females, 16 in each group) went to the final analysis, for which the results are shown. The participants were randomly assigned to one of two groups: the REAL group (anodal tDCS) or the SHAM group (sham stimulation). tDCS was administered during the second cycle of REM sleep for a duration of 15 minutes over the vmPFC.

Transcranial Direct Current Stimulation (tDCS)

tDCS was administered via a Starstim stimulator (Neuroelectrics, Barcelona, Spain) using five electrodes (1 cm radius each), with one anodal electrode and four cathodal electrodes positioned on the scalp and covered with conductive cream. ROAST modeling (Huang et al., 2019) optimized the electrode configuration to target the vmPFC, placing the anodal electrode over the nasion and cathodal electrodes at F7, F8, Ex19, and Ex20.

Real tDCS included a 30-second ramp-up, 15 minutes stimulation at 2 mA, and a 30-second ramp-down. This was delivered during the second and, if needed, third REM cycles, skipping the first cycle due to its short duration. For sham stimulation, a similar ramp-up and brief 30-second stimulation at 2 mA were applied, followed by ramp-down. EMLA cream was used to numb the skin under electrodes to avoid arousal due to sensations.

2.3. Fear Conditioning Protocol

A 3-day differential fear conditioning protocol adapted from Milad et al. (2007) was used. On Day 1, participants underwent fear acquisition by differentiating between a danger stimulus (CS+, paired with an electrical shock at a 62.5% reinforcement rate) and a safety stimulus (CS-), which was never paired with the shock. This phase included 32 trials (16 CS+ and 16 CS-).

The extinction phase on Day 2 involved presenting the CS+ without the shock in 16 trials (8 CS+ and 8 CS-), followed by a recall session on the same day and another on Day 3, both mirroring the extinction phase.

Each trial lasted for 37.5 seconds, beginning with a variable fixation cross, followed by an office scene where a desk lamp turned blue or yellow for 12 seconds, signaling the CS+/-. For the blue lamp, the unconditioned stimulus (US) was presented at the end of CS+ presentation with a calibrated intensity rated as "highly uncomfortable but not painful."

Stimuli were displayed using Presentation software, and the US was delivered via a ring electrode on the participant's right index finger using a constant current stimulator.

Skin conductance responses (SCR)

Regarding statistical analysis, we applied a mixed-model ANOVA separately in each phase of the experiment. For fear acquisition, 'stimulus' (CS+ or CS-), trial (time point of the trial) as within-subjects factors, and 'group' (tDCS or Sham) as between-subjects factor were entered in the analysis. For extinction, recall, and the second recall, 'block' (early extinction/recall/second recall, containing the first half of CS+/CS- trials and late extinction/recall/second recall, containing the second half of CS+/CS- trials),'stimulus' (CS+ or CS-), trial (time point of the trial) as within-subject factors and 'group' (tDCS or Sham) as between-subjects factors were entered in the analysis. A square root transformation (sqrt) was applied to normalize distributions. Least significant difference (LSD) post-hoc comparisons were applied in cases of significant results of the ANOVA.

Results

Acquisition

The analysis identified a significant main effect of the factor 'stimulus' ($F(1,30) = 69.366$, $p < .001$). SCR for CS+ ($M = .691$, $SE = .073$) was significantly larger compared to CS- ($M = .385$, $SE = .057$). Additionally, a significant main effect for the factor 'trial' was found ($F(6.928,207.52) = 4.756$, $p < .001$). No further statistically significant main effects and interactions were observed.

These findings indicate effective fear learning in both groups, and that no SCR differences appeared between groups during fear acquisition (*Figure 1*).

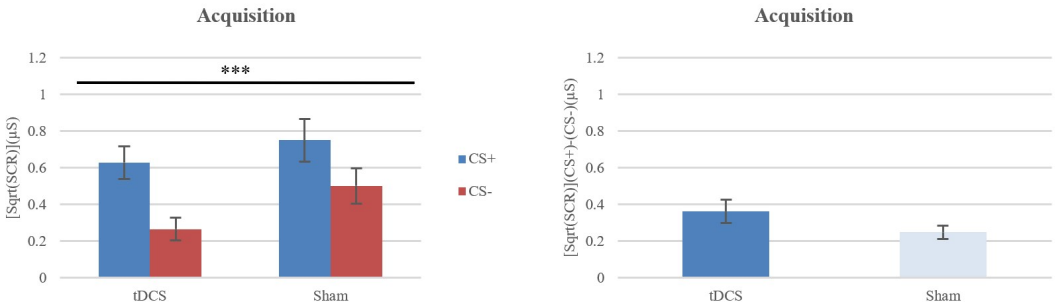

Figure 1. Skin conductance responses during acquisition. *Note*. The left chart shows CS+ versus CS- differences, while on the right, the differential score (CS+ minus CS-) is shown. Both groups show successful acquisition of fear responses. Least significant difference (LSD) post-hoc comparisons were applied to explore differences. *** p<0.001, ** p<0.01, * p<0.05; Error bars show standard error of mean.

Extinction

The analysis identified significant main effects of 'block' ($F(1,30)= 19.784$, $p < .001$), 'trial' ($F(3,30) = 4.637$, $p = .005$) and 'stimulus' ($F(1,30) = 20.085$, $p < .001$), as well as a significant 'block x stimulus' interaction ($F(1,30) = 21.460$, $< .001$). The SCR associated with CS+ ($M = .823$, $SE = .105$) was significantly higher compared to CS- ($M = .428$, $SE= .098$) in the early extinction ($p<.001$), but this difference was not significant ($p = .138$) in the late extinction phase, with CS+ ($M = .428$, $SE = .069$) and CS- ($M = .350$, $SE = .066$). No additional significant main effects or interactions were noted.

These findings indicate successful extinction in both groups, as shown by the reduced differences in responses to CS+ and CS- during late extinction, without any significant differences between groups in either the early or late phases of extinction (*Figure 2*).

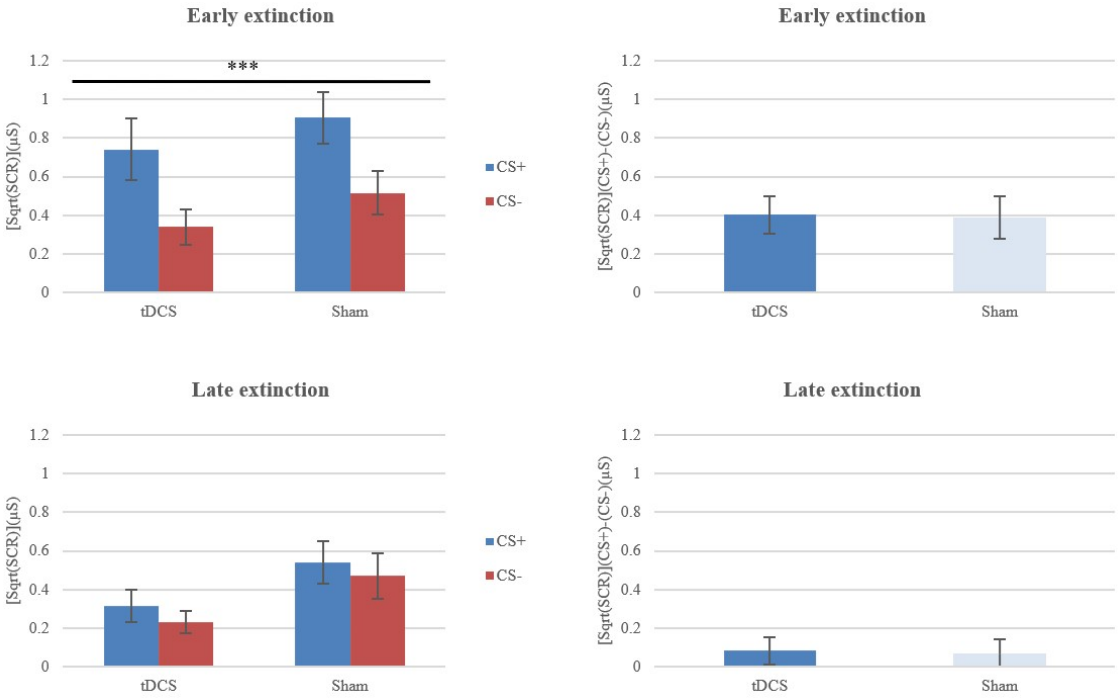

Figure 2. Skin conductance responses during early and late extinction. *Note.* The left chart shows CS+ versus CS- differences, while on the right chart, the differential score (CS+ minus CS-) is shown. Lack of CS+ vs. CS- differences during late extinction suggests that extinction took place. No differences between groups during early and late extinction were detected. Least significant difference (LSD) post-hoc comparisons were applied to explore differences. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$; Error bars show standard error of mean.

First recall

Significant main effects of 'block' ($F(1,30) = 25.290$, $p < .001$), 'trial' ($F(3,30) = 10.118$, $p < .001$) and 'stimulus' ($F(1,30) = 12.423$, $p = .001$) were observed, accompanied by significant 'block x trial' ($F(3,30) = 7.271$, $p < .001$) and 'block x stimulus' ($F(1,30) = 5.039$, $p = .032$) interactions. The SCR for CS+ ($M = .627$, $SE = .090$) was significantly higher compared to CS- ($M = .363$, $SE = .060$) in the early (first) recall ($p = .001$), whereas during the late (first) recall, no significant ($p = .144$) differences were observed between CS+ ($M = .341$, $SE = .075$) and CS- ($M = .259$, $SE = .055$). Furthermore, no differences emerged between intervention groups during both the early and late (first) recall phases. No further statistically significant main effects and interactions were observed.

A specific effect of tDCS on fear extinction memory consolidation was not detected during the first recall (*Figure 3*).

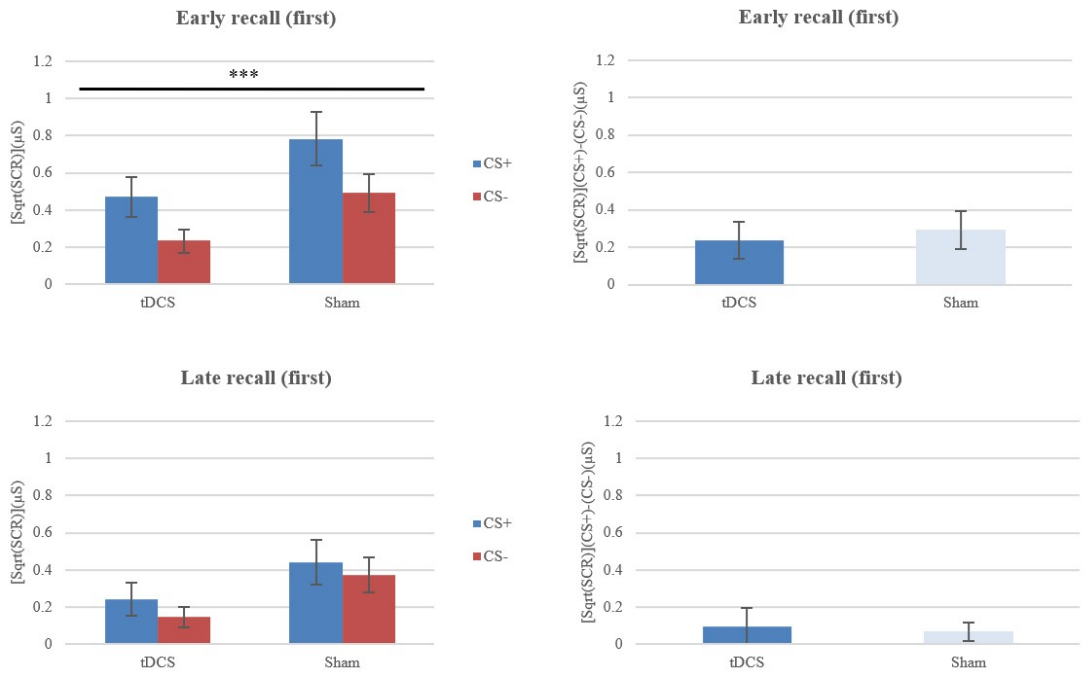

Figure 3. Skin conductance responses during early and late (first) recall. *Note.* The left chart shows CS+ versus CS- differences while on the right chart, the differential score (CS+ minus CS-) is shown. During early recall, both groups show significant CS+ vs. CS- differences, suggesting expression of fear responses, while during late recall, such a result is not observed. No differences between groups during early and late recall were detected. Least significant difference (LSD) post-hoc comparisons were applied to explore differences. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$; Error bars show standard error of mean.

Second recall

Significant main effects were identified for 'block' ($F(1,30) = 19.974$, $p < .001$), 'trial' ($F(2.311,69.330) = 10.310$, $p < .001$) and 'stimulus' ($F(1,30) = 18.363$, $p < .001$), along with significant 'block x stimulus' ($F(1,30) = 10.749$, $p = .003$) and 'block x trial' interactions ($F(3,90) = 3.094$, $p = .031$). Notably, a significant group x stimulus interaction emerged ($F(1,30) = 6.295$, $p = .018$).

Follow-up post-hoc tests examining intra-group differences (CS+ versus CS-) showed that the sham group had a significantly ($p < .001$) higher SCR for CS+ ($M = .763$, $SE = .102$) compared to CS- ($M = .517$, $SE = .079$), whereas the real tDCS group showed no significant ($p = .219$) difference between CS+ ($M = .211$, $SE = .102$) and CS- ($M = .147$, $SE = .079$). Furthermore, inter-group differences in SCR for CS+ and CS- were observed. The real tDCS group showed lower SCR toward the CS+ ($Mdif = .552$, $p < .001$) and CS- ($Mdif = .37$, $p = .002$) compared to the sham group. Additional post-hoc tests on differential scores (CS+ minus CS-) indicated a significantly lower SCR in the real tDCS group compared to the sham group ($Mdif = .182$, $p = .018$). No further statistically significant main effects and interactions were observed.

The results show that tDCS applied during REM sleep enhances fear extinction memory consolidation, with effects observable one day post-intervention (*Figure 4*).

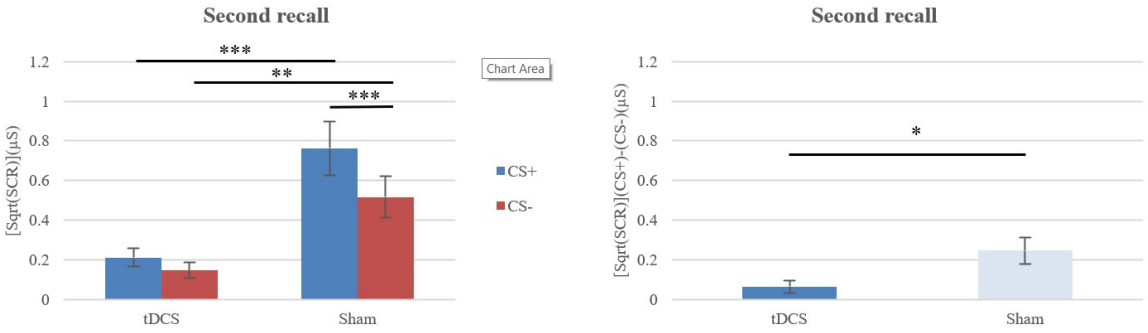

Figure 4. Skin conductance responses during the second recall. Note. The left chart shows CS+ versus CS- differences, while on the right chart, the differential score (CS+ minus CS-) is shown. During the second recall, the sham group showed significant CS+ versus CS- differences, suggesting the expression of fear responses, contrary to tDCS. Between-group comparisons reveal that the tDCS group shows a significantly lower SCR for CS+ and CS-, and furthermore, significantly lower SCR differential score. Results show enhancement of fear extinction memory consolidation with tDCS during REM sleep. Least significant difference (LSD) post-hoc comparisons were applied to explore differences. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$; Error bars show standard error of mean.

Conclusions

The findings lead to several key conclusions. First, the SCR results show successful fear learning and fear extinction in both groups and no differences between them in acquiring and extinguishing fear. Second, the impact of tDCS was not immediately apparent post-intervention. Third, and most crucially, **the effect of tDCS was evident a day later during the second recall, demonstrating that real tDCS effectively enhanced fear extinction memory consolidation during REM sleep.**

Experiment 2

Participants

We recruited a cohort of 30 healthy adults, aged between 18 and 40 years, for the fMRI phase of the study. The participants were randomly assigned to one of two groups: the REAL group (anodal tDCS) or the SHAM group (sham stimulation). tDCS was administered during the second cycle of REM sleep for a duration of 15 minutes over the vmPFC.

fMRI acquisition

MRI data were collected using a Siemens 3T MRI-system (Siemens Magnetom Trio TIM, Erlangen, Germany). Head movement was restricted using foam cushions (>2 mm, 2). T1-weighted images were acquired to improve normalization of the functional data, with a total of 176 slices at a thickness of 1 mm and in-plane resolution of 0.98 x 0.98 mm (TR = 1900 ms; TE = 2.52 ms; flip angle = 9; FOV = 250 x 250 mm^2). During fMRI participants viewed the back-projected stimuli via a 45° mirror placed atop the head coil. Task presentation and recording of behavioral responses were performed with Presentation 14.2 (NeuroBehavioral Systems, Inc.).

fMRI data analysis

The task-based fMRI data were processed using the FEAT module within the FSL toolbox (version 6.0.1). The preprocessing pipeline included several key steps: correction for head motion and slice timing, spatial smoothing using a Gaussian kernel with a fullwidth at half-maximum (FWHM) of 6 mm, and high-pass filtering with a cutoff of 50 seconds. Additionally, the images underwent linear registration to each participant's high-resolution T1-weighted anatomical image, followed by a linear registration to the Montreal Neurological Institute (MNI) standard space. For the first-level analysis, a general linear model (GLM) was employed to generate statistical maps representing functional activation for the contrasts incorporating data from all 16 presentations of both CS+ and CS-. Regressors (REAL; SHAM, CS+; CS-; REAL > SHAM; SHAM > REAL; CS+ > CS-; CS- > CS+) were convolved with the hemodynamic response function. Second-level analyses were conducted using random-effects estimation via FLAME (FMRIB's Local Analysis of Mixed Effects). The contrasts of interest were examined across the whole brain volume. To control for multiple comparisons, a cluster-based thresholding approach was applied with family-wise error (FWE) correction, setting a significance level of $p < .05$ and a Z-statistic threshold greater than 3.1.

Results

The primary outcomes measured were **skin conductance responses** (SCRs) and **patterns of brain activation** identified via fMRI during the extinction recall phase.

a) Skin Conductance Response (SCR)

The analysis of skin conductance responses (SCRs) to stimuli CS+ and CS- was conducted across three phases: Acquisition, Extinction, and Recall.

Acquisition Phase

During the acquisition phase, the analysis of variance (ANOVA) revealed a significant main effect of stimulus type, with participants exhibiting consistently greater skin conductance responses (SCRs) to the CS+ stimulus compared to the CS- ($F(1, 28) = 40.329$, $p < .001$). This strong differentiation in response was evident across both the REAL and SHAM groups, indicating that physiological responses were more pronounced for the CS+ than for the CS-. The interaction between stimulus type and group was not significant ($F(1, 28) = 1.960$, $p = .173$), suggesting that the difference in SCR responses between CS+ and CS- was comparable across the two groups. A significant main effect of time was also observed ($F(1, 28) = 9.799$, $p = .004$), reflecting a significant change in SCR responses over time, independent of stimulus type or group assignment. However, no significant interactions were found between time and group ($F(1, 28) = 0.005$, $p = .946$), stimulus type and time ($F(1, 28) = 0.040$, $p = .842$), or in the three-way interaction among stimulus type, time, and group ($F(1, 28) = 0.018$, $p = .896$). Additionally, the main effect of group was not significant ($F(1, 28) = 0.755$, $p = .392$), indicating no overall difference in SCR responses between the REAL and SHAM groups during this phase (see Fig. 1).

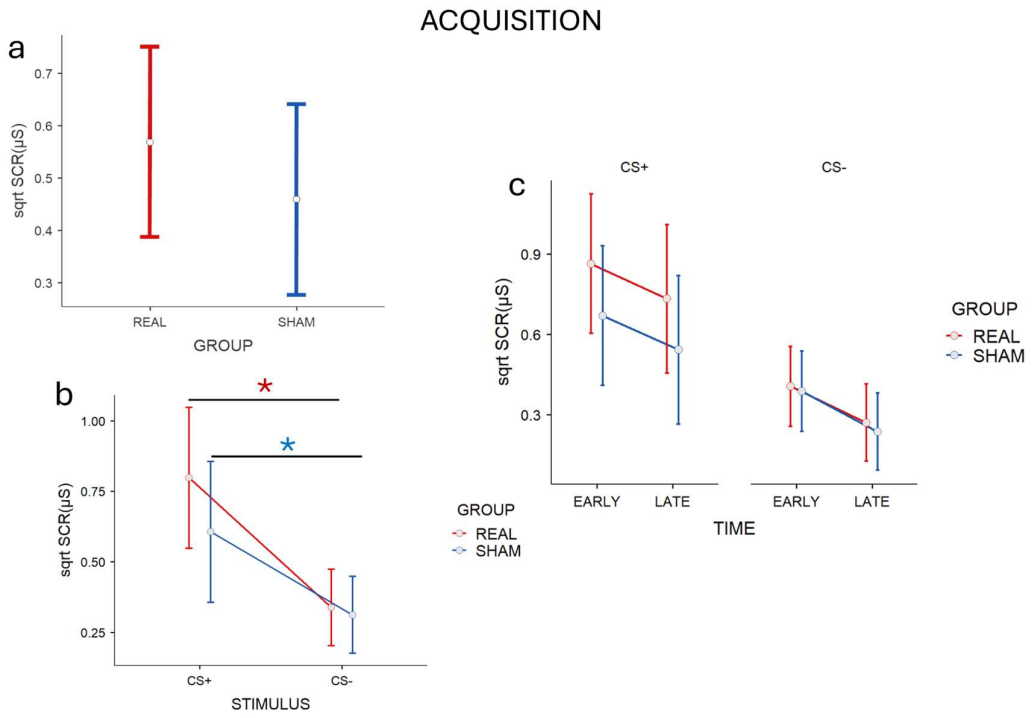

Fig. 1 SCR in Acquisition. ***a)*** The upper left panel shows the mean SCR for CS+ and CS- in both the REAL and SHAM groups, with no significant difference observed between groups. ***b)*** The lower left panel demonstrates a significant difference in SCR between CS+ and CS- in both the REAL (Mean Difference = 0.4601, SE = 0.0840, $t(28) = 5.480$, $p < .001$) and SHAM (Mean Difference = 0.4859, SE = 0.1387, $t(28) = 3.504$, $p = .008$) groups, with higher SCR for CS+ in both groups. ***c)*** The right panels display SCR changes over time for both CS+ and CS- in the REAL and SHAM groups.

Extinction Phase

During the extinction phase, the analysis revealed a significant main effect of stimulus type ($F(1, 28) = 13.913$, $p < .001$), with the CS+ stimulus eliciting a higher skin conductance response (SCR) than the CS- stimulus. A significant main effect of time was also found ($F(1, 28) = 18.860$, $p < .001$), indicating a general decrease in SCR from the early to late phases of extinction. Additionally, a significant interaction between stimulus type and time was observed ($F(1, 28) = 7.466$, $p = .011$), suggesting that the difference in SCR responses between CS+ and CS- stimuli varied over time. However, there was no significant main effect of group ($F(1, 28) = 0.012$, $p = .912$), indicating no overall difference in SCR responses between the REAL and SHAM groups. No significant interactions were detected between stimulus type and group, time and group, or the three-way interaction among stimulus type, time, and group (see Fig. 2).

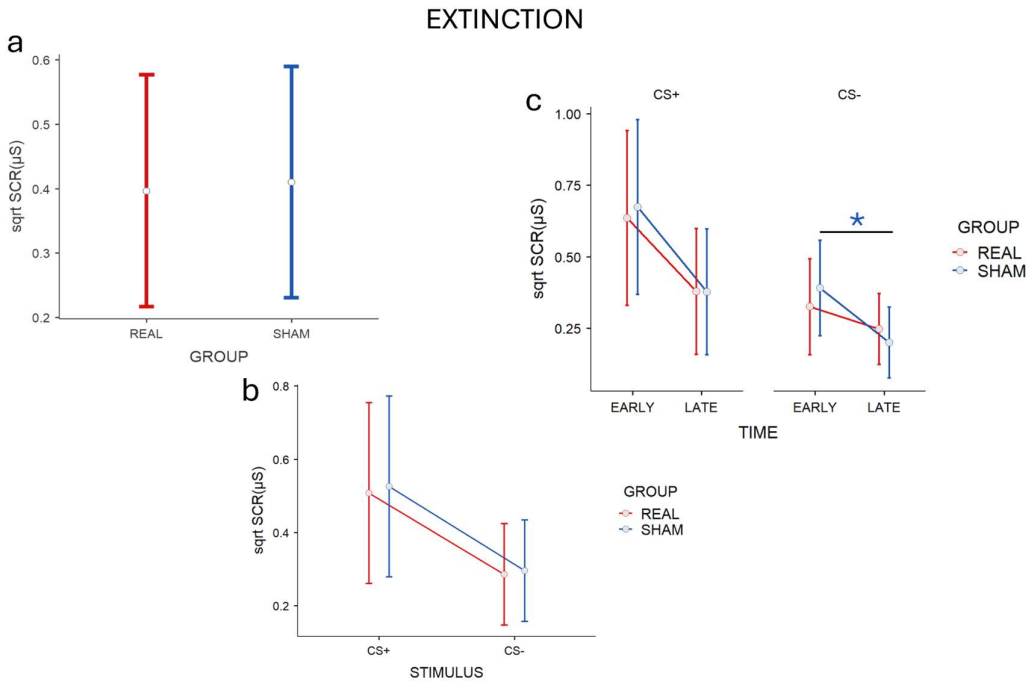

Fig. 2 SCR in Extinction a) In the top left panel, the mean sqrt SCR is displayed separately for the REAL and SHAM groups, showing no significant difference between the groups. b) The bottom left panel depicts the STIMULUS effect. c) The top right panel shows the effect of TIME on the sqrt SCR for CS+ and CS- stimuli, with a significant reduction in response from the Early to the Late phase in SHAM group (Mean difference = 0.2939, SE = 0.0840, $t(28) = 3.501$, $p = .008$).

Recall Phase

In the recall phase, the analysis demonstrated a significant main effect of time ($F(1, 28) = 15.95$, $p < .001$), indicating notable changes in SCR responses over time. A significant interaction between time and group was also found ($F(1, 28) = 10.28$, $p = .003$), suggesting that the evolution of SCR responses over time differed between the REAL and SHAM groups. The main effects for stimulus type ($F(1, 28) = 3.66$, $p = .066$) and the interaction between stimulus type and group ($F(1, 28) = 3.61$, $p = .068$, $\eta^2p = 0.114$) did not reach statistical significance, implying no significant differences in responses to CS+ and CS- stimuli between the two groups. Furthermore, no significant interactions were observed between stimulus type and time ($F(1, 28) = 2.73$, $p = .110$) or in the three-way interaction among stimulus type, time, and group ($F(1, 28) = 1.52$, $p = .228$). However, a significant between-subjects effect of group was detected ($F(1, 28) = 11.6$, $p = .002$), indicating overall differences in SCR responses between the REAL and SHAM groups during the recall phase (see Fig. 3).

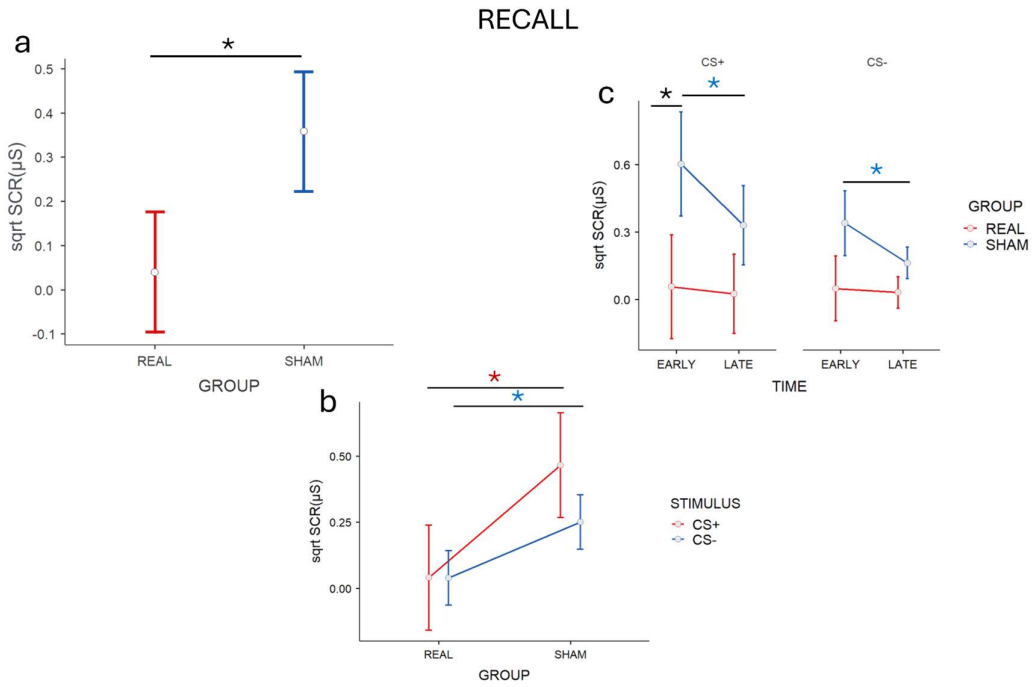

Fig. 3 SCR in Recall. a) The upper left panel illustrates overall group differences, where a significant effect of GROUP was observed (Mean difference = -0.318, SE = 0.0934), t (28) = -3.41, p = .002). ***b)*** The bottom left panel shows the interaction between STIMULUS and GROUP, indicating that the Sham group had a higher response to CS+ (Mean Difference = -0.426, SE = 0.1370), t (28) = -3.11, p = .021). and CS- (Mean Difference = -0.211, SE = 0.0711, t (28) = -2.97, p = .029). compared to the Real group. ***c)*** The right panels display time course data separately for CS+ and CS- stimuli, further detailing the interaction effects. During the early phase, the Sham group exhibit a higher response to CS+ compared to the Real group (Mean Difference = -0.546, SE = 0.1597, t (28) = -3.4207, p = .035).). Moreover, within the Sham group, a significant difference was observed between

early and late phases in both CS+ (Mean Difference = 0.2727, SE = 0.0533, t (28) = 5.1205, $p < .001$.) and CS- (Mean Difference = 0.3080, SE = 0.0783, t(28) = 3.9319, p = .014)

These findings suggest that overall differences in SCR responses between the REAL and SHAM groups were only evident during the recall phase. This suggests that tDCS has improved fear extinction consolidation, modulating physiological responses over time accordingly.

b) fMRI Findings

The fMRI analysis revealed distinct patterns of brain activation between the REAL and SHAM groups during the presentation of stimuli (CS+ and CS-) across different regressors during the Recall phase.

CS+ Presentation (REAL and SHAM)

During the presentation of the CS+ stimulus, significant differences in brain activation were found between the REAL and SHAM conditions. The REAL group showed extensive activation across several brain regions, including the occipital cortex (MNI coordinates: 28, -94, 4; $Z = 7.7$, $p < .05$), lingual gyrus (-23, -78, -9; $Z = 6.9$, $p < .05$), fusiform gyrus (-19, -85, -7; $Z = 6.5$, $p < .05$), insula (52, 15, -6; $Z = 5.2$, $p < .05$), angular gyrus (-50, -51, 35; $Z = 4.7$, $p < .05$), and frontal operculum (-52, 24, 3; $Z = 4.6$, $p < .05$), characterized by broad activation clusters. In contrast, the SHAM condition demonstrated significant activation in the calcarine cortex (MNI coordinates: -12, -100, 5; $Z = 6.8$, $p < .05$) and fusiform gyrus (24, -82, -12; $Z = 5.8$, $p < .05$) (see Fig.4).

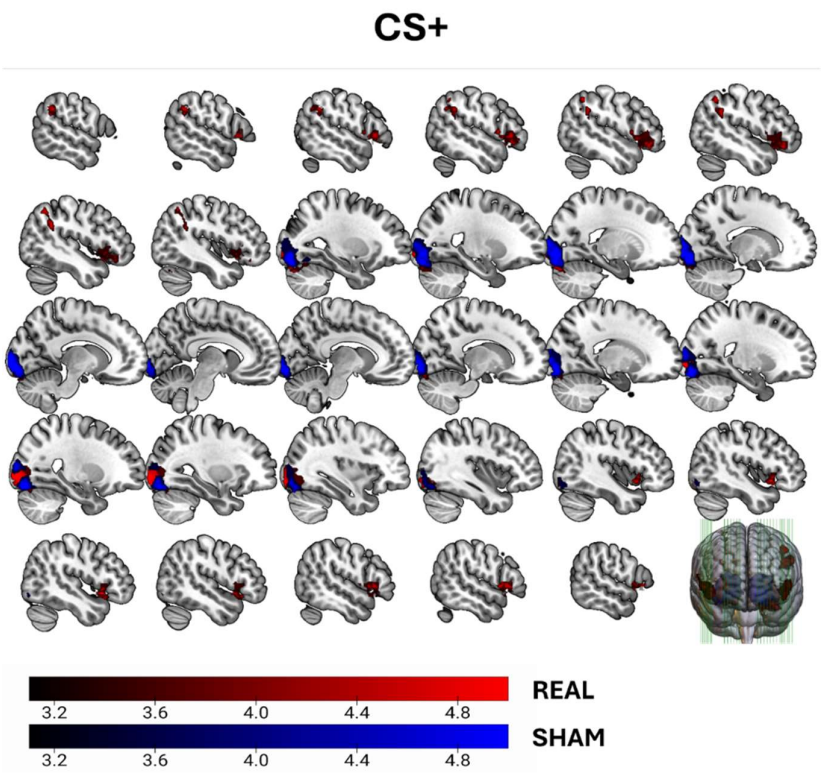

Fig. 4 CS+ Activation Clusters in REAL and SHAM. This figure presents sagittal views of significant activation clusters ($Z > 3.2$, $p < .05$, FWE-corrected) observed in the REAL (red) and SHAM (blue) conditions during CS+ presentation. The color bars represent the Z-scores ranging from 3.1 to 5.1.

CS+ Presentation (REAL > SHAM)

Comparing the REAL condition to the SHAM condition during CS+ presentation, the REAL group exhibited significantly increased activation, with the largest cluster in the right precentral gyrus extending into the postcentral gyrus and insula (MNI coordinates: 50.3, -0.1, 30.0; $Z = 5.8$, $p < .05$) over 30,742 voxels. Additional activations were seen in the right supplementary motor area (SMA) (5.4, 4.3, 70.6; $Z = 5.2$, $p < .05$), left SMA (-10.1, -0.1, 72.0; $Z = 5.6$, $p < .05$), right temporal pole (superior temporal gyrus, 47.4, 13.9, -12.0; $Z = 4.5$, $p < .05$), right precuneus (40.8, -10.5, 30.7; $Z = 5.8$, $p < .05$), right angular gyrus (-46.3, -48.1, 31.5; $Z = 4.7$, $p < .05$), right insula (51.1, 16.8, 2.4; $Z = 5.2$, $p < .05$), and frontal operculum (49.6, -8.3, 51.4; $Z = 5.1$, $p < .05$) (see Fig. 5).

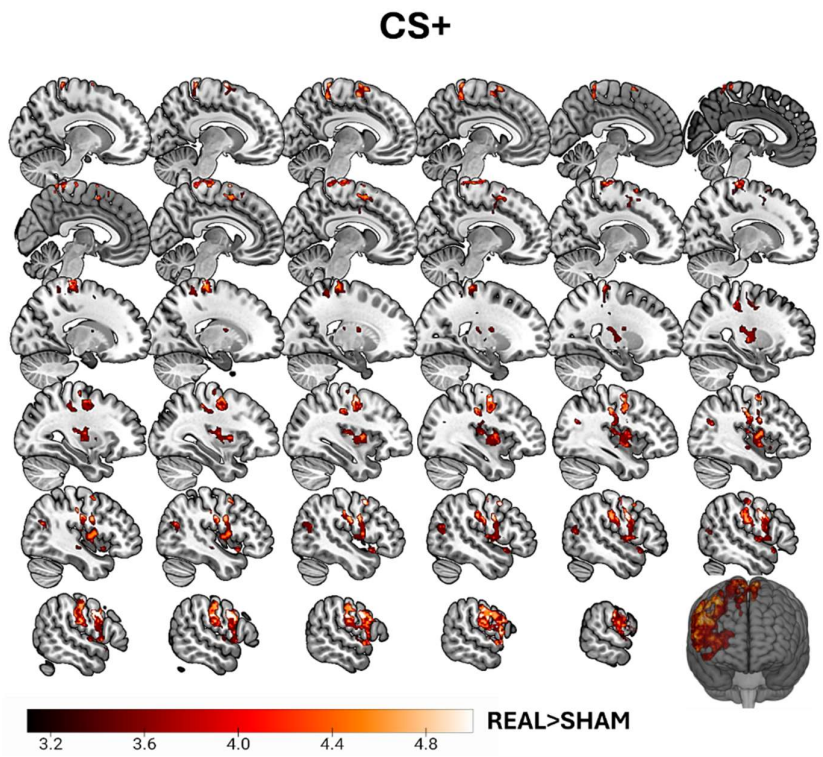

Fig. 5 CS+ activation clusters in REAL>SHAM. This figure presents sagittal views of significant activation clusters ($Z > 3.1$, $p < .05$, FWE-corrected) observed in the REAL>SHAM condition during CS+ presentation. The most prominent activations were found in the right Precentral gyrus, right Insula, right Supplementary Motor Area (SMA), and associated motor and sensory regions. The color bars represent the Z-scores ranging from 3.1 to 5.1.

CS- Presentation (REAL and SHAM)

For the CS- stimulus, the REAL group showed significant activation in the right superior occipital cortex (20, -94, 6; Zmax = 6.0, $p < .05$), extending into the fusiform gyrus and cerebellum, left middle occipital cortex (-22, -86, -0.2; Zmax = 6.0, $p < .05$), and right fusiform gyrus (42, -68, -19; Zmax = 3.8, $p < .05$). Conversely, the SHAM condition exhibited more extensive activation in the right lingual gyrus (22, -88, -16; Zmax = 9.3, $p < .05$), involving the occipital and fusiform cortices and cerebellum (see Fig. 6).

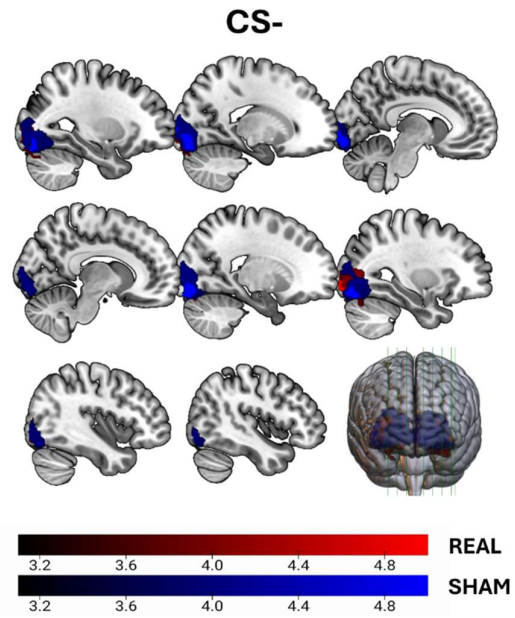

Fig. 6 CS- activation clusters in REAL and SHAM This figure presents sagittal views of significant activation clusters ($Z > 3.1$, $p < .05$, FWE-corrected) observed in REAL and SHAM during CS-. The REAL condition highlights significant activation in the right superior occipital cortex, left middle occipital cortex, and right fusiform gyrus). The SHAM condition shows extensive activation in the right lingual gyrus. The color bars represent the Z-scores ranging from 3.1 to 5.1.

CS+ vs. CS- (REAL)

Within the REAL group, comparing CS+ and CS- stimuli revealed increased activity in the right superior middle frontal gyrus (9.8, 33.8, 54.3; Zmax = 5.6, $p < .05$), left insula (39.6, 16.1, -6.1; Zmax = 4.6, $p < .05$), and left supplementary motor area (0.2, 19.8, 58.0; Zmax = 4.0, $p < .05$) (see Fig. 7).

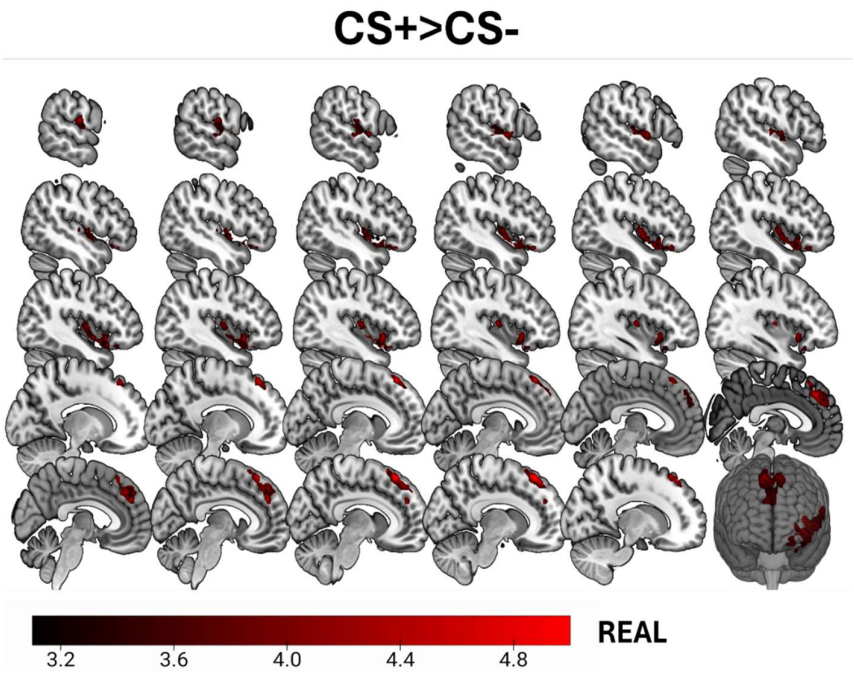

Fig 7 CS+>CS- activation clusters in REAL. This figure presents sagittal views of significant activation clusters ($Z > 3.1$, $p < .05$, FWE-corrected) observed in the right superior medial frontal gyrus, left insula, and left supplementary motor area. The color bars represent the Z-scores ranging from 3.1 to 5.1.

CS+ vs. CS- (REAL > SHAM)

In the REAL vs. SHAM comparison of CS+ to CS-, significant activation was noted in the right postcentral gyrus (62.1, -12.7, 27.8; Zmax = 4.7, $p < .05$), extending into the precentral gyrus and including parts of the supramarginal gyrus and the rolandic operculum (see Fig. 8).

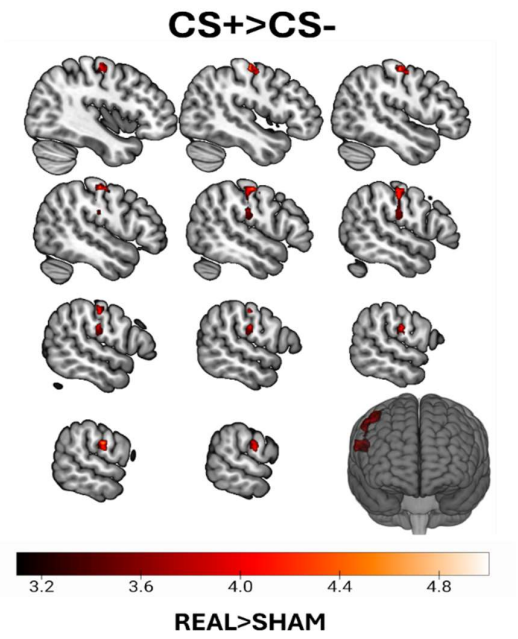

Fig.8 CS+>CS- activation clusters in REAL> SHAM This figure presents sagittal views of significant activation clusters (Z > 3.1, p < .05, FWE-corrected) observed in the right postcentral gyrus, extending to the precentral gyrus, supramarginal gyrus, and rolandic operculum The color bars represent the Z-scores ranging from 3.1 to 5.1

CS->CS+ (REAL and SHAM)

Analysis of the transition from CS- to CS+ in both conditions revealed significant activation in the REAL group in the right precuneus (12, -46, 41; Zmax = 4.6, p < .05), right middle occipital gyrus (33, -60, 33; Zmax = 4.6, p < .05), and right superior frontal gyrus (23, -8, 42; Zmax = 4.7, p < .05). The SHAM group showed activations in the right postcentral gyrus (37, -19, 37; Zmax = 5.9, p < .05), right middle occipital gyrus (35, -66, 16; Zmax = 5.2, p < .05), right lingual gyrus (5, -88, -9; Zmax = 5.1, p < .05), and left cuneus (3, -87, 20; Zmax = 4.5, p < .05) (see Fig. 9).

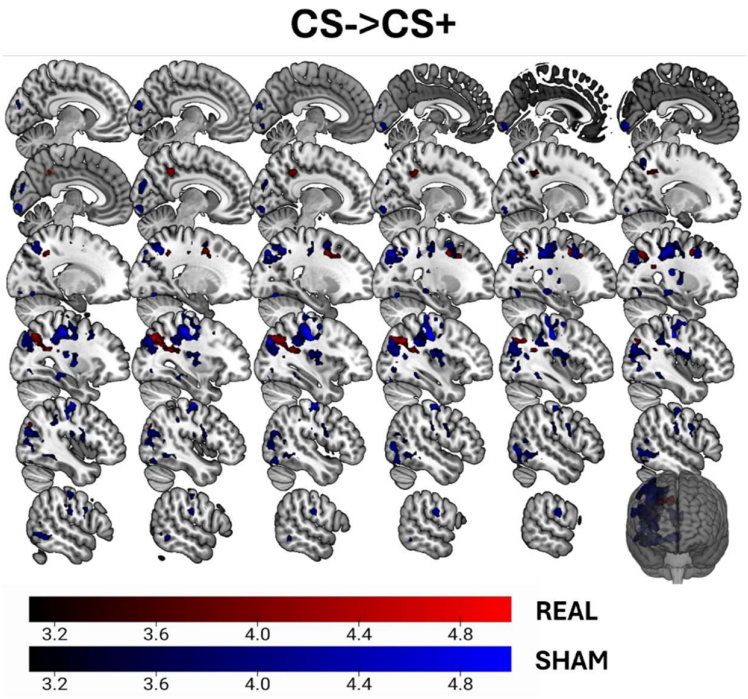

Fig. 9 CS->CS+ activation clusters in REAL and SHAM This figure presents sagittal views of significant activation clusters ($Z > 3.1$, $p < .05$, FWE-corrected) observed in the right precuneus, middle occipital gyrus, and superior frontal gyrus in the REAL condition (in red). The SHAM condition (in blue) shows significant activations in the right postcentral gyrus, middle occipital gyrus, lingual gyrus, and left cuneus. The color bars represent the Z-scores ranging from 3.1 to 5.1.

CS->CS+ (SHAM > REAL)

In the contrast of CS- to CS+ for the SHAM greater than REAL condition, increased activation was observed in the right precentral gyrus (45.9, -11.9, 58.0; $Z = 5.2$, $p < .05$), extending into the right postcentral gyrus and supramarginal gyrus, with additional clusters in the right insula (40.0, -2.4, 5.7; $Z = 4.6$, $p < .05$) and right putamen (28.2, -11.9, 2.0; $Z = 3.9$, $p < .05$) (see Fig. 10).

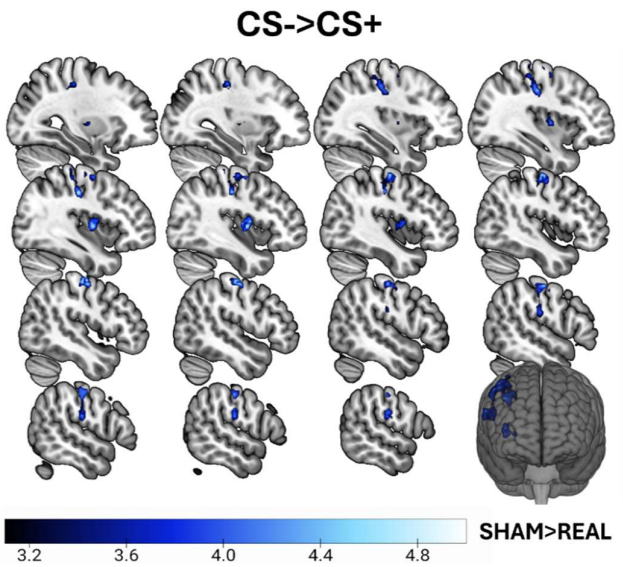

Fig. 10 CS->CS+ activation clusters in SHAM>REAL This figure presents sagittal views of significant activation clusters ($Z > 3.1$, $p < .05$, FWE-corrected) observed in the right precentral gyrus, right insula, and right putamen. The color bars represent the Z-scores ranging from 3.1 to 5.1.

Conclusion

The results show that tDCS delivered during REM sleep significantly enhanced fear extinction recall, as evidenced by a greater decrease in skin conductance responses (SCRs) to the conditioned stimulus (CS+) during the recall phase compared to the sham group. Furthermore, fMRI analyses revealed distinct patterns of brain activation between the tDCS and sham groups, with the tDCS group exhibiting greater activation in areas associated with motor control, sensory processing, and emotional regulation, particularly in the context of REM sleep. These findings suggest that tDCS may enhance the neural substrates involved in fear of extinction learning during REM sleep.

General conclusion

Overall, the reported findings suggest that PFC tDCS during REM sleep promotes fear extinction learning by enhancing respective neural substrates.

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