Offline rTMS inhibition of the bilateral dorsolateral prefrontal cortex impairs reappraisal efficacy

Miroslaw Wyczesany (m.wyczesany@uj.edu.pl)
Jagiellonian University
Agnieszka K Adamczyk
Jagiellonian University
Justyna Hobot
Jagiellonian University
Giansalvo Barbalinardo
Jagiellonian University
Przemysław Adamczyk
Jagiellonian University
Adam Labaza
Jagiellonian University
Tomasz S Ligeza
Jagiellonian University

Research Article

Keywords:

Posted Date: March 9th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1411756/v1

Offline rTMS inhibition of the bilateral dorsolateral prefrontal cortex impairs reappraisal efficacy

Miroslaw Wyczesany[1*], Agnieszka K Adamczyk[1,2], Justyna Hobot[1,3], Giansalvo Barbalinardo[1], Przemysław Adamczyk[1], Adam Labaza[1], Tomasz S Ligeza[1,4]

[1] Institute of Psychology, Jagiellonian University, Kraków, Poland

[2] Behavioural Science Institute, Radboud University, Nijmegen, The Netherlands

[3] Center of Functionally Integrative Neuroscience, Aarhus University, Aarhus, Denmark

[4] Department of Psychology, Northeastern University, Boston (MA), United States

[*] corresponding author at: Ingardena 6, PL-30060 Kraków; m.wyczesany@uj.edu.pl

ABSTRACT

In the study we verified the causal role of the dorsolateral prefrontal cortex in performing reappraisal strategy of emotional control. Subjects underwent three sessions of inhibitory offline theta burst TMS stimulation that was targeted at either left dorsolateral, right dorsolateral or vertex area. During the experimental task they were asked to either passively watch or reappraise affective pictures. Efficacy of emotional control was quantified using the late positivity potential of the evoked brain responses. After both left and right dorsolateral stimulation, reappraisal was compromised compared to the vertex session. Worsening of affective control was reflected in earlier but not later time windows of the late positivity. It is suggested that inhibitory stimulation primarily affected top-down mechanism of attentional deployment necessary to focus on the content of experimental stimuli that enables elaboration of new, less negative interpretation of emotional content.

Introduction

It is commonly agreed that inhibiting emotional responses using a wide range of affective control strategies is generally initiated and controlled by the prefrontal cortex (PFC). Indeed, existing data consistently point at engagement of PFC during both voluntary and automatic attempts to regulate emotions [1–7]. Reappraisal, which is a class of emotion regulation (ER) strategies based on reinterpretation of stimuli meaning, is typically used to down-regulate emotional responses to negative stimuli. As a result, negative feelings are decreased along with central and peripheral markers of emotional processing [2,8]. Reappraisal increases activations in several brain areas that can be considered parts of the regulatory network, mainly in the dorsolateral (DLPFC) and the ventrolateral PFC (VLPFC), but other regions, including the supplementary motor area (SMA), the dorsomedial PFC (DMPFC), and the anterior cingulate cortex (ACC), are also often reported [2,3,5,9]. The increased DLPFC/VLPFC activations that are observed during efficient down-regulation with reappraisal tend to be negatively correlated with the activity of brain regions associated with affective processing [9,10]. This effect is most consistently observed for the bilateral amygdala, while the ventral striatum and the left anterior insula are also often reported in this context [2,11,12]. Similarly, decreased activations can be seen in sensory cortices, which indicates that affective control affects early, sensory and attentional stages of information processing [13–15]. Scores of self-report measures of emotional responding have also been demonstrated to show a negative correlation with PFC activity [9]. Importantly, in affective disorders the pattern of activations described above is often disturbed, which suggests a problem with efficient recruitment of regulatory mechanisms in patients [16,17].

Existing data suggest that the DLPFC's contribution to reappraisal covers multiple cognitive domains. Directing attention to relevant information and maintaining and manipulating strategy-relevant content held in working memory (WM) are considered especially important during the first phase of reappraisal, when a new interpretation is generated [1,6,18]. Other types of function that are attributed to the DLPFC and are necessary to perform emotional regulation are essentially related to cognitive control [19–21]. They perform initiation of modulatory actions, maintenance of regulatory goals, and supervision over the whole process [5,7,22,23]. Last but not least, the DLPFC is claimed to be a source of multidimensional top-down influences (both direct or indirect) towards numerous brain regions, both cortical (including prefrontal, anterior cingulate, posterior attentional and visual areas) [24] and subcortical (amygdala, ventral striatum) [12,25].

The DLPFC serves multiple crucial functions whilst being richly functionally connected. It is not surprising that this region is widely acknowledged as playing a key role in reappraisal [26]. However, the evidence to support this claim is mostly correlational and hardly provides direct causal support.

Important experimental methods that allow the inference of causal properties of a brain region are transcranial magnetic stimulation (TMS) or direct current (electric) stimulation (DCS), both of which can either enhance or inhibit the activity of a selected region for minutes to hours following a single administration [27]. To our knowledge, only a few reports involving non-invasive brain stimulation have been published in which the effects of DLPFC stimulation on reappraisal efficacy were assessed. In the study of [28] two targets were selected, DLPFC and VLPFC, to examine the impact of transcranial DCS (tDCS) on reappraisal of negative pictures. However, contrary to the expectations, the DLPFC stimulation showed no effect compared to the sham condition. Only VLPFC stimulation resulted in a decrease of self-reported negativity of reappraised emotional pictures along with changes in cardiac reactivity measured by interbeat interval (IBI). In another experiment, the right DLPFC and VLPFC regions were stimulated using an offline 10Hz excitatory repetitive TMS (rTMS) protocol before participants were asked to use reappraisal to down-regulate their emotional responses to affective social stimuli [29]. The modulatory effects were quantified using the late positive potential (LPP; an ERP marker of the depth of emotional responses) and self-report scores. Stimulation of either of the areas brought an increase in ER efficacy compared to vertex stimulation. In another study, the right DLPFC was excited using anodal tDCS, which resulted in decreased skin-conductance responses and decreased emotional arousal ratings during downregulation of negative images when compared to sham conditions [30]. Finally, improved reappraisal efficacy along with cognitive control performance were reported after ten sessions of anodal (excitatory) tDCS targeted at the left DLPFC [31]. However, these results should be interpreted with caution as the effects were based on self-reports only and were carried out in a group of bipolar disorder patients. The aforementioned studies are generally consistent with theoretical expectations and supplement them with some causal argumentation; on the other hand, they have some limitations that still need to be addressed, like the specific groups that were recruited [31] or the application of bi-hemispheric stimulation, which simultaneously causes opposite effects for each hemisphere (either anodal/excitatory on the left and cathodal/inhibitory on the right or the opposite side), which makes the interpretation of the findings in terms of hemispheric specialization difficult. The latter issue remains important due to the well-known hemispheric differences of DLPFC in emotional processing [32,33], but much less recognized in the domain of emotional regulation.

Due to the scarcity of existing reports that can directly support the causal role of the DLPFC and consider hemispheric differences in the down-regulating of emotions by reappraisal, we designed this study to provide more data on this issue. We intended to observe the effects of a focal continuous Theta Burst Stimulation (cTBS) protocol delivered on either the left or right DLPFC on the efficiency of emotional downregulation in a classic reappraisal paradigm with presentation of affective pictures. cTBS was chosen because of its predominant inhibitory impact on cognitive performance when applied to the DLPFC [34]. Specifically, we aimed to assess the effects after separate left or right inhibition of the DLPFC in order to compare these conditions with control (vertex) stimulation in healthy participants. The reappraisal condition was limited to downregulation only as this can be

considered more ecologically valid and more closely related to everyday psychological functioning. Emotional responses were quantified using the Late Positive Potential (LPP) component, one of the best-validated EEG markers of emotional responding and regulation [35], which was supplemented with self-reports on the emotionality of the presented pictures.

As an experimental check, we expected to see the typical effect of reappraisal after the sham stimulation reflected in decreased LPP response and more positive subjective ratings after negative stimuli in reappraisal conditions. The main hypothesized experimental effect would be seen as decreased efficacy of reappraisal after inhibitory stimulation of either the L or R DLPFC compared to the sham condition. We expected this to be a lack of attenuation of LPP responses after the reappraisal of negative images. Additionally, some exploratory comparisons between the conditions of left and right inhibition of the DLPFC were planned, as the current literature data were insufficient to draw more directional hypotheses. Finally, as reports of more general alterations in bottom-up perceptual processes after rTMS stimulation of DLPFC are present in the literature [33], we could not rule out that such a nonspecific influence also affects LPP amplitudes in the passive watching trials. Therefore, such effects were also checked.

Results

Behavioral data

Fig. 1 shows the behavioral data results. Due to a technical issue, behavioral data from one TMS session were lost from one subject. Comparison of the NEG-WATCH and NEU-WATCH conditions revealed that participants reported more negative emotions when watching negative versus neutral pictures in the V session: $t(22) = -6.97$, $p < 0.0001$; $p_{FDR} = 0.0001$. As expected, comparison of the NEG-REAP vs. NEG-WATCH conditions in the V session revealed that participants reported less negative emotions in the reappraisal compared to the watch condition: $t(22) = -3.62$, $p < 0.002$; $p_{FDR} = 0.006$. Importantly, self-reported negative emotions did not differ as a function of session in the NEU-WATCH condition. This indicates that TMS did not affect emotional perception. Contrary to our predictions, comparison of differences in valence ratings (NEG-WATCH minus NEG-REAP) revealed no effect of TMS session.

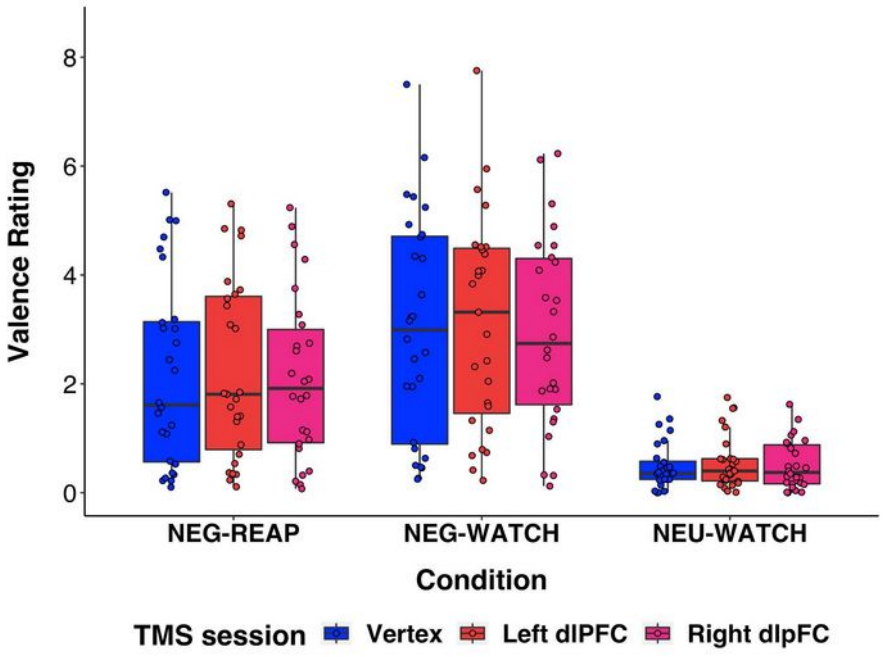

Figure 1

EEG data

Manipulation check

Our reappraisal manipulation was shown to be successful as a significantly lower LPP amplitude in NEG-REAP trials compared to NEG-WATCH trials in the V session was observed in the LPP2 time window over the CPz ($t(24) = -2.349$; $p < 0.027$; $p_{FDR} < 0.04$) and POz ($t(24) = 2.823$; $p < 0.009$; $p_{FDR}=0.03$) electrodes, and over POz electrode in the LPP3 ($t(24) = 2.561$; $p < 0.017$; $p_{FDR} < 0.05$) and LPP4 ($t(24) = 2.031$; $p < 0.053$; $p_{FDR} < 0.15$) time windows. However, the modulation in the latter time window did not survive the FDR correction and. No reappraisal effect was detected in the earliest LPP1 window.

The effect of TMS session on passive watching trials

Data analysis revealed that TMS did not influence basic perceptual processes as there were no effects of the TMS session on the WATCH-NEG or WATCH-NEU waveforms for any considered electrode and time window.

The detrimental effect of DLPFC inhibition on reappraisal efficacy

Confirming the main experimental hypothesis, comparison of NEG-WATCH minus NEG-REAP LPP amplitude differences between left/right DLPFC versus vertex TMS sessions demonstrated that inhibition of the right DLPFC, disrupted reappraisal-induced down-regulation of the LPP amplitude over the fronto-central and centro-parietal midline sites for the LPP2 (FCz: $t(49.5) = 2.26$; $p < 0.028$; $p_{FDR} < 0.028$; CPz: $t(50.2) = 2.56$; $p < 0.013$; $p_{FDR} < 0.019$) and LPP3 time-windows (FCz: $t(48.1) = 2.43$; $p < 0.019$; $p_{FDR} < 0.045$; CPz: $t(50.0) = 2.23$; $p < 0.030$; $p_{FDR} < 0.045$). Additionally, in the earliest LPP1 time window, marginally significant effects were observed at the fronto-central electrodes (FCz: $t(49.4) = 1.99$; $p < 0.05$; $p_{FDR} < 0.10$; CPz: $t(49.0) = 2.32$, $p < 0.02$, $p_{FDR} < 0.07$) for the R session, but it was considered beyond the significance threshold after the FDR correction. Interestingly, inhibition of the left DLPFC disrupted reappraisal-induced modulation more posteriorly: over the centro-parietal (CPz: $t(49.5) = 2.03$; $p < 0.046$; $p_{FDR} < 0.06$) and parieto-occipital (POz: $t(4.91) = 2.20$; $p < 0.032$; $p_{FDR} < 0.06$) electrodes in the LPP2 time window. However this modulation was only marginally significant after the FDR correction. In the LPP3 window, a close-to-significance effect was detected over the POz electrode, but this effect did not survive FDR correction ($t(48.3) = 1.95$; $p < 0.056$; $p_{FDR} < 0.15$). No effects of right or left DLPFC stimulation on LPP amplitude difference between WATCH-NEG and REAP-NEG trials were seen in the late LPP4 window. The EEG results are shown in Tab 1.

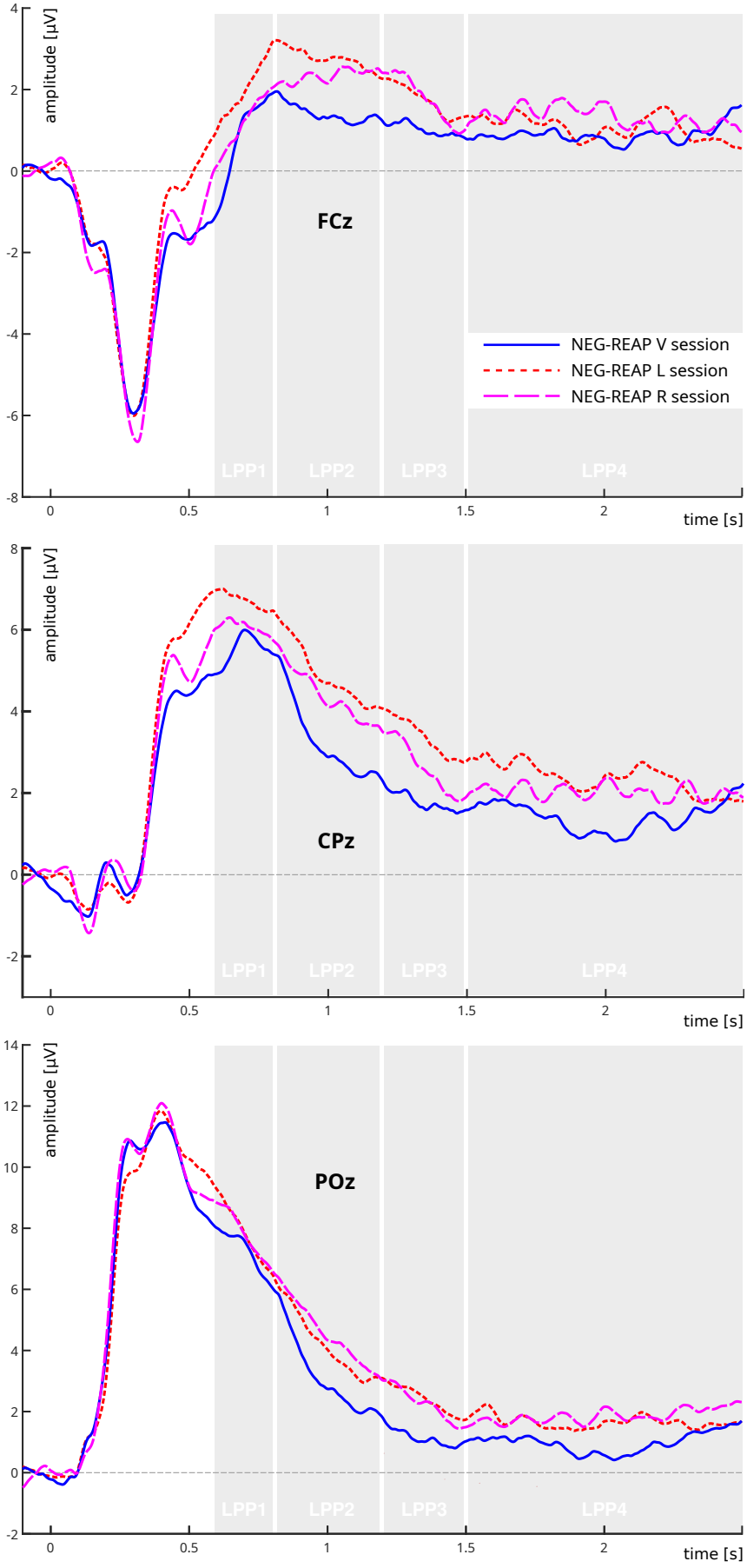
amplitude [µV]
FCz
NEG-REAP V session
NEG-REAP L session
NEG-REAP R session
LPP1
LPP2
LPP3
LPP4
time [s]
amplitude [µV]
CPz
LPP1
LPP2
LPP3
LPP4
time [s]
amplitude [µV]
POz
LPP1
LPP2
LPP3
LPP4
time [s]

Figure 2

Discussion

The study explored the effects of cTBS on either left or right DLPFC on reappraisal effectiveness. It was intended to verify whether inhibiting these areas compromises affective down-regulation, thus supporting a causal role of the DLPFC in reappraisal strategies. Firstly, as an experimental check, reappraisal effects were verified using EEG and behavioral data from the control (vertex) session. As expected, during reappraisal of negative images, the amplitude of the LPP component was decreased compared to the passive watching condition. Specifically, the effects were observed over centro-parietal and parieto-occipital electrode sites in the mid-early window, while at the parieto-occipital sites in the later time windows. Down-regulation of electrocortical responses was accompanied by a decrease in self-reported negative experience following reappraisal. Before determining the effects of cTBS/TMS on reappraisal efficiency, we verified whether the TMS affected basic perceptual processes and emotional responses in passive viewing conditions. This is important for the interpretation of the results regarding the main experimental hypothesis as it allows to determine whether emotion regulation effects are not partly driven by general alterations in perceptual processing. As the data analysis did not prove any influence of the TMS on the LPP component during passive watching conditions, we could assume that emotional perception remained relatively intact, at least regarding late evoked brain responses.

Primarily, our results confirmed the main experimental hypothesis: the inhibitory stimulation at both sides of the DLPFC resulted in decreased efficiency of reappraisal compared to the control (vertex) session. This effect was reflected in smaller differences of LPP amplitudes between reappraisal and watch trials after either right or left DLPFC inhibition compared to the control session. In other words, inhibition of the DLPFC resulted in impairment of affective down-regulation, as indicated by a lack of LPP attenuation in the reappraisal condition. As the *difference* between NEG-WATCH and NEG-REAP conditions was considered, we can conclude that this effect was driven by enhanced emotional processing of negative images during reappraisal rather than changes in basic perceptual processing, despite the stimuli valence. Our results replicate the findings of Zhao and colleagues [29], who demonstrated the enhancement of reappraisal effects after excitatory TMS stimulation applied over the right DLPFC. Importantly, our results also extend these findings by showing that reappraisal can be impaired after inhibitory stimulation of the left DLPFC.

Interestingly, inhibition of the left DLPFC resulted in *weaker* disruption of reappraisal effects than inhibition of the right DLPFC. The stimulation of the left and right DLPFC also affected reappraisal at different electrode sites, which was observed more posteriorly or more frontally, respectively. Although interpretation of this different scalp distribution can be challenging, some studies suggested distinct processes indexed by frontal and centro-parietal LPPs. While the centro-parietal LPP seems to reflect motivated attention or subjectively perceived stimulus significance [36], the

fronto-central LPP has been suggested to reflect enhanced attentional control processing [37,38] or increased cognitive effort [39] during reappraisal. Accordingly, it is possible that inhibition of the right and left DLPFC impacted the divergent cognitive processes that are necessary for the efficient execution of this strategy. Speculatively, given the engagement of the left DLPFC in linguistic processing [40], its inhibition could have impacted semantic elaboration, whereas inhibition of the right DLPFC could have compromised the attention allocation processes necessary for efficient reappraisal. This latter interpretation seems to be consistent with a study by Zwanzger and colleagues [33] which revealed enhanced attentional processing of *task-relevant* emotional stimuli after inhibitory stimulation to the right DLPFC. It is also in agreement with the data on the effects of DLPFC stimulation on bottom-up attention, which demonstrated that inhibition of the right (but usually not the left) DLPFC amplifies attentional processing of negative stimuli that are *task-irrelevant* (i.e. serve as distractors to another task) [32,41]. Nonetheless, the above speculation about the different processes that were affected by the TMS in our study should be treated with caution and could serve as an interesting direction for future studies rather than a warranted conclusion.

High temporal resolution of EEG recording enables us to comment on specific processes that were putatively disturbed by the TMS. To do so, we will now briefly consider the consecutive processes that underlie the successful implementation of reappraisal. One of the important prerequisites involves directing attention to important features of the examined scene in order to understand and semantically elaborate its meaning. After the stimulus is appraised, its initial negative interpretation must be maintained in working memory (WM). This interpretation can be then used as a template for the construction of an alternative, more neutral interpretation of the depicted scene. After the alternative reinterpretation is prepared, it must be implemented and its effectiveness tested. Thus, the following reappraisal stages involve (a potentially iterative process of) implementation and monitoring the effectiveness of the constructed re-interpretation(s). Finally, if the newly constructed interpretation is efficient at down-regulating negative experience, one must inhibit the initial interpretation to complete the reappraisal task successfully.

Although some session differences were already emerging in the earliest LPP1 time window, this should be treated with caution, as these effect did not reach the corrected significance threshold (see Fig. 2). Our analysis revealed that TMS mostly influenced the mid-early stages of reappraisal, while at the later stages the stimulation effects were again reduced (CPz, POz) or disappeared completely (FCz). These observations, along with the significant effects in the middle time windows (800 to 1500 ms), imply that TMS interfered with early/middle-latency reappraisal stages. Given the relatively early onset of the session effects, we suggest that a possible sub-function that was affected was top-down attention control. Our indication is supported by several pieces of evidence. Firstly, DLPFC remains active during both reappraisal and distraction emotional-regulation strategies, both of which involve specific requirements for attentional deployment: while reappraisal requires directing attention toward the affective content, distraction involves diverting attention away from the emotional

scene, despite bottom-up attraction by high-arousal content [42]. Secondly, the functional significance of early LPP (up to approximately 1000 ms) is associated with attention allocation, while later time windows are more often linked with memory and reinterpretation stages [38]. Thus, the TMS effect could be interpreted as a disturbance in the efficient allocation of top-down attention to the presented stimuli that is necessary to process the presented scene in order to understand its meaning, and prepare a new, less negative interpretation. Finally, this interpretation is further supported by the results of Vierheilig and colleagues [43], who observed specific attentional task-related enhancement of LPP amplitude after right anodal (excitatory) tDCS of the DLPFC, but more general modulation of emotional perception during a passive viewing task was not observed.

The role of working memory (WM) processes in the interpretation of our results requires some consideration. It is engaged during both early and late stages, however the WM load seemingly increases in the later phase due to the maintenance of new interpretation that has to neutralize stimulus negativity [44]. Indeed, the relationships between WM efficiency and reappraisal success have been reported [45,46]. As our effects of TMS diminished in the later LPP time windows, the interpretation which indicates the disruption of WM seems less plausible. However, taking into account that the DLPFC is a widely established neural substrate for WM [47] and that reappraisal consistently activates brain regions implicated in the WM [2,9], we encourage future studies to design an experiment that would be optimized to disentangle the unique contribution of this region to reappraisal efficacy.

Despite the observed modulation of electrocortical responses after TMS, we failed to observe the effects of session at the behavioral level. A possible reason for this discrepancy is that the effects of stimulation could have been too subtle to be detected at the subjective experience level. Although the impact of affective downregulation on self-report is widely reported in typical reappraisal procedures, it is often not observed in paradigms where the modulatory goal is implicit and remains undisclosed. In such cases, similarly to our data, self-report ratings tend not to differ even though brain markers or physiological markers show attenuation of affective processing [7,48,49]. Additionally, it has been reported that even vertex stimulation that is often considered as a control condition can impact the estimation of affective stimuli, which raises further doubts regarding the reliability of the first-person methods in our (and similar) procedures [50].

To summarize, our study showed that selective inhibition of either the left or right DLPFC can disrupt reappraisal efficacy, as evidenced by the reduced attenuation of LPP responses to negative pictures in the reappraisal versus passive watch condition. Our study offers a novel contribution by demonstrating that stimulation of not only the right but also the left DLPFC can impact reappraisal, starting from its earlier stages. We point at attentional deployment as a potential source of these disturbances. Moreover, our results tentatively suggest that inhibition of the left and right DLPFC might bring about distinct effects, thus implicating the disruption of different cognitive processes. However, this possibility warrants further investigation and replication.

Finally, some advantages and possible drawbacks of our design should be discussed. The paradigm used here was based on inhibiting rather than boosting a single neural area during the performance of a specific cognitive task. As such, it could be conducted on group of healthy participants, thus allowing for an interpretation of results that is similar to well-controlled temporary 'loss of function' or lesion studies, which enable more straightforward interpretation of results [51]. Moreover, contrary to the majority of previous studies, we used a repeated-measures design, which provided us with higher power to detect even subtle stimulation effects. Regarding the limitations, it should be remembered that although TMS allows for the selective disruption of activity in a unilateral brain region, if the specific cognitive function is distributed bilaterally, the remaining neural activity can potentially 'take over' and account for the less-pronounced activation in its mirror structure. As meta-analytic results suggest the bilateral involvement of the DLPFC in reappraisal, this might be one of the reasons why our effects were limited to early/middle-latency time windows and why some of the effects were only marginally significant. Also, although we tested a moderate size sample of participants in our study, our sample size did not deviate significantly from those used in the TMS/tDCS studies mentioned previously. Finally, we used the same pictorial stimuli throughout all three TMS sessions. Although this design choice facilitated between-session comparison and controlling for both low- and high-level stimulus features, it also ran the risk of observing weaker emotion-induction effects (or stronger emotion regulation effects) due to repeated exposure to the same images. Finally, our choice of TMS targets, although based on meta-analytical works, could not fully cover the spatial extent of the DLPFC areas reported in the reappraisal literature. Thus, we advise future studies to replicate our findings, especially exploring slightly modified targets and differently designed stimuli material.

Materials and methods

Participants

The experimental procedure was compliant with the directives of the Helsinki Declaration (1975, revised 2000) and was approved by the The Jagiellonian University Institute of Psychology Research Ethics Committee. Informed consent was obtained from all participants. 26 volunteers participated in the study (mean age 25.4, SD 3.5; 15 females, 10 males, 1 other gender). A similar sample size was used in previous studies that investigated the effect of TMS stimulation on emotion regulation [29]. Data from two subjects were discarded due to technical malfunctions during recording. The reimbursement paid was €40.

Pictorial material

A total of 90 pictures were selected from the Nencki Affective Pictures System (NAPS) [52] and the International Affective Picture System (IAPS) [53]: 45 were neutral (food, landscapes, households, neutral animals, people in daily activities) and 90 were negative (disgusting food, sad people, accidents, violent images, animal mutilations, surgical procedures). We attempted to match unpleasant and neutral stimuli in terms of both color content and complexity (e.g., number of faces, number of body parts, etc.). Normative ratings of the pictures (pulled from normalized NAPS and IAPS values) were as follows: for neutral pictures, mean valence 4.99; SD 1.07, mean arousal 4.94; SD 1.13; for negative pictures, mean valence 2.89, SD 1.29, mean arousal 6.93; SD 1.42. The neutral and negative sets differed in their normative ratings of valence and arousal ($p < 0.01$). Another 21 negative pictures were selected to enable reappraisal training before the experimental session. To facilitate comparisons, the same pictures were used for each TMS session, but it was ensured that negative stimuli were always associated with the same (either WATCH or REAP) instruction.

Behavioral Procedure

An overview of an experimental session is provided in Fig. 3. The study applied a reappraisal paradigm and stimuli presentation timing similar to Ochsner et al. [54]. All except one participant underwent three rTMS sessions on separate days in counterbalanced order, with either left DLPFC, right DLPFC or sham (vertex) stimulation, further referred to as L, R, V sessions, respectively. A single experimental session lasted for ~60-80 mins, of which ~25 min was the experimental task itself (the first session was longer, as it included determination of individual RMT; see TMS procedure). Upon arrival, a brief description of the procedure and TMS stimulation was provided and participants were asked to give informed consent and complete a qualification safety questionnaire. Before the initial experimental session, participants underwent reappraisal training. As reappraisal is an attention engagement strategy [55], we used situation-focused reappraisal to distinguish it from other strategies that involve attention disengagement [54,56]. Using sample stimuli, participants were taught to find and apply new interpretations of emotional scenes that were related to more positive outcomes. The subjects' ability to reappraise according to the instructions was verified by the experimenter. Directly before the experimental task, subjects underwent a TMS stimulatory session. The task was presented in full-screen mode on an LED screen (62 cm diameter, viewing distance of 60 cm approx). The task consisted of 90 trials in pseudo-randomized order, with a half-minute break after the 30th and 60th presentation. Each trial started with the presentation of a cue (3 sec) that could be either 'WATCH' or 'REAPPRAISE', which meant passively watching pictures vs using the previously trained reappraisal strategy, respectively. Then a fixation cross was shown (2 sec), followed by a picture (2.5 sec). Trials concluded with the subjective rating of the negative affective experience on a continuous visual analogue scale (until response); this was followed by a blank screen before the next trial (3 sec) (Fig.

3B). Summarizing the design of the experiment, there were 3 conditions (depending on picture valence and instruction cue, i.e. NEU-WATCH, NEG-WATCH, NEG-REAP) and 3 TMS sessions (L, R, V).

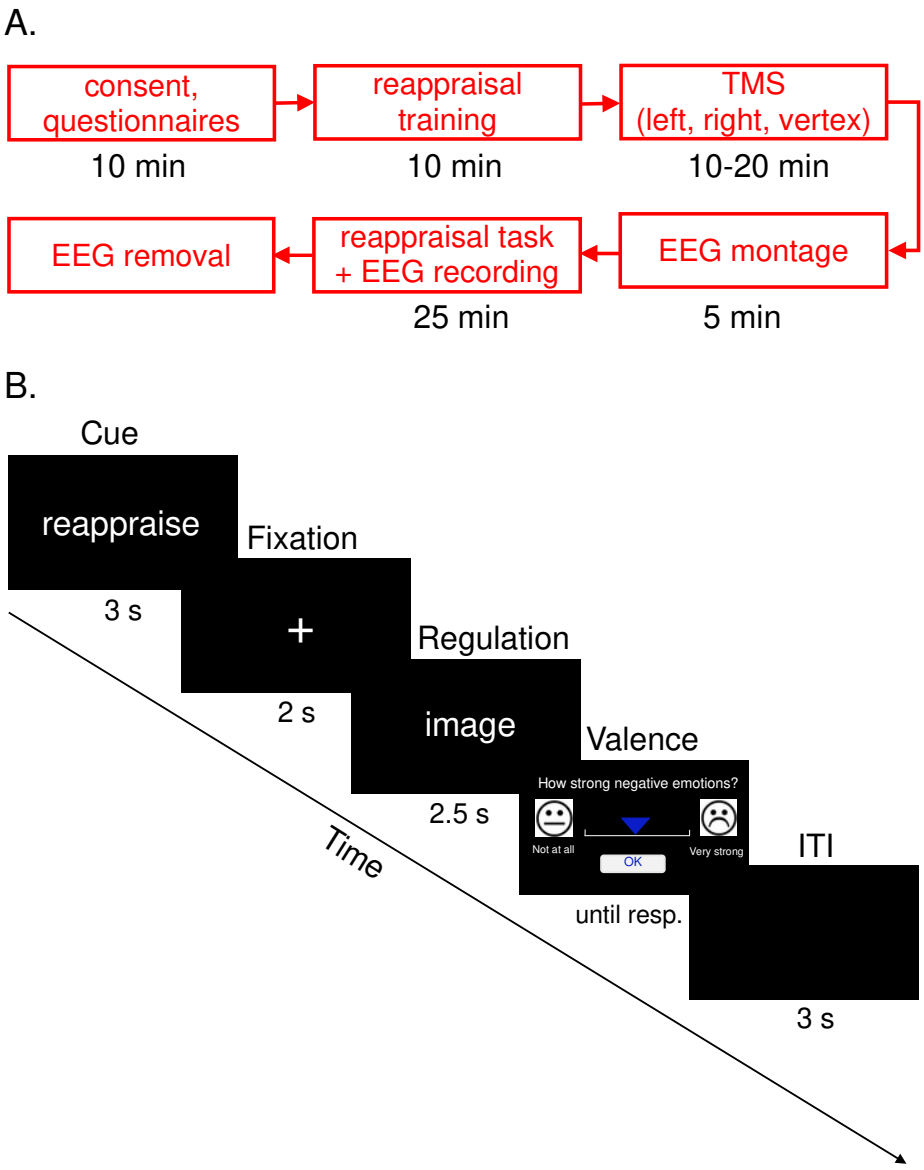

Figure 3

TMS Procedure

Biphasic TMS (~ 400 µs) was delivered with a Magstim Super Rapid2 Plus1 stimulator using a 70 mm figure-of-eight air-cooled Double Air Film Coil. For the purpose of resting motor threshold (RMT) estimation, the electromyographic signal was recorded with a Brainsight unit from the first

dorsal interosseous (FDI) muscle of the right index finger during the motor threshold (MT) estimation. In the first experimental session, the individual RMT was estimated. Stimulation started from applying 30% of MSO single-pulse TMS to the left primary motor cortex and adjusting the stimulation intensity. Then, by varying the stimulation intensity, the site where the suprathreshold TMS induced the maximal twitch in the right index FDI muscle was established. Subsequently, the lowest intensity that resulted in a motor-evoked potential (MEP) of more than 50 μV peak-to-peak amplitude in 5 out of 10 consecutive trials was determined. The stimulation sites were localized with the Brainsight 2.3 neuronavigation software (Rogue Research Inc.) using individual structural MRI images. Each participant's brain was transformed into standard MNI space using Brainsight software. Depending on the session, TMS was applied to either the left DLPFC (middle frontal gyrus; MNI coordinates: 51, 15, 48; L session), the right DLPFC (middle frontal gyrus; -33, 3, 54; R session), or vertex (0, -28, 90), with the coil handle pointing backward. The DLPFC stimulation sites were selected based on reappraisal meta analysis of [2]. The site of stimulation and the tangential position of the coil in relation to the scalp were ensured by using the neuronavigation system. Throughout the RMT determination procedure and subsequent application of cTBS to the DLPFC, the coil was held at a 45° angle off the midline, with the handle pointing down, while for the vertex stimulation the coil remained untitled. The current induced in the brain was PA–AP. During the cTBS stimulation, participants kept their eyes open. The cTBS protocol had a conventional pattern and duration, i.e. 3-pulse bursts at 50 Hz were applied at 5 Hz for 40 seconds [57]. The cTBS was delivered at 80% of the individual RMT and the average intensity equalled 61.1% (SD 12.2) of the maximal stimulator output (MSO). Participants wore earplugs for noise protection throughout the duration of TMS.

EEG recording and analysis

The EEG signal was recorded using a BiosemiTwo device and a set of 64 cap electrodes, supplemented by two mastoid and four oculomotor electrodes. Data from two participants had to be discarded due to technical problems that arose during the procedure. The EEG signal was processed using FieldTrip-based custom routines. The following steps were applied: reference to the linked mastoid; 0.2 Hz high-pass and 46 Hz low-pass filtering; bad channel screening using trimmed variance extreme values; eye-movement correction with the recursive least square (RLS) method [58]; segmentation relative to stimulus onset in the -0.1 to 2.5 sec window; baseline correction using the prestimulus period; either interpolation of bad channels or whole trial removal (depending on the number of electrodes affected). Range (100 μV threshold), excess variance or muscle signal contamination (detected by spectral power value for frequencies >35 Hz) artifacts were finally removed, and the remaining trials (78.9 on average) were averaged across conditions. LPP magnitude was quantified as the average value within the following time windows (LPP1: 600-800 ms, LPP2: 800-1200 ms, LPP3: 1200-1500 ms, LPP4: 1500-2500 ms), which were selected based on examination of a collapsed localizer (i.e., a grand average waveform collapsed across conditions) [59]. As previous

studies have localized reappraisal effects over fronto-central, centro-parietal and parieto-occipital regions, mean LPP amplitudes were measured from electrodes FCz, CPz, POz.

Statistical analysis

Statistical analyses were performed by fitting a series of linear mixed-effects (LME) models using the lme4 package, and the lmerTest library was used to estimate related p-values. Each model included condition as a fixed effect and a participant-specific intercept. Resulted p-values were subjected to additional False Discovery Rate (FDR) correction. LME modeling was used to account for missing data, while the *lmerTest* package applied the Satterthwaite method for estimating degrees of freedom.

As a manipulation check for emotional regulation, we compared NEG-REAP vs. NEG-WATCH data in the V session, which was considered a session without any DLPFC stimulation. To further verify that the TMS stimulation did not alter perceptual processes *per se*, which would affect the interpretation of the results, we also determined the effect of the stimulatory session on the NEU-WATCH waveforms. Moreover, to determine whether emotional responding itself could be affected by stimulation, we checked the effect of session on NEG-WATCH LPP amplitudes, which reflects the overall amount of resources captured by the processing of emotional stimuli and the intensity of emotional response. Crucially, to verify that the TMS stimulation affected emotion regulation, we compared the session's effect on the difference between the NEG-REAP and the NEG-WATCH conditions. We considered the differences between the watch and reappraisal conditions instead of absolute LPP amplitudes to minimize the possible confounding effects of more general post-TMS alterations of perceptual or emotional processing that could still be present, even when not detected by previously described tests.

References

1. Braunstein, L. M., Gross, J. J. & Ochsner, K. N. Explicit and implicit emotion regulation: a multi-level framework. *Soc. Cogn. Affect. Neurosci.* **12**, 1545–1557 (2017).
2. Buhle, J. T. *et al.* Cognitive Reappraisal of Emotion: A Meta-Analysis of Human Neuroimaging Studies. *Cereb. Cortex* **24**, 2981–2990 (2014).
3. Etkin, A., Büchel, C. & Gross, J. J. The neural bases of emotion regulation. *Nat. Rev. Neurosci.* **16**, 693–700 (2015).

4. Frank, D. W. *et al.* Emotion regulation: quantitative meta-analysis of functional activation and deactivation. *Neurosci. Biobehav. Rev.* **45**, 202–211 (2014).
5. Kohn, N. *et al.* Neural network of cognitive emotion regulation — An ALE meta-analysis and MACM analysis. *NeuroImage* **87**, 345–355 (2014).
6. Ochsner, K. N., Silvers, J. A. & Buhle, J. T. Functional imaging studies of emotion regulation: a synthetic review and evolving model of the cognitive control of emotion. *Ann. N. Y. Acad. Sci.* **1251**, E1–E24 (2012).
7. Wyczesany, M., Adamczyk, A. K., Ligeza, T. S., Bereś, A. & Marchewka, A. Implicit induction of emotional control—A comparative fMRI investigation of self-control and reappraisal goal pursuit. *Emotion* **21**, 1379–1391 (2021).
8. Fuentes-Sánchez, N., Jaén, I., Escrig, M. A., Lucas, I. & Pastor, M. C. Cognitive reappraisal during unpleasant picture processing: Subjective self-report and peripheral physiology. *Psychophysiology* **0**, e13372 (2019).
9. Berboth, S. & Morawetz, C. Amygdala-prefrontal connectivity during emotion regulation: A meta-analysis of psychophysiological interactions. *Neuropsychologia* **153**, 107767 (2021).
10. Wang, H.-Y. *et al.* Neural mechanisms of implicit cognitive reappraisal: preceding descriptions alter emotional response to unpleasant images. *Neuroscience* **347**, 65–75 (2017).
11. Staudinger, M. R., Erk, S., Abler, B. & Walter, H. Cognitive reappraisal modulates expected value and prediction error encoding in the ventral striatum. *Neuroimage* **47**, 713–721 (2009).
12. Urry, H. L. *et al.* Amygdala and Ventromedial Prefrontal Cortex Are Inversely Coupled during Regulation of Negative Affect and Predict the Diurnal Pattern of Cortisol Secretion among Older Adults. *J. Neurosci.* **26**, 4415–4425 (2006).
13. Adam, R., Schönfelder, S., Forneck, J. & Wessa, M. Regulating the blink: Cognitive reappraisal modulates attention. *Front. Psychol.* **5**, 143 (2014).
14. Wessing, I. *et al.* Cognitive emotion regulation in children: Reappraisal of emotional faces modulates neural source activity in a frontoparietal network. *Dev. Cogn. Neurosci.* **13**, 1–10 (2015).
15. Wyczesany, M., Ligęza, T., Tymorek, A. & Adamczyk, A. The influence of mood on visual perception of neutral material. *Acta Neurobiol. Exp. (Warsz.)* **78**, (2018).

16. Ferdek, M. A., Adamczyk, A. K., van Rijn, C. M., Oosterman, J. M. & Wyczesany, M. Pain catastrophizing is associated with altered EEG effective connectivity during pain-related mental imagery. *Acta Neurobiol. Exp. (Warsz.)* **79**, 53 (2019).
17. Zilverstand, A., Parvaz, M. A. & Goldstein, R. Z. Neuroimaging cognitive reappraisal in clinical populations to define neural targets for enhancing emotion regulation. A systematic review. *NeuroImage* **151**, 105–116 (2017).
18. Morawetz, C., Bode, S., Derntl, B. & Heekeren, H. R. The effect of strategies, goals and stimulus material on the neural mechanisms of emotion regulation: A meta-analysis of fMRI studies. *Neurosci. Biobehav. Rev.* **72**, 111–128 (2017).
19. Hendricks, M. A. & Buchanan, T. W. Individual differences in cognitive control processes and their relationship to emotion regulation. *Cogn. Emot.* **30**, 912–924 (2016).
20. Mincic, A. M. Neural substrate of the cognitive and emotional interference processing in healthy adolescents. *Acta Neurobiol. Exp. (Warsz.)* **70**, 406–422 (2010).
21. Ochsner, K. N. & Gross, J. J. Cognitive emotion regulation. *Curr. Dir. Psychol. Sci.* **17**, 153–158 (2008).
22. Morawetz, C., Bode, S., Baudewig, J., Kirilina, E. & Heekeren, H. R. Changes in effective connectivity between dorsal and ventral prefrontal regions moderate emotion regulation. *Cereb. Cortex* **26**, 1923–1937 (2016).
23. Petrides, M. Lateral prefrontal cortex: architectonic and functional organization | Philosophical Transactions of the Royal Society B: Biological Sciences. *Philos. Trans. R. Soc. B* **360**, 781–795 (2005).
24. Ligeza, T. S., Wyczesany, M., Tymorek, A. D. & Kamiński, M. Interactions Between the Prefrontal Cortex and Attentional Systems During Volitional Affective Regulation: An Effective Connectivity Reappraisal Study. *Brain Topogr.* **29**, 253–261 (2016).
25. Quirk, G. J. & Beer, J. S. Prefrontal involvement in the regulation of emotion: convergence of rat and human studies. *Curr. Opin. Neurobiol.* **16**, 723–727 (2006).
26. Fang, F., Potter, T., Nguyen, T. & Zhang, Y. Dynamic Reorganization of the Cortical Functional Brain Network in Affective Processing and Cognitive Reappraisal. *Int. J. Neural Syst.* **30**, 2050051 (2020).
27. Bergmann, T. O. & Hartwigsen, G. Inferring causality from noninvasive brain stimulation in cognitive neuroscience. *J. Cogn. Neurosci.* **33**, 195–225 (2021).

28. Marques, L. M., Morello, L. Y. N. & Boggio, P. S. Ventrolateral but not Dorsolateral Prefrontal Cortex tDCS effectively impact emotion reappraisal – effects on Emotional Experience and Interbeat Interval. *Sci. Rep.* **8**, 15295 (2018).
29. Zhao, J. *et al.* The VLPFC versus the DLPFC in Downregulating Social Pain Using Reappraisal and Distraction Strategies. *J. Neurosci.* **41**, 1331–1339 (2021).
30. Feeser, M., Prehn, K., Kazzer, P., Mungee, A. & Bajbouj, M. Transcranial Direct Current Stimulation Enhances Cognitive Control During Emotion Regulation. *Brain Stimulat.* **7**, 105–112 (2014).
31. Molavi, P. *et al.* Repeated transcranial direct current stimulation of dorsolateral-prefrontal cortex improves executive functions, cognitive reappraisal emotion regulation, and control over emotional processing in borderline personality disorder: A randomized, sham-controlled, parallel-group study. *J. Affect. Disord.* **274**, 93–102 (2020).
32. De Raedt, R. *et al.* Neurocognitive effects of HF-rTMS over the dorsolateral prefrontal cortex on the attentional processing of emotional information in healthy women: An event-related fMRI study. *Biol. Psychol.* **85**, 487–495 (2010).
33. Zwanzger, P. *et al.* Inhibitory repetitive transcranial magnetic stimulation (rTMS) of the dorsolateral prefrontal cortex modulates early affective processing. *NeuroImage* **101**, 193–203 (2014).
34. Ngetich, R., Zhou, J., Zhang, J., Jin, Z. & Li, L. Assessing the Effects of Continuous Theta Burst Stimulation Over the Dorsolateral Prefrontal Cortex on Human Cognition: A Systematic Review. *Front. Integr. Neurosci.* **14**, 35 (2020).
35. Schupp, H. T. *et al.* Affective picture processing: The late positive potential is modulated by motivational relevance. *Psychophysiology* **37**, 257–261 (2000).
36. Hajcak, G. & Foti, D. Significance?... Significance! Empirical, methodological, and theoretical connections between the late positive potential and P300 as neural responses to stimulus significance: An integrative review. *Psychophysiology* **57**, (2020).
37. Bernat, E. M., Cadwallader, M., Seo, D., Vizueta, N. & Patrick, C. J. Effects of Instructed Emotion Regulation on Valence, Arousal, and Attentional Measures of Affective Processing. *Dev. Neuropsychol.* **36**, 493–518 (2011).
38. Moser, J. S., Hartwig, R., Moran, T. P., Jendrusina, A. A. & Kross, E. Neural markers of positive reappraisal and their associations with trait reappraisal and worry. *J. Abnorm. Psychol.* **123**, 91 (2014).

39. Shafir, R., Schwartz, N., Blechert, J. & Sheppes, G. Emotional intensity influences pre-implementation and implementation of distraction and reappraisal. *Soc. Cogn. Affect. Neurosci.* **10**, 1329–1337 (2015).

40. Hertrich, I., Dietrich, S., Blum, C. & Ackermann, H. The Role of the Dorsolateral Prefrontal Cortex for Speech and Language Processing. *Front. Hum. Neurosci.* **15**, (2021).

41. Leyman, L., Raedt, R. D., Vanderhasselt, M.-A. & Baeken, C. Influence of high-frequency repetitive transcranial magnetic stimulation over the dorsolateral prefrontal cortex on the inhibition of emotional information in healthy volunteers. *Psychol. Med.* **39**, 1019–1028 (2009).

42. Strauss, G. P., Ossenfort, K. L. & Whearty, K. M. Reappraisal and Distraction Emotion Regulation Strategies Are Associated with Distinct Patterns of Visual Attention and Differing Levels of Cognitive Demand. *PLOS ONE* **11**, e0162290 (2016).

43. Vierheilig, N., Mühlberger, A., Polak, T. & Herrmann, M. J. Transcranial direct current stimulation of the prefrontal cortex increases attention to visual target stimuli. *J. Neural Transm.* **123**, 1195–1203 (2016).

44. Kalisch, R. The functional neuroanatomy of reappraisal: time matters. *Neurosci. Biobehav. Rev.* **33**, 1215–1226 (2009).

45. Pe, M. L., Raes, F. & Kuppens, P. The Cognitive Building Blocks of Emotion Regulation: Ability to Update Working Memory Moderates the Efficacy of Rumination and Reappraisal on Emotion. *PLOS ONE* **8**, e69071 (2013).

46. Zaehringer, J., Falquez, R., Schubert, A.-L., Nees, F. & Barnow, S. Neural correlates of reappraisal considering working memory capacity and cognitive flexibility. *Brain Imaging Behav.* **12**, 1529–1543 (2018).

47. Barbey, A. K., Koenigs, M. & Grafman, J. Dorsolateral prefrontal contributions to human working memory. *Cortex* **49**, 1195–1205 (2013).

48. Wang, Y. & Li, X. Temporal course of implicit emotion regulation during a Priming-Identify task: an ERP study. *Sci. Rep.* **7**, (2017).

49. Yuan, J., Ding, N., Liu, Y. & Yang, J. Unconscious emotion regulation: Nonconscious reappraisal decreases emotion-related physiological reactivity during frustration. *Cogn. Emot.* **29**, 1042–1053 (2015).

50. Jung, J., Bungert, A., Bowtell, R. & Jackson, S. R. Vertex stimulation as a control site for transcranial magnetic stimulation: a concurrent TMS/fMRI study. *Brain Stimulat.* **9**, 58–64 (2016).

51. Pascual-Leone, A., Walsh, V. & Rothwell, J. Transcranial magnetic stimulation in cognitive neuroscience – virtual lesion, chronometry, and functional connectivity. *Curr. Opin. Neurobiol.* **10**, 232–237 (2000).
52. Marchewka, A., Żurawski, \Lukasz, Jednoróg, K. & Grabowska, A. The Nencki Affective Picture System (NAPS): introduction to a novel, standardized, wide-range, high-quality, realistic picture database. *Behav. Res. Methods* **46**, 596–610 (2014).
53. Lang, P. J., Bradley, M. M. & Cuthbert, B. N. *International affective picture system (IAPS): Instruction manual and affective ratings*. (University of Florida, The Center for Research in Psychophysiology, 1999).
54. Ochsner, K. N. *et al.* For better or for worse: neural systems supporting the cognitive down- and up-regulation of negative emotion. *NeuroImage* **23**, 483–499 (2004).
55. Sheppes, G. *et al.* Emotion regulation choice: A conceptual framework and supporting evidence. *J. Exp. Psychol. Gen.* **143**, 163–181 (2014).
56. Willroth, E. C. & Hilimire, M. R. Differential effects of self- and situation-focused reappraisal. *Emotion* **16**, 468–474 (2016).
57. Huang, Y.-Z., Edwards, M. J., Rounis, E., Bhatia, K. P. & Rothwell, J. C. Theta burst stimulation of the human motor cortex. *Neuron* **45**, 201–206 (2005).
58. Gómez-Herrero, G. Automatic Artifact Removal (AAR) toolbox v1. 3 (Release 09.12. 2007) for MATLAB. *Tamp. Univ. Technol.* (2007).
59. Luck, S. J. & Gaspelin, N. How to get statistically significant effects in any ERP experiment (and why you shouldn't): How to get significant effects. *Psychophysiology* **54**, 146–157 (2017).
60. Keselman, H. J., Algina, J., Kowalchuk, R. K. & Wolfinger, R. D. The analysis of repeated measurements: a comparison of mixed-model satterthwaite f tests and a nonpooled adjusted degrees of freedom multivariate test. *Commun. Stat. - Theory Methods* **28**, 2967–2999 (1999).

Acknowledgments

The study was supported by the National Science Center, Poland (grant no. 2016/21/B/HS6/03723).

Author contributions

MW – experimental design, data analysis, results interpretation and discussion, manuscript writing; AKA – data analysis, discussion, manuscript writing; JH – preparing the TMS procedure, manuscript editions; GB – statistical analysis, manuscript editions; PA - performing experiment, manuscript editions; AL – preparing data analysis pipeline; TSL – results interpretation and discussion, manuscript editions.

Data availability statement

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Competing Interests Statement

The authors declare no competing interests.

Figure Legends

- Figure 1. Results of valence ratings. *Note.* Valence Rating (0 = neutral, 10 = unpleasant). NEG-REAP = negative-reappraisal; NEG-WATCH = negative-watch; NEU-WATCH = neutral-watch.
- Figure 2. ERP plots for the NEG-WATCH condition depending on the session.
- Figure 3. The experimental procedure. Note. A. Overview of a single experimental session. B. Sample trial sequence.

Tables

Table 1. Statistics for the LPP component

time window	electrode	t	df	p	p_{FDR}
NEG-WATCH vs NEG-REAP (V session)					
LPP2 (800-1200 ms)	CPz	2.35	24	0.03	0.04
	POz	2.82	24	0.01	0.03
LPP3 (1200 -1500 ms)	POz	2.56	24	0.02	0.05
LPP4 (1500-2500 ms)	POz	2.03	24	0.05	0.15 (ns)

NEG-WATCH - NEG-REAP (V vs R session)					
LPP1 (500-800 ms)	FCz	1.99	49.4	0.05	0.10 (ns)
	CPz	2.32	49.0	0.02	0.07 (ns)
LPP2 (800-1200 ms)	FCz	2.26	49.5	0.03	0.03
	CPz	2.56	50.2	0.01	0.02
LPP3 (1200 -1500 ms)	FCz	2.43	48.1	0.02	0.04
	CPz	2.23	50.0	0.03	0.04
NEG-WATCH - NEG-REAP (V vs L session)					
LPP2 (800-1200 ms)	CPz	2.03	49.5	0.05	0.06 (ns)
	POz	2.20	49.1	0.03	0.06 (ns)
LPP3 (1200 -1500 ms)	POz	1.95	48.3	0.05	0.15 (ns)