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Research article

Anodal tDCS effects over the left dorsolateral prefrontal cortex (L-DLPFC) on the rating of facial expression: evidence for a gender-specific effect

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ABSTRACT

The accurate recognition of others' facial expressions is a core skill for social interactions. The left Dorsolateral Prefrontal Cortex (L-DLPFC) represents a key node in the network for facial emotion recognition. However, its specific role is still under debate. As such, the aim of the current neuromodulation study was to assess the causal role of the L-DLPFC in humans' rating of facial expressions of emotions and implicit attitudes toward other races. In this sham-controlled single-blind between-subject experiment, we offline administered L-DLPFC transcranial direct current stimulation (tDCS) to 69 healthy participants who were divided into three groups of 23 (each receiving anodal 1 mA tDCS, anodal 2 mA tDCS, or Sham), before completing an "Emotion Rating task and two Implicit Association Tests (IATs). The former required the intensity rating of 192 faces (half black and half white) displaying happiness, sadness, anger, or fear. The IATs were designed to assess participants' automatic associations of positive or negative attributes with racial contents. Results on the Emotion Rating task showed participants' gender-specific effect of tDCS. Specifically, a *gender bias,* with only males showing a tendency to underestimate negative emotions was found in Sham, and absent in the tDCS groups. When considering the race of the stimuli, females but not males in Sham exhibited a *racial bias*, that is, the tendency to overestimate negative emotions of other-race faces. Again, the bias disappeared in the tDCS groups. Concerning the IATs, no significant effects emerged. We conclude that the L-DLPFC plays a critical role in humans' rating of facial expressions, and for variability in other-race emotional judgements. These results shed light on the neural bases of the human emotional system and its gender-related differences, and have potential implications for interventional settings.

1. Introduction

Emotions are complex psychological states involving distinct components, such as a subjective experience, a physiological reaction, and a behavioral (or expressive) response (Hockenbury and Hockenbury, 2010). Within the range of 'channels' that people use to express themselves, facial expressions represent an innate aspect of emotions, and provide others with instantaneous information about a person's reaction and state (Ekman et al., 1987). Thus, human's ability to recognize emotions from others' faces is a critical skill for typical social cognition.

Neuroscientific evidence from patients with brain lesions and from functional magnetic resonance imaging (fMRI) has highlighted a network of cortical and subcortical areas involved in various aspects of facial expression processing. Different high-order cortical regions, such as the orbitofrontal cortex (OFC), dorsolateral prefrontal cortex (DLPFC), dorsal medial prefrontal cortex (DMPFC), ventrolateral prefrontal cortex (VLPFC), and anterior cingulate cortex (ACC) are part of the large brain network involved in the processing of emotional faces (Beauregard et al., 2001; Lévesque et al., 2003; Ochsner et al., 2002, 2004; Phan et al., 2005; Urry et al., 2006). Among these prefrontal regions, the left dorsolateral prefrontal cortex (L-DLPFC) plays a key role in mood (Davidson and Irwin, 1999), attentional processing of emotional information (Jacob et al., 2014), and processing of both emotional scenes (Ueda et al., 2003) and facial expressions (Sergerie et al., 2005; Nitsche et al., 2012). In addition, it represents an important corticolimbic hub to down- and up-regulate limbic responses, and plays a pivotal role in the interplay

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between emotional states, attention, and evaluative processes (Allaert et al., 2020; Banks et al., 2007; Mondino et al., 2015). The DLPFC, together with ACC, is assumed to be associated also with social decision-making processes (Balconi and Canavesio, 2014; Chee et al., 2000). Specifically, the ACC seems to detect conflicts between intentions and automatic social evaluations, with the DLPFC being engaged in a regulatory mechanism to control implicit attitudes (Richeson et al., 2003; Stanley et al., 2008).

Human's ability to recognize emotions from others' faces has been tested with various experimental manipulations, such as "*expression-labelling tasks*" (i.e., choosing the name of the emotion that matches a presented face) (Nowicki and Carton, 1993; Sedda et al., 2013; Wilhelm et al., 2014), and "*intensity-judgment*" tasks (i.e., rating the intensity of a given emotion) (Peña-Gómez et al., 2011; Wang et al., 2014). These have highlighted specific and robust behavioural effects, such as the *gender bias* (Wang, 2013) and the *race bias* (Wang et al., 2014). The *gender bias* shows that women are more accurate than men in judging emotional meaning from nonverbal information, even under situations of minimal stimulus details (Hall and Matsumoto, 2004; Montagne et al., 2005; Hoffmann et al., 2010), Furthermore, neuroimaging studies show that male and female subjects use a rather different set of neural correlates when processing emotional faces, particularly for positive vs. negative facial expressions and, with respect to event-related potential, exhibit differences in neural components for attentional orientation toward emotional faces (Lee et al., 2002; Campanella et al., 2004). Gender-related differences also emerge in event-related potentials (ERPs) when considering the "early" vs. "late" processing stage of facial stimuli (Knyazev et al., 2010). These results suggest that the generalizability of findings on facial emotion recognition should consider the gender of participants (Lee et al., 2002).

The *race bias* indicates that people are more accurate in recognising facial expressions belonging to the perceiver's race (Johnson and Fredrickson, 2005; Elfenbein and Ambady, 2002). In addition, recognition biases are also supported by studies on ERP's elicited by own-race vs. other-race faces (Ito and Urland, 2005). This bias is strictly linked to stereotypes and implicit attitudes, since implicit prejudice toward out-groups exerts a systematic influence on emotion judgements and results in higher ratings of negative expressions of faces from a different racial group (Hugenberg and Bodenhausen, 2003; Hutchings and Haddock, 2008). People's implicit attitudes can be effectively measured with the Implicit Association Test (IAT), designed to assess positive and negative automatic evaluations underlying implicit attitudes through a series of target-concept discrimination tasks (Greenwald et al., 1998).

Despite a right hemispherical lateralization (referred as the '*Right-Hemisphere Hypothesis*) has been proved for different aspects of emotional face processing (Dimberg and Petterson, 2000; Shobe, 2014; Levy et al., 1983) and related *gender* and *race biases* (Prete et al., 2016, 2017; Prete and Tommasi, 2019), much evidence demonstrates the left hemisphere plays a pivotal role in facial expressions recognition (Stone et al., 1996; Ahern and Schwartz, 1979; Harmon-Jones, 2004; Najt et al., 2013), even in relation to gender-specific differences (Proverbio et al., 2006; Ran et al., 2014) and racial factors (Wiese et al., 2014). However, given the heterogeneity of findings concerning the left hemisphere's role, the debate over hemispheric asymmetries for emotion perception is still ongoing (Alves et al., 2009; Brüne et al., 2013; Fetterman et al., 2013). A meta-analysis (Abbott et al., 2013) concluded that both hemispheres process emotions through a cross-hemispheric "collaboration", with the left one being specialized in the positive emotions. A broader range of evidence suggests that the left hemisphere may play a more prominent role in emotional processing at levels beyond simple identification of emotionality that has yet to be extensively explored. Specifically, it has been proposed that the left hemisphere may be involved in additional or secondary interpretation, and to make an important contribution to the regulation of negative emotion and social interaction (Shobe, 2014). Another interpretation in this debate is the hypothesis of differential hemispheric specialization as a function of affective valence (known as '*Valence-Specific Hypothesis*'), which postulates that left and right hemispheres are dominant for positive and negative emotions, respectively (Davidson et al., 1987; Adolphs et al., 2001).

A technique that has recently been used to causally determine the role of a brain area in visual cognition is transcranial direct current stimulation (tDCS) (Barbieri et al., 2016; Nitsche et al., 2012). In its simplest form, tDCS consists in delivering a small current (1–2 mA) through the scalp that results in a neuronal excitability shift via de- or hyper-polarization of neuronal membranes, with anodal and cathodal stimulation respectively increasing and decreasing cortical excitability at the macroscopic level (Nitsche and Paulus, 2000; Nitsche et al., 2003). Due to its key role in high-order processes related to emotional content, the DLPFC has often been targeted in tDCS protocols (Vierheilig et al., 2016; Penton et al., 2017; Prete et al., 2017). tDCS effects on cortical activity and connectivity of the DLPFC have been investigated in relation to the impact of mood, emotional processing, and attention on emotional information (Tremblay et al., 2014). Even though variability of tasks and stimulation parameters resulted in heterogeneous outcomes, the use of transcranial electrical stimulation (tES) over the DLPFC has led to consistent evidence for its role in emotional content processing. In line with the *Valence-Specific Hypothesis*, anodal stimulation over the L-DLPFC has been shown to decrease negative emotions perception (Maeoka et al., 2012), to enhance cognitive control for positive affective stimuli (Vanderhasselt et al., 2013), and to improve positive emotional face identification (Nitsche et al., 2012). Concerning emotional intensity, Peña-Gómez et al. (2011) found that L-DLPFC stimulation led to an underestimation of affective pictures with a negative valence, explicable in terms of enhancement of cognitive control of emotional expression, with a modulatory effect of individual personality differences. These findings corroborate the role of the L-DLPFC as a core hub in the emotional regulatory circuit.

Albeit tDCS over the L-DLPFC has been shown to enhance facial expression recognition in labelling tasks (Nitsche et al., 2012), it still remains important to corroborate this evidence with different, and potentially more compelling, paradigms such as emotional intensity ratings; in addition, it is largely unknown whether L-DLPFC plays a role in known behavioural effects, such as *gender bias* and the *race bias*. As such, in the current single-blind sham-controlled between-subjects design study, we tested whether the L-DLPFC is causally involved in intensity judgements of facial expressions, and in the behavioural findings of *gender bias* and *race bias*. Furthermore, we were interested in assessing L-DLPFC involvement in implicit negative attitudes toward other-race faces. For this purpose, we administered anodal tDCS (a-tDCS) before participants completed an emotion-rating task and two racial IATs. We adopted three stimulation intensities (i.e., Sham, 1 mA, and 2 mA) to test the effects of current injection on behavioural outcomes. In line with previous studies (Nitsche et al., 2012; Wang et al., 2014), we expected that excitability-enhancing stimulation of the L-DLPFC can (i) modulate participants' rating of emotions, with underestimation of negative expressions especially for other-race faces, and higher performance of female participants, and (ii) exert an effect on implicit attitudes toward other-race faces. However, given the limited research available on the topic, we were unable to make specific predictions on the interaction effects.

2. Materials and methods

2.1. Participants

Sixty-nine Caucasian right-handed volunteers (36 females, mean age 23.24 ± 3.76 SD) with normal or corrected-to-normal vision and without any recorded history of psychiatric or neurological disorder were recruited for the experiment. Participants were assigned to one of three groups (N = 23 in each group) with the same M/F ratio (i.e., 12 F per group) receiving different stimulation protocols (see section below). All participants were naïve to the research hypotheses and the experimental condition they were assigned to. Prior to the testing session, they received a verbal and written explanation of the procedure and the typical adverse effects of brain stimulation (i.e., itching and tingling skin sensation, skin reddening, and headache). Participants gave their written informed consent to participate in the study. This research was conducted according to the ethical standards of the World Medical Association Declaration of Helsinki, and the study protocol received approval from the Ethics Committee of the University of Bari 'Aldo Moro' (protocol number: ET-19-01).

2.2. Experimental design

In this sham-controlled between-subjects design, participants were assigned to one of three groups: Group-1 ("Sham"; N = 23), Group-2 ("tDCS_1mA"; N = 23), and Group-3 ("tDCS_2mA"; N = 23) (see next section for a detailed description of the stimulation protocol). Given that females show similar cortical excitability compared to males only during the follicular phase of the menstrual cycle (when progesterone levels are low and estrogen levels are high), they were tested only during this phase (Inghilleri et al., 2004). Participants from all three groups completed the Emotions Rating Task and two racial IATs (facial IAT and control IAT) immediately after tDCS (i.e., *offline*). The tasks were set up with SuperLab 5.0 (Version 5.0.5, Cedrus Corporation, USA) and administered on a Fujitsu computer running Windows 10 and with a 1920 × 1080 pixels 23-inches monitor. Counterbalancing across tasks (Emotion Rating task vs IATs, and facial IAT vs control IAT) was adopted to control for order effects.

The Emotion Rating task was designed with pictures of facial expressions selected from the Binghamton University 3D Facial Expression Database (BU-3DFE) (Yin et al., 2006). A total of 192 photos (size: 600 × 650 pixels) of 4 actors were chosen (i.e., 1 Caucasian female, 1 Afro-American female, 1 Caucasian male, and 1 Afro-American male). From each of these identities, pictures showing happy, sad, fearful, and angry faces were selected, including different levels of intensity of each emotion (25%, 50%, 75%, 100%). The emotion selection was based on the *Approach/Withdrawal Hypothesis* (anger and happiness as 'approach' emotions vs. fear and sadness as 'withdrawal' emotions), to take into account both emotional valence and social connotation (Kemp and Guastella, 2011). Each stimulus had three versions depicting different view angles: (i) full-face, (ii) 45° to the right, and (iii) 45° to the left (Figure 1), in order to adequately mimic humans' naturalistic face evaluation (Palermo et al., 2013). Four additional stimuli were selected from the same database to create a brief practice session to be completed before the actual task.

In each trial, a face was presented at the centre of a black screen, preceded by a 2.5 s prompt indicating which emotion the following facial expression would show. Participants were instructed to rate the emotion intensity of each face on a Likert-like scale from 1 (very low intensity) to 4 (very high intensity) by pressing the corresponding button on a computer keyboard. Pictures disappeared after response within a time limit of 8 s; then, a black screen was presented for 1 s before the beginning of the next trial (Figure 2). The presentation order of the stimuli was randomized, and the task took approximately 35–40 min to complete.

Two *good-bad* evaluative IATs were created: the 'facial' version included attributes and faces, whilst the control version had attributes and proper names. To design the face-IAT, 10 pictures of white and 10 of black neutral faces were casually chosen from The Chicago Face Database (Ma and Wittenbrink, 2015). The attributes consisted of 10 positive (e.g. friendly, intelligence) and 10 negative words (e.g. enemy, terror). To design the control IAT, 10 typical Italian (participants' in-group) and 10 typical African (participant's out-group) proper names were selected.

Both IATs contained two critical double-categorization blocks (Figure 3). In one double-categorization block ("congruent" condition), participants were asked to press one button ("D" or "K") for either positive words and white faces (in-group names in the control version), whereas the other button was pressed for either negative words and black faces (out-group-names in the control version). Faces and words appeared at the centre of the screen until participants gave their response. In another block ("incongruent" condition), participants pressed one button for either "positive words and out-group faces/names", and pressed another button for either "negative words and in-group faces/names". In case of a wrong classification, an "X" appeared on the screen until participants corrected their answer. If participants held more positive attitudes toward their in-group than out-group members, such attitudes would be manifested via a better performance during the positive+in-group and negative + out-group block than during the opposite one. Half of the participants received the positive+in-group block first, whereas the other half received the positive + out-group block first. The task took approximately 10 min to complete.

At the end of the experimental session, all participants were debriefed and asked to fill in a 'tDCS adverse effects questionnaire' about potential uncomfortable sensations experienced during or after the stimulation protocol (Sellaro et al., 2015).

2.3. tDCS stimulation

TDCS was delivered by a battery driven, constant current stimulator (BrainSTIM stimulator; E.M.S. s.r.l.) via a pair of surface sponge electrodes (25 cm^2) soaked in saline solution (0.9% NaCl) and applied to the scalp at the target location. According to the experimental condition, electrodes delivered a constant current of 1 mA (tDCS_1mA) or 2 mA (tDCS_2mA) (current density: 0.04 and 0.08 mA/cm^2, respectively). Participants received *offline* stimulation; that is, tDCS (or Sham) was

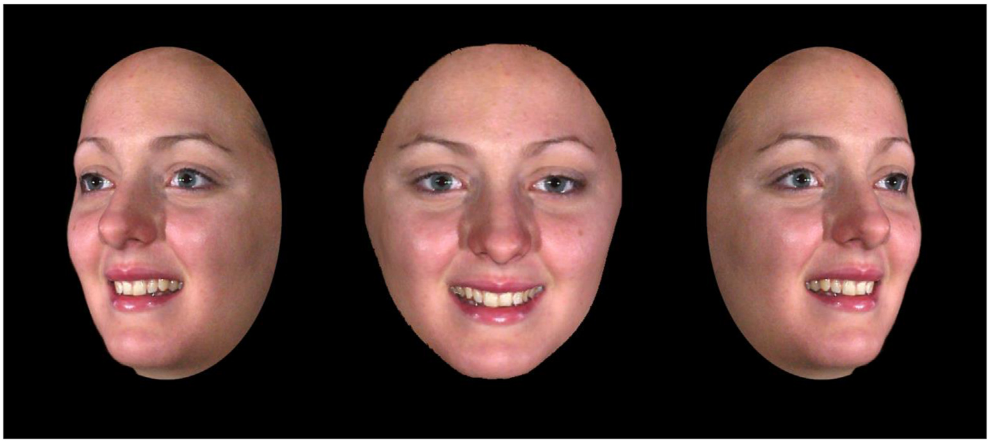

Figure 1. Example of a female, happy face model (emotion intensity 50%) from the BU-3DFE in different angles.

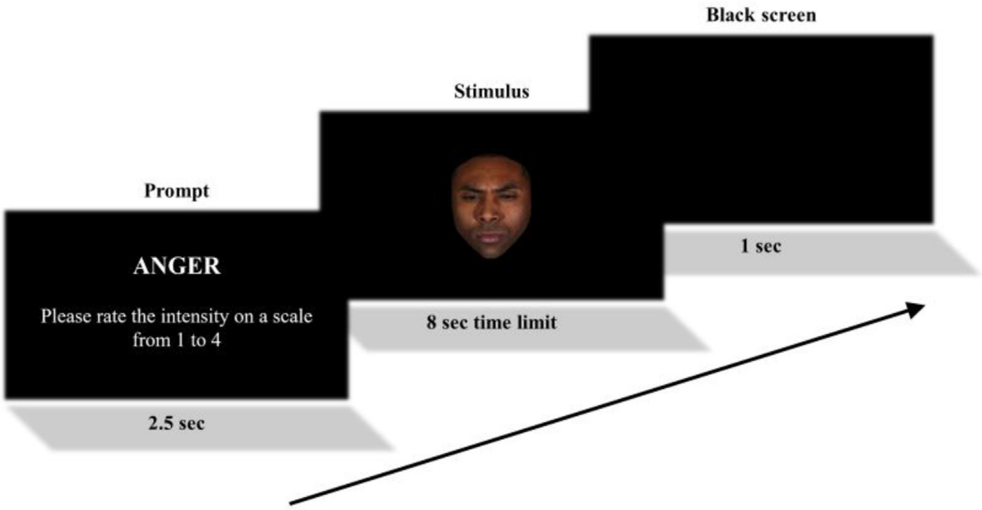

Figure 2. Schema of the trial structure in the Emotion Rating task. Faces were presented at the centre of a black screen, preceded by information indicating which emotion the following facial expression would show. Participants were asked to rate the emotion intensity of each face from 1 to 4 by pressing the corresponding button. A rating index was calculated.

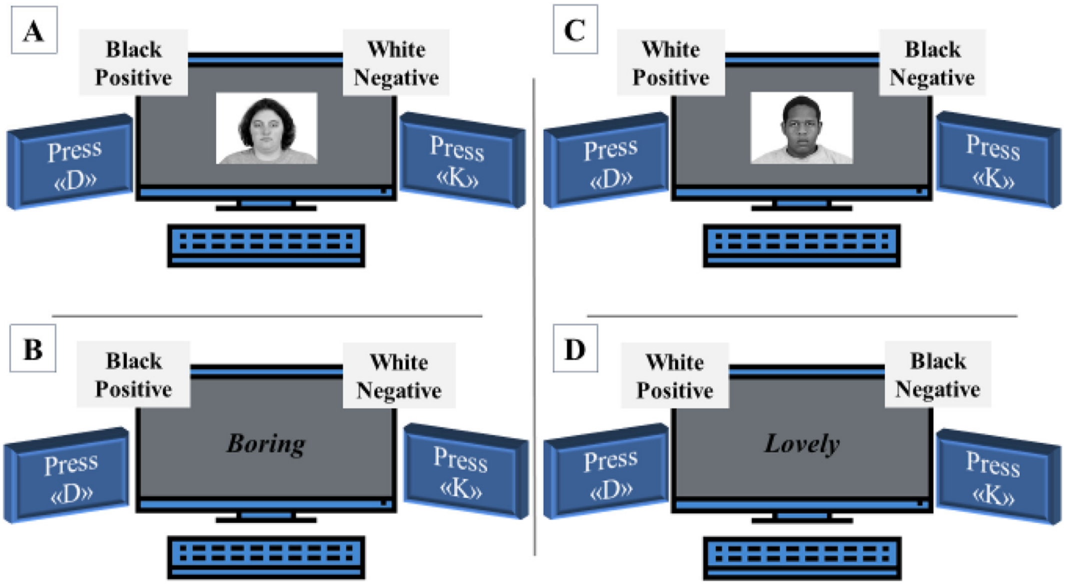

Figure 3. Schematic representation of the racial IAT. Both pictures of black or white individuals (A, C) and words representing good or bad evaluative attributes (B, D) were randomly presented in the centre of the screen. Participants were asked to classify these as to the category pairs shown in the upper left or the upper right of the screen using the "D" or "K" key on their computer keyboard. When participants pressed the wrong button, an "X" appeared on the screen until they corrected their answer. Performance in the positive + outgroup (A)/negative + ingroup (B) associations (incongruent) and positive + ingroup (C)/negative + outgroup (B) (congruent) are compared by the Greenwald's algorithm as a measure of implicit race preference (Greenwald et al., 2003).

applied for 20 min *before* any task execution, with 10 s ramp-up and 10 s ramp-down. We chose offline stimulation to be consistent with our previous tDCS work (Costantino et al., 2017; Willis et al., 2019), and because there is direct evidence of more effective offline tDCS in visual cognition (Barbieri et al., 2016).

We adopted a "unilateral-monopolar-montage" (Nasseri et al., 2015), with the anode located over F3 (the electrode traditionally adopted to target the L-DLPFC) (Edgcumbe, 2018; Herwig et al., 2003), and the reference (i.e., cathode) over the right deltoid muscle. The site of active stimulation was identified using the Electroencephalography 10–20 system. To successfully blind participants, in the Sham condition the stimulator was turned on for only 20 s to elicit a short-lasting skin sensation. None of the participants reported major complains or intolerable discomfort during or after tDCS.

2.4. Statistical analyses

All analyses were conducted using RStudio (R Team, 2015). A stepwise multiple regression mixed model was adopted to test for tDCS effects on participant's emotion ratings and IAT scores. With respect to the Emotion Rating task, a *rating index* was calculated for each trial as the difference between participants' responses and the correct emotion intensity. This value (range interval: ± 3) was submitted to the analyses as a dependent variable. Experimental group ("Group": Sham, tDCS_1mA, tDCS_2mA) and participants' gender ("Gender": M, F) were between-subject independent variables. Three within-subject independent factors related to the face stimuli were also included: facial expressions of emotions ("Emotion": happiness, anger, sadness, fear), emotion intensity ("Intensity": 25%, 50%, 75%, 100%) and race of the shown faces (“Race”: black, white). Both the view angle of the stimuli and stimuli’ gender have not been included in the analysis. Post-hoc comparisons (using Tukey HSD p-value correction) were performed in order to probe statistically significant interactions.

The scoring of the IATs consisted of an estimate of the implicit association effect based on the algorithm developed by Greenwald and colleagues, named *D-score* (Greenwald et al., 2003). A lower D-score indicated a larger “anti-black” bias revealed by faster and more accurate performances in the positive+in-group and negative + out-group blocks than in the reverse ones.

3. Results

3.1. Effects that did not include participants’ gender

All main effects and interactions of the mixed model for the Emotion Rating Task are reported in Table 1. Post-hoc tests were performed for all effects in highest-level interaction, as main and high-level effects are further specified by higher-level interactions (e.g., post-hoc tests for the emotion main effect and the emotion*race interaction are not reported because the effect is further specified by the emotion*race*intensity interaction effect). For concision and clarity purposes, only the statistically significant post-hoc tests will be reported.

Results not including participants’ gender showed significant *main effects* of emotion [F (3, 13121) = 50.7, $p < .001$], race [F (1, 13121) = 206.6, $p < .001$] and intensity [F (1, 13121) = 9274.9, $p < .001$]. Five *two-way interactions* were statistically significant: (i) group*intensity [F (2, 13121) = 9.4, $p = .002$], (ii) group*emotion [F (6, 13121) = 10.1, $p < .001$], (iii) emotion*race [F (3, 13121) = 45, $p < .001$], (iv) emotion*intensity [F (3, 13121) = 107.1, $p < .001$] and (v) race*intensity [F (1, 13121) = 7.8, $p = .005$]. The *three-way interaction* emotion*race*intensity was statistically significant [F (3, 13121) = 18.5, $p < .001$].

Post-hoc tests conducted for the (ii) group*intensity interaction showed that underestimation and overestimation errors in all groups significantly increased and decreased in relation to emotion intensity in all groups (Sham: b = -0.8926; tDCS_1mA: b = -0.8939; tDCS_2mA: b = -0.9515). Contrasts performed for the (ii) group*emotion interaction showed a general trend in each group, with happy faces being rated as more intense than fearful, fearful more intense than angry, and angry more intense than sad (all $zs > 3$, all $ps < .001$).

Concerning the IATs, both effects in the mixed model and correlations between IATs’ D-scores and Emotion Rating indexes were explored. Neither effects nor interactions were statistically significant (all $ps > .05$).

3.2. Evidence for a gender-specific effect

The first-level interaction group*gender was statistically significant [F (2, 69) = 3.2, $p = .048$]. *Post-hoc* comparisons based on gender revealed that men and women significantly differed in the Sham group ($z = 2.043$, $p = .041$) with female participants obtaining a higher *rating index* than males (M = 0.024; SD = 1.339; F = 0.282: SD = 1.267); this gender difference was attenuated and non-significant in the tDCS_1mA (M = 0.098; SD = 1.348; F = 0.226: SD = 1.341; $z = 1.011$, $p = .312$) and further numerically mitigated in the tDCS_2mA ($z = 0.361$, $p = .718$) conditions. The two tDCS intensities did not result in statistically significant effects ($p = .254$). Three *three-way interactions* including the gender variable were statistically significant: (i) group*gender*emotion [F (6, 13121) = 7.1, $p < .001$], (ii) group*gender*race [F (2, 13121) = 4.5, $p = .011$], (iii) gender*emotion*race [F (3, 13121) = 3.5, $p = .016$]. *Post-hoc* contrasts based on gender performed for the (i) group*gender*emotion interaction show that males, as compared to females, attributed lower ratings to the emotions of anger (M = -0.208; SD = 1.319; F = 0.155; SD = 1.251; $z = 2.724$, $p = .006$), fear (M = 0.211; SD = 1.398; F = 0.498; SD = 1.291; $z = 2.178$, $p = .029$) and sadness (M = -0.379; SD = 1.362; F = -0.026; SD = 1.349; $z = 2.646$, $p = .008$) in the Sham condition only. With the exception of fear in tDCS_1mA ($z = 2.078$, $p = .04$), this difference between genders was not statistically significant in the tDCS_1mA group (all $|z|s < 1.5$, all $ps > .1$), and in tDCS_2mA group (all $|z|s < .9$, all $ps > .3$) (Figure 4). *Post-hoc* contrasts based on emotion revealed significant differences among emotions, with angry and particularly happy faces receiving

Table 1. ANOVA – Fixed factors.

	Sum Sq	Mean Sq	Num DF	Den DF	F-value	Pr (>F)
Group	1.4	0.7	2	86.8	10.968	0.338521
Gender	1.1	1.1	1	86.8	18.210	0.180701
Emotion	54.1	18.0	3	13121.0	292.552	<2.2e-16***
Race	0.3	0.3	1	13121.0	0.4119	0.521024
Intensity	11436.6	11436.6	1	13121.0	185.496.692	<2.2e-16***
Group*Gender	0.9	0.4	2	63.0	0.7198	0.490833
Group*Emotion	37.9	6.3	6	13121.0	102.380	2.456e-11***
Gender*Emotion	0.1	0.0	3	13121.0	0.0563	0.982426
Group*Race	0.4	0.2	2	13121.0	0.3593	0.698186
Gender*Race	0.4	0.4	1	13121.0	0.6554	0.418196
Emotion*Race	294.4	98.1	3	13121.0	1.591.730	<2.2e-16***
Group*Intensity	5.9	2.9	2	13121.0	47.815	0.008398**
Gender*Intensity	0.9	0.9	1	13121.0	14.884	0.222490
Emotion*Intensity	198.2	66.1	3	13121.0	1.071.366	<2.2e-16***
Race*Intensity	4.8	4.8	1	13121.1	77.641	0.005337**
Group*Gender*Emotion	44.1	7.3	6	13121.0	119.188	2.182e-13***
Group*Gender*Race	5.5	2.7	2	13121.1	44.541	0.011648*
Gender*Emotion*Race	6.4	2.1	3	13121.2	34.532	0.015771*
Gender*Emotion*Intensity	1.2	0.4	3	13121.0	0.6355	0.592063
Gender*Race*Intensity	0.3	0.3	1	13121.0	0.4155	0.519182
Emotion*Race*Intensity	34.3	11.4	3	13121.1	185.452	5.318e-12***
Gender*Emotion*Race*Intensity	5.5	1.8	3	13121.2	29.965	0.029468*

Signif. codes: 0 ‘***’ 0.001 ‘**’ 0.01 ‘*’.

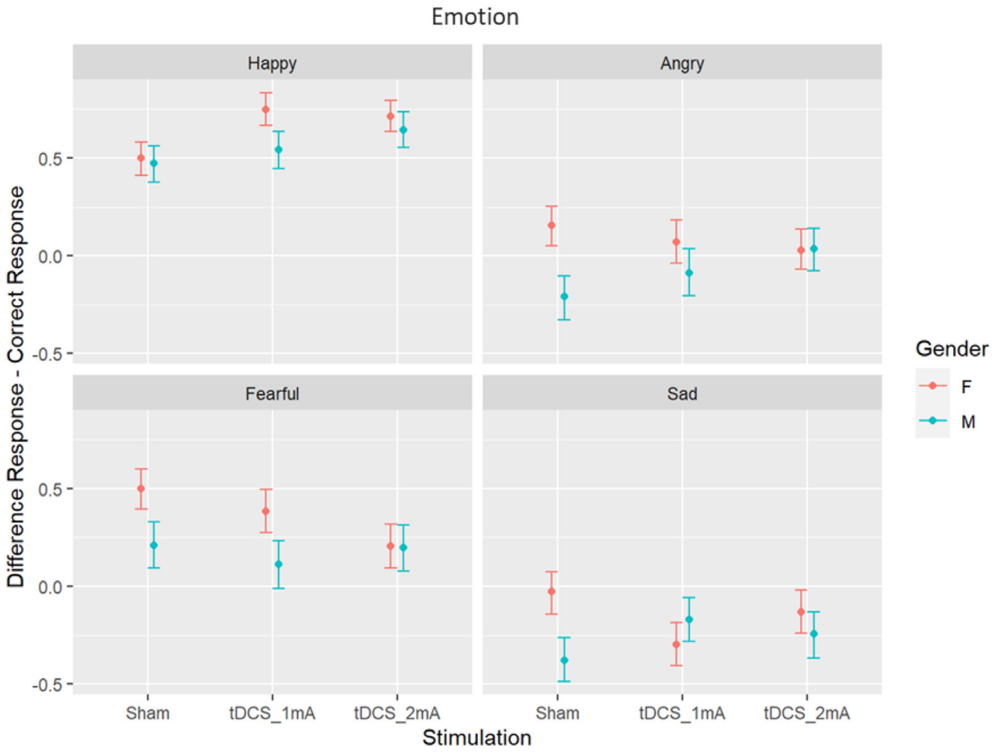

Figure 4. Emotion Rating Task: Group x gender x emotion. Contrasts based on gender. Y-axis: rating index (difference between participant's response and correct response); x-axis: stimulation groups ('Sham', 'tDCS_1mA', 'tDCS_2mA'). Subfigures: average responses for Happy (upper left), Angry (upper right), Fearful (lower left) and Sad (lower right) faces. Red bars: female responses; blue bars: male responses. The '0.0' on the y-axes indicates the higher rating correctness (null difference between participants' response and correct intensity). Male participants (blue bars), as compared to females (red bars), gave lower ratings to the emotions of anger (z = 2.724, p = .006), fear (z = 2.178, p = .029) and sadness (z = 2.646, p = .008) in the Sham condition only. With the exception of fear in tDCS_1mA (z = 2.078, p = .04), this gender difference was absent in the tDCS_1mA (all |z|s < 1.5, all ps > .1) and tDCS_2mA conditions (all |z|s < .9, all ps > .3).

higher ratings than the other emotions in all groups (3.392 < all |*z*|s < 22.670, all *ps* < .003, with the exception of (i) the happy–fearful contrast for females in Sham (*z* = 0.078 *p* = .999), and (ii) the angry–sad contrast for males in tDCS_1mA group (*z* = 1.764, *p* = .291)). *Post-hoc* contrasts based on gender for the (ii) group*gender*race interaction showed a statistically significant higher rating of female compared to male participants for black faces in the Sham group only (M = -0.037; SD = 1.352; F = 0.239; SD = 1.265*; z* = 2.152, *p* = .031), and a trend towards significance for white faces (M = 0.084; SD = 1.326; F = 0.325; SD = 1.26*; z* = 1.864, *p* = .062) (Figure 5). Contrasts based on race showed a lower rating of emotional intensity of black faces compared to the white ones for both females (Sham: *z* = 2.597, *p* = .009; tDCS_1mA: *z* = 5.715, *p* < .001; tDCS_2mA: *z* = 2.137, *p* = 0.03) and males (Sham: *z* = 3.572, *p* = .0004; tDCS_2mA: *z* = 3.464, *p* < .001), with the exception of men in the tDCS_1mA group, which revealed no significant results (*z* = 1.668, *p* = .096) (Figure 6).

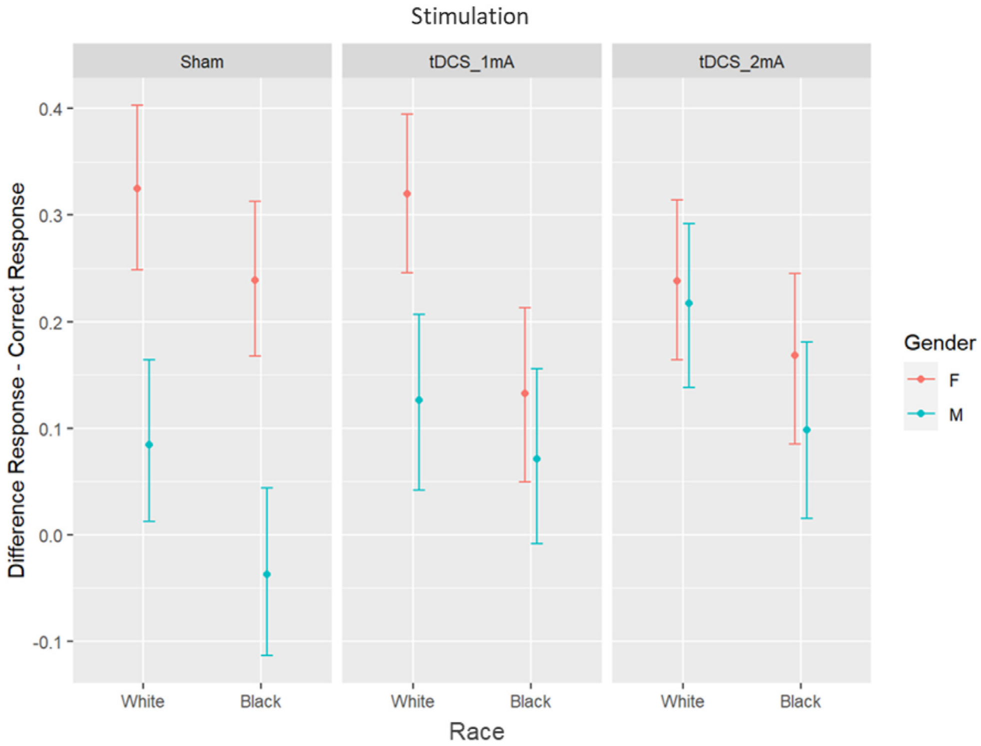

Figure 5. Emotion Rating Task: Group x gender x race. Contrasts based on gender.Y-axis: rating index (difference between participat's response and correct response); x-axis: race of the stimuli ('White', 'Black'). Subfigures: Sham stimulation (left), tDCS_1mA (center) and tDCS_2mA (right). Red bars: female responses; blue bars: male responses. The '0.0' on the y-axes indicates the higher rating correctness (null difference between participants' response and correct intensity). Female participants (red bars), as compared to males (blue bars), exhibited a higher rating for black faces in the Sham group only (z = 2.152, p = .031).

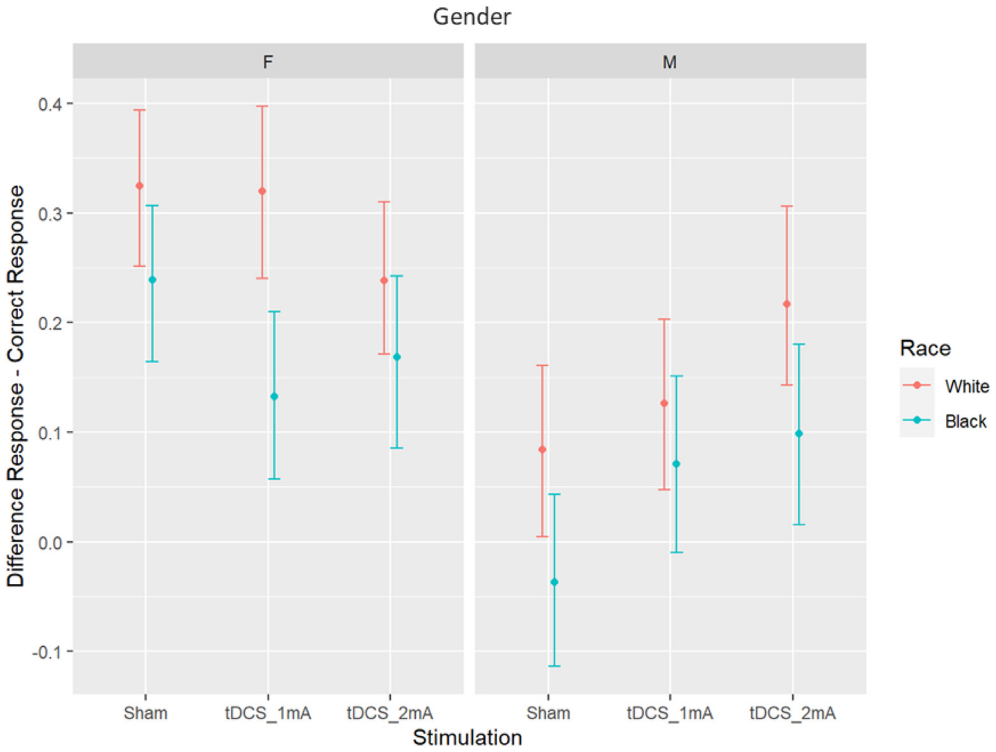

Figure 6. Emotion Rating Task: Group x gender x race. Contrasts based on race. Y-axis: rating index (difference between participat's response and correct response); x-axis: stimulation groups ('Sham', 'tDCS_1mA', 'tDCS_2mA'). Subfigures: average responses for female (left) and male (right) participants. Red bars: white faces; blue bars: black faces. The '0.0' on the y-axes indicates the higher rating correctness (null difference between participants' response and correct intensity). Both females and males showed a lower rating of emotional intensity of black faces (blue bars) compared to the white ones (red bars) (Females: Sham: z = 2.597, p = .009; tDCS_1mA: z = 5.715, p < .001; tDCS_2mA: z = 2.137, p = 0.03; Males: Sham: z = 3.572, p = .0004; tDCS_2mA: z = 3.464, p < .001), with the exception of men in the tDCS_1mA group, which revealed no significant results (z = 1.668, p = .096).

Concerning the three-way interactions that did not include stimulation, post-hoc comparisons based on race performed for the (iii) gender*emotion*race interaction revealed a higher rating of emotion intensity of happy and angry black faces compared to the white ones (all $|z|$s < -3.8, all *ps* < .001). Conversely, fearful and sad expressions of *white* faces were rated as more intense than the black ones (all $|z|$s > 11.7, all *ps* ≤ .0001). Contrasts based on gender revealed that male participants rated white angry ($z = 3.004$, $p = .002$) and fearful ($z = 2.26$, $p = .023$) faces, and black fearful ($z = 2.420$, $p = .015$) and sad faces ($z = 2.024$, $p = .043$) as less intense than female counterparts. Post-hoc contrasts based on emotion showed that both males and females gave higher ratings to fearful than happy than sad and angry white faces, while black faces were rated as more happy than angry than fearful and sad faces (4.105 < all $|z|$s < 35.184, all *ps* < .001, except the angry–sad contrast for white faces from females ($z = -1.518$ $p = .426$).

Concerning the IATs, both effects in the mixed model and correlations between IATs' D-scores and Emotion Rating indexes were explored. Neither effects nor interactions were statistically significant (all *ps* > .05). Results from the 'tDCS adverse effects questionnaire' indicated no significant differences between the stimulation conditions ($p > .05$).

4. Discussion

Lesion studies and neuroimaging experiments indicate that the L-DLPFC is a critical hub in emotional cognition (Banks et al., 2007; Conson et al., 2015). However, causal evidence for this involvement along with its features is still scarce. Thus, the aim of the current study was to investigate whether 20 min of 1 and 2 mA offline anodal-tDCS over the L-DLPFC results in a change of (i) intensity judgement of emotional faces and (ii) implicit racial preference in neurotypical subjects. Our findings show a L-DLPFC tDCS gender-specific effect. Specifically, male and female participants differed in their rating indexes of negative emotions only in the Sham group; this difference disappeared in both tDCS_1mA and tDCS_2mA. In addition, a higher rating in female compared to male participants was found for black faces in the Sham group only. This greater sensitivity of females to negative emotions in the Sham condition was eliminated by tDCS at 1 and 2 mA.

4.1. Gender differences in the sham group

Male and female participants in the Sham condition exhibited significant differences in their rating indexes. Specifically, as compared to females, males showed a tendency to underrate negative but not positive emotions (i.e., anger, fear, and sadness). The origin of gender differences in emotional processing is under conceptual debate, specifically with respect to its source, which includes biologically driven factors as well as socialisation processes (McClure, 2000). Previous studies revealed a *gender bias*, with females being better at recognizing emotional expressions, especially for expressions' subtle cues (known as the *Emotional Sensitivity Hypothesis)* (Campbell et al., 2002; Hoffmann et al., 2010; Montagne et al., 2005; Thayer and Johnsen 2000).

We have, however, found no support for a female advantage. In contrast, males were numerically, albeit not significantly, closer to the rating index' *zero*, which represents the highest accuracy in intensity judgement (i.e., perfect match between perceived intensity and real intensity). In line with our results, Hall and Matsumoto (2004) found that men and women produce a different pattern of rating emotions in facial expressions, with females giving significantly higher ratings than men on all emotions except happiness. As a matter of fact, the literature on sex differences in emotional processing reveals a female higher sensitivity to emotionally negative stimuli irrespectively of the stimuli valence intensity, which is in line with our results (Li et al., 2008; Yuan et al., 2007). This can be linked to what is often seen in real-life settings: males show fewer emotional responses to negative events that, however, generally elicit strong emotional reactivity in females (Yuan et al., 2009).

Another relevant gender difference in the Sham group emerged in relation to race, with female participants exhibiting higher ratings for black faces than male counterparts. Previous research on implicit attitudes toward outgroups suggests that biases exert an effect on facial expressions judgement: a higher *race bias* corresponds to an intensified perception of negative expressions from other-race faces (Hugenberg and Bodenhausen, 2004; Hutchings and Haddock, 2008; Wang et al., 2014). In our study, female participants gave higher ratings for blacks as compared to males; this might suggest a higher *other race* bias in women. Interestingly, this effect was mitigated by the intervention (see paragraph 4.2).

With respect to the *Approach/Withdrawal Hypothesis,* approach-oriented emotions (happiness and anger) received higher ratings and were rated as more intense in black faces compared to the white ones, while withdrawal-oriented emotions (fear and sadness) showed an opposite trend. This result is in line with neuroimaging studies showing differential processing of emotional faces as a function of race (Lee et al., 2008; Ran et al., 2014), and with the idea of 'approach' and 'withdrawal' as the best explanation for the way the brain processes emotional stimuli (Barrett and Wager, 2006).

4.2. The role of tDCS over L-DLPFC

Neuromodulation (1 mA and 2 mA) over the L-DLPFC eliminated the gender differences that were observed in the Sham condition. In other words, *tDCS made the two genders more similar in their ability to rate emotion intensities*. The significant gender difference observed in the Sham condition was not found in both tDCS_1mA and tDCS_2mA groups. This implies that anodal stimulation effectively modulated emotional intensity perception, and exerted a differential effect on male versus female participants. Precisely, tDCS determined a numerical increase of the rating indexes for males, and decreased the rating indexes for female participants. This result supports (i) the gender-related diversity in emotion processing, and (ii) the role of the L-DLPFC in facial expression judgement. The inconsistency and complexity of results from studies investigating the role of this brain area imply the need for further investigations, particularly to better understand the role of prefrontal regions in gender dimorphisms and emotional processing (Tremblay et al., 2014).

Since the activation of both the right and left DLPFC has been associated with the processing of emotions and facial expressions (Peña-Gómez et al., 2011; Mondino et al., 2015) future studies should ascertain potential site-specific effects on the contralateral cortex. The choice to target the L-DLPFC has been based on a literature review revealing intriguing effects of tDCS over this area in relation to both gender (Conson et al., 2015) and valence of the stimuli (Nitsche et al., 2012). Overall, as stated in the Introduction, the DLPFC modulates emotional processing, both for stimuli identification and retrieval, and this is particularly true for the left hemisphere (Banks et al., 2007). L-DLPFC plays a crucial role in the interplay between emotional processing, attention and decision making (Stanley et al., 2008; Allaert et al., 2020), and in the process of implicit associations (Marini et al., 2018). However, a future contralateral tDCS protocol would exceptionally account for the evidence deriving from the *Right-Hemisphere Hypothesis* (Levy et al., 1983), and the *Valence-Specific Hypothesis* (Davidson et al., 1987) and would be of great interest for addressing the ongoing debate in this field of emotional processing.

Concerning the race of the face stimuli, results indicate that L-DPLFC tDCS reduced the gender-related difference in the ratings for black faces observed in the Sham stimulation condition. Previous studies have shown the potential of tDCS to modulate the "*Other-race effect (ORE)*" (Costantino et al., 2017). This is a cognitive domain in which lateral cortical regions - and particularly the DLPFC – have a key role in the inhibition of stereotype-consistent responses (e.g., implicit racial bias) (Forbes et al., 2012). In our study, there was a significant gender bias in the interaction between stimulation and race, with female participants giving higher ratings for black faces as compared to the male counterparts in the Sham group only. Interestingly, this effect was mitigated by the stimulation at both 1 and 2 mA. This supports the role of the L-DLPFC in biased cognition, and further investigation is needed to unveil the mechanisms involved in biased judgements in combination with affective aspects of face processing. Only a few studies explored gender differences in emotional processing through brain stimulation before. Conson et al. (2015) found that females were more accurate than males in recognizing all expressions, but anodal right/cathodal left stimulation of the DLPFC enhanced processing of fearful faces in male participants only. Differences in prefrontal activation patterns may contribute to the gender-specific effects of stimulation over the DLPFC (Weisenbach et al., 2014).

Exploring the cognitive and neurophysiological features of facial expression perception in relation to gender would be interesting also from a psychiatric perspective. Sex differences in emotion processing occur in affective disorders that characterise various neuropsychiatric conditions (e.g., autism, depression, and schizophrenia) (Frank, 2008; Halladay et al., 2015; Montagne et al., 2005), and should be investigated with the purpose to find new venues for treatment. In fact, from the '*Sex Differences in Brain, Behavior, Mental Health and Mental Disorders*' workshop of the National Institute of Mental Health (2011) emerged that there is a paucity of research examining sex differences at a neurobiological and mechanistic level, and a need for more neuroscientists to incorporate "gender" as a variable in experimental designs was identified (McCarthy et al., 2012). Concerning the use of brain stimulation for treatment, tDCS has shown its efficacy in addressing clinically significant emotion recognition deficits associated with depression, with the majority of protocols targeting the L-DLPFC (Bennabi and Haffen, 2018; Brennan et al., 2017). The methodological variability in this field implies a need for treatments optimization, especially through the implementation of personalized protocols.

Our results provide also support for differential efficacies of tDCS at different current intensities. As a matter of fact, we found a trend-wise effect with higher significance in the tDCS_2 mA group as compared to tDCS_1 mA, revealing potential linearity between current injection increase and behavioural outcomes. Since previous evidence has demonstrated non-linearity between current intensity and behaviour (at least in the Motor Cortex) (Agboada et al., 2019; Hassanzahraee et al., 2020), unravelling this aspect may have important methodological and theoretical implications for the implementation of effective neuromodulation interventions.

4.3. Implicit bias

Our results provide no evidence for an effective modulation of implicit racial preferences through anodal L-DLPFC stimulation. In fact, no significant effects emerged from the analyses on the IATs. Previous fMRI studies have revealed the DLPFC, together with other prefrontal regions, to be involved in social decision-making processes, and stereotypic attitudes (Marini et al., 2018; Wood and Grafman, 2003). Knutson et al. (2007) demonstrated that the L-DLPFC is recruited during the inhibition of complex social associations, specifically automatic beliefs about race and gender. In spite of this evidence, L-DLPFC stimulation via TMS and tDCS has led to inconsistent results: bias increase and both improvement and decrease of reaction times in congruent and incongruent conditions were observed (Crescentini et al., 2014; Gladwin et al., 2012). Variability in tasks and study designs may have led to those inconsistencies in the literature, and be the reason for the divergence between our results from those in tDCS studies targeting the same area. To the best of our knowledge, this is the first study investigating L-DLPFC tDCS for a racial IAT. Future studies should replicate the protocol with higher sample sizes, and with the implementation of a between-subjects design in which the effects are not subject to interindividual variability. Thus, further research is needed to specify the precise role of this area in the prefrontal hub for stereotypical implicit bias.

A potential limitation of the current study consists of the exclusion of the stimuli' gender from the main analysis of the manuscript. Given both the complexity of a *five-way* interaction and the increased risk for *Type II* errors, the analysis including stimuli' gender has been reported in the 'Supplementary material' section only. Critically, results confirm the significant interactions reported and discussed in the manuscript.

4.4. Conclusions and future directions

Our results support the feasibility of tDCS as an effective non-invasive brain stimulation tool to probe the role of the DLPFC in emotional processing. Our results provide evidence for L-DLPFC involvement in gender-related differences in the processing of emotional faces. Given that facial expression of emotions is a core topic in the field of affective and social neuroscience, with outstanding questions that still have no answer, future studies should test the same protocol with higher sample sizes, and potentially targeting different nodes of the "facial-expression network". In addition, multimodal neuroimaging studies, adopting for instance concomitant electroencephalography (EEG) recordings, will allow gaining a better insight into the neurophysiological bases of the observed behavioural outcomes.

Declarations

Author contribution statement

Annalisa Palmisano: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Francesco Bossi: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

Cecilia Barlabà, Francesco Febbraio, Riccardo Loconte, Antonella Lupo: Performed the experiments.

Michael A. Nitsche, Davide Rivolta: Conceived and designed the experiments; Analyzed and interpreted the data; Wrote the paper.

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Data availability statement

Data will be made available on request.

Declaration of interests statement

The authors declare no conflict of interest.

Additional information

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