Received: 31 January 2020 | Accepted: 24 July 2020

DOI: 10.1111/ejn.14932

RESEARCH REPORT

EJN European Journal of Neuroscience FENS WILEY

A multimodal study of the effects of tDCS on dorsolateral prefrontal and temporo-parietal areas during dichotic listening

Lynn Marquardt[1,2] | **Isabella Kusztrits**[1,2] | **Alexander R. Craven**[1,2,3] | **Kenneth Hugdahl**[1,2,4,5] | **Karsten Specht**[1,6,7] | **Marco Hirnstein**[1,2]

[1]Department of Biological and Medical Psychology, University of Bergen, Bergen, Norway

[2]NORMENT Center of Excellence, University of Bergen, Haukeland University Hospital, Bergen, Norway

[3]Department of Clinical Engineering, Haukeland University Hospital, Bergen, Norway

[4]Department of Radiology, Haukeland University Hospital, Bergen, Norway

[5]Division of Psychiatry, Haukeland University Hospital, Bergen, Norway

[6]Mohn Medical and Imaging Visualization Centre, Haukeland University Hospital, Bergen, Norway

[7]Department of Education, UiT/The Arctic University of Norway, Tromsø, Norway

Correspondence
Lynn Marquardt, Department of Biological and Medical Psychology, University of Bergen, Jonas Lies vei 91, 5009 Bergen, Norway.
Email: lynn.marquardt@uib.no

Funding information
Bergen Research Foundation, Grant/Award Number: BFS2016REK03; European Research Council (ERC), Grant/Award Number: 693124

Abstract

The underlying neural mechanisms of transcranial direct current stimulation (tDCS), especially beyond the primary motor cortex, remain unclear. Several studies examined tDCS effects on *either* functional activity, neurotransmitters *or* behavior but few investigated those aspects together to reveal how the brain responds to tDCS. The objective is to elucidate the underlying mechanisms of tDCS using a multimodal approach that extends from behavioral to neurotransmitter levels of explanation. Thirty-two healthy participants performed an auditory dichotic listening task at two visits, one session with sham and one session with real tDCS (2 mA) while simultaneously undergoing functional magnetic resonance imaging (fMRI). The anode and cathode were placed over the left temporo-parietal cortex (TPC) and dorsolateral prefrontal cortex, respectively. Before and after simultaneous dichotic listening/fMRI/tDCS, combined glutamate and glutamine (Glx) and myo-inositol levels were assessed in the stimulated areas. While fMRI and dichotic listening showed expected functional activity and behavioral effects, neither method demonstrated differences between real and sham stimulation. Glx only showed a statistical trend towards higher levels after real tDCS in both stimulated brain areas. There were no significant correlations between behavior and Glx. Despite a reasonable sample size, electrical field strength, and replication of behavioral and functional activity results, tDCS had little to no effect on dichotic listening, Glx, and functional activity. The study emphasizes that findings about the underlying neural mechanisms of the primary motor cortex cannot simply be generalized to other brain areas. Particularly, the TPC might be less sensitive to tDCS. Moreover, the study demonstrates the general feasibility of multimodal approaches.

Abbreviations: DLPFC, dorsolateral prefrontal cortex; fMRI, functional magnetic resonance imaging; GABA, γ-aminobutyric acid; Glx, glutamine and glutamate; MR, magnetic resonance; MRS, magnetic resonance spectroscopy; NAA, N-acetylaspartate; PRESS, point resolved spectroscopy sequence; SNR, signal to noise ratio; tDCS, transcranial direct current stimulation; TE, echo time; TPC, temporo-parietal cortex; TR, time to repeat.

Edited by Gregor Thut

KEYWORDS
electric field simulation, functional magnetic resonance imaging, magnetic resonance spectroscopy, transcranial direct current stimulation

1 | INTRODUCTION

Despite substantial progress, the underlying neural mechanisms of transcranial direct current stimulation (tDCS) are still not well understood. In humans, the effects of tDCS are typically studied with respect to behavior (Ditye, Jacobson, Walsh, & Lavidor, 2012; Westwood, Olson, Miall, Nappo, & Romani, 2017), brain activity (assessed with functional magnetic resonance imaging, fMRI) (Antal et al., 2012) and neurotransmitters/neurometabolites (Kim, Stephenson, Morris, & Jackson, 2014; Stagg et al., 2009). For instance, several studies investigated tDCS effects on gamma-aminobutyric acid (GABA) and glutamate, the main inhibitory and excitatory neurotransmitters, respectively. Some reported an increase of Glx (glutamate + glutamine) levels after anodal stimulation (Clark, Coffman, Trumbo, & Gasparovic, 2011; Hunter et al., 2015) and a Glx decrease after cathodal stimulation (Stagg et al., 2009). However, there are also null findings on glutamate or Glx (Antonenko et al., 2017; Dwyer et al., 2018; Kim et al., 2014). Another study found a significant increase in myo-inositol under the anode (Rango et al., 2008). Myo-inositol is a carbocyclic sugar, derived from glucose and involved in signal transmission in the brain. Others have examined tDCS effects on fMRI measures in motor or cognitive tasks. For example, Antal, Polania, Schmidt-Samoa, Dechent, & Paulus (2011) found a significant decrease of the blood oxygen level-dependent signal in the supplementary motor cortex when participants performed a finger-tapping task and were stimulated with anodal tDCS over the primary motor cortex. Weber, Messing, Rao, Detre, & Thompson-Schill (2014) reported changes in brain connectivity, as assessed with fMRI, due to tDCS during a risk assessment paradigm.

However, few studies looked at tDCS effects on behavior, brain activity and neurotransmitters *together*, although such a multimodal, neuroscientific approach may be more promising to reveal the associations between different aspects of brain functioning (Hunter, Coffman, Trumbo, & Clark, 2013; Tremblay et al., 2014). For instance, participants in Antonenko et al. (2017) received tDCS over the sensorimotor cortex during resting-state fMRI and GABA levels were measured before and after. The results showed reduced GABA levels after anodal tDCS compared to sham. Another study by the same group found that both anodal and cathodal tDCS decreased GABA levels and increased sensorimotor network connectivity and the tDCS induced changes in GABA levels correlated with the simulation of the tDCS electric field strength (Antonenko et al., 2019).

Studies investigating the underlying mechanisms of tDCS often focus on stimulation of the primary sensory/motor cortex (Antal et al., 2011, 2012; Antonenko et al., 2019; Kim et al., 2014; Stagg et al., 2009). We aimed to extend that work and examined the left dorsolateral prefrontal cortex (DLPFC) and left temporo-parietal cortex (TPC) with respect to tDCS effects on behavior, neurotransmitter levels and functional brain activity—within the same study. We chose those two brain areas to test a model that seeks to explain auditory hallucinations in patients with schizophrenia by assuming that *hyper*active temporo-parietal areas give rise to auditory hallucinations and *hypo*active prefrontal areas limit an individual's capacity to control the hallucinations (Hugdahl, 2009, 2015). Tentative evidence for the model comes from treatment studies where anodal tDCS over the prefrontal areas (with a supposedly excitatory effect) and cathodal stimulation of temporal areas (with a supposedly inhibitory effect) reduced hallucinations in patients with schizophrenia (Brunelin et al., 2012).

We aimed to test the model in healthy individuals using the Bergen dichotic listening task, in which simple speech sounds are presented to the left and right ear (Hugdahl et al., 2009). It was chosen because it is a reliable and well-established behavioral paradigm that involves both the DLPFC and the TPC, as revealed by functional neuroimaging (Westerhausen, Kompus, & Hugdahl, 2014). Behaviorally, it produces a right ear advantage that is modulated by participants' attention (Hugdahl, 2004). This right ear advantage and attentional modulation are typically reduced in schizophrenia patients (Hugdahl et al., 2013; Ocklenburg, Westerhausen, Hirnstein, & Hugdahl, 2013). Thus, by placing the excitatory anode over the TPC and the inhibitory cathode over the DLPFC in healthy individuals, we intended to "mimic" the reduced right ear advantage/generally fewer correct responses and the corresponding hypertemporal/hypofrontal activity pattern in schizophrenia patients as a test for the model.

More specifically, we hypothesized that excitatory, anodal stimulation of the left TPC and inhibitory, cathodal stimulation of the left DLPFC would lead to higher and lower levels of Glx, respectively. Functional brain activity would increase in the left TPC due to anodal excitation and Glx increase, and decrease in the left DLPFC, during tDCS as compared to sham. The right ear advantage would be reduced due to interference caused by increased Glx levels and reduced Glx levels in the left TPC and DLPFC, respectively. Based on findings showing Glx increase under the anode and decrease under the cathode (Clark et al., 2011; Stagg et al., 2009), we predicted that Glx in the left DLPFC

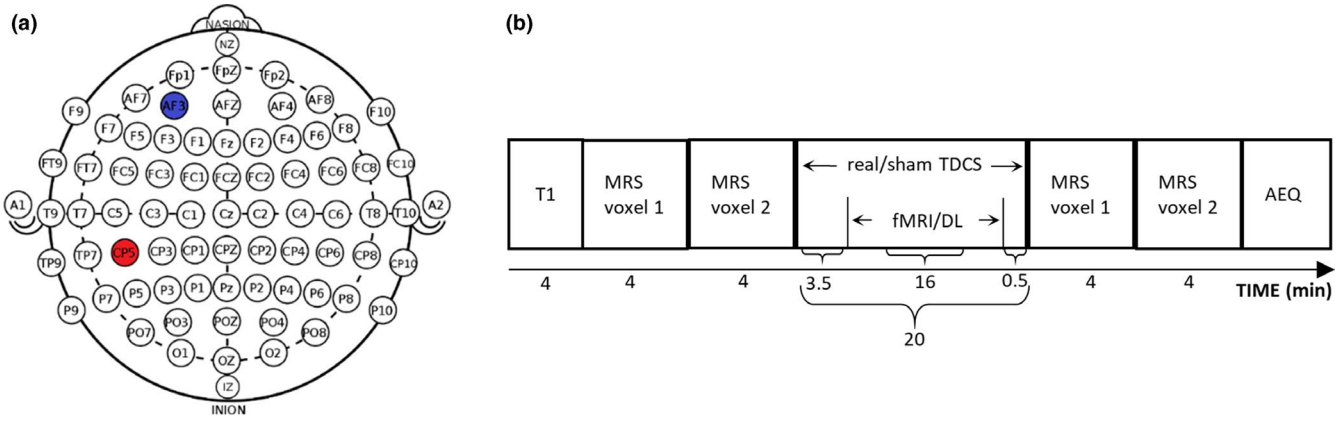

FIGURE 1 Electrode Montage and Experimental setup of one visit. Panel a) The cathode (blue) was placed over AF3 and the anode (red) over CP5. Panel b) Participants completed a dichotic listening task while undergoing simultaneous tDCS/fMRI. Before and afterwards, MR spectroscopy was performed in both stimulated areas. T1 was a structural scan that was used for modelling the electric field. At the end, participants completed an adverse effects questionnaire. Each participant visited twice, receiving once sham and once real tDCS.

(cathode) should be correlated negatively with a stronger, more focal electric field. In turn, Glx concentrations in the left TPC (anode) should be correlated positively with a stronger and focal electric field.

2 | METHODS

2.1 | Participants

Initially, 38 participants were recruited via flyers and word-of-mouth at the Haukeland University Hospital, Bergen, Norway. Exclusion criteria were past/present neurological or psychological disorders, head trauma, metallic implants, epilepsy in first degree relatives, pregnancy, claustrophobia, acute consumption of drugs or alcohol at time of testing, and severe skin diseases in the area of the electrode placement. Six participants had to be removed from the analysis due to incomplete data ($n = 1$), insufficient magnetic resonance spectroscopy (MRS) quality, ($n = 4$) and incorrect stimulation protocol ($n = 1$).

The mean age of the remaining 32 participants (18 male/14 female) was 26 ± 4.8 years (range = 20–39). Participants had a mean of 16 ± 2 years of education. All participants were screened for hearing deficits and could detect frequencies between 250 and 3,000 Hz at an intensity of <20 dB. Further, none of the participants had an interaural acuity difference of more than 10 dB (see also Hirnstein, Hugdahl, & Hausmann, 2014; Hirnstein, Westerhausen, Korsnes, & Hugdahl, 2013). All participants gave written informed consent in accordance with the Declaration of Helsinki and were reimbursed for their participation. The study was approved by the Regional Committee for Medical Research Ethics in Western Norway (REK Vest) # 2013/2342.

2.2 | Procedure

The dichotic listening paradigm was carried out during fMRI to assess tDCS effects on functional brain activity. Moreover, immediately before and after simultaneous tDCS/fMRI/dichotic listening, participants underwent MRS to measure glutamate. Finally, we took inter-individual differences in electric field parameters into account through simulation of tDCS effects based on structural MR scans.

A reporting checklist with an overview of the study's design, following the recommendations by Buch et al. (2017), is provided in the Appendix S1. Participants were tested twice, once with real and once with sham stimulation in a counter-balanced double-blind design. Fifteen participants received real, 17 sham tDCS in the first session. The real and sham tDCS sessions were separated by 8.4 ± 3.2 days on average (range: 4–16 days).

Only at the first session, participants provided informed consent and completed the hearing test as well as the dichotic listening task practice trials. In both sessions, they completed questionnaires concerning tDCS and MR safety, and electrode positions for tDCS were located with EEG caps (EASYCAP GmbH), based on the 10/20 system (Figure 1a), before entering the MR scanner. Rectangular, MR compatible tDCS electrodes made of rubber (5 cm × 7 cm) were used. The cathode and anode were placed over AF3 (left DLPFC) and CP5 (left TPC), respectively. Electrodes were coated with conductive paste Ten20 (Weaver and Company) and a 9 mg/ml NaCl solution to decrease impedance and attached to the scalp via a rubber band. Impedance was kept below 14.2 kΩ, which was tested outside the MR scanner.

After the impedance check, participants entered the GE 750 3T Scanner. In both sessions, the MR sequences

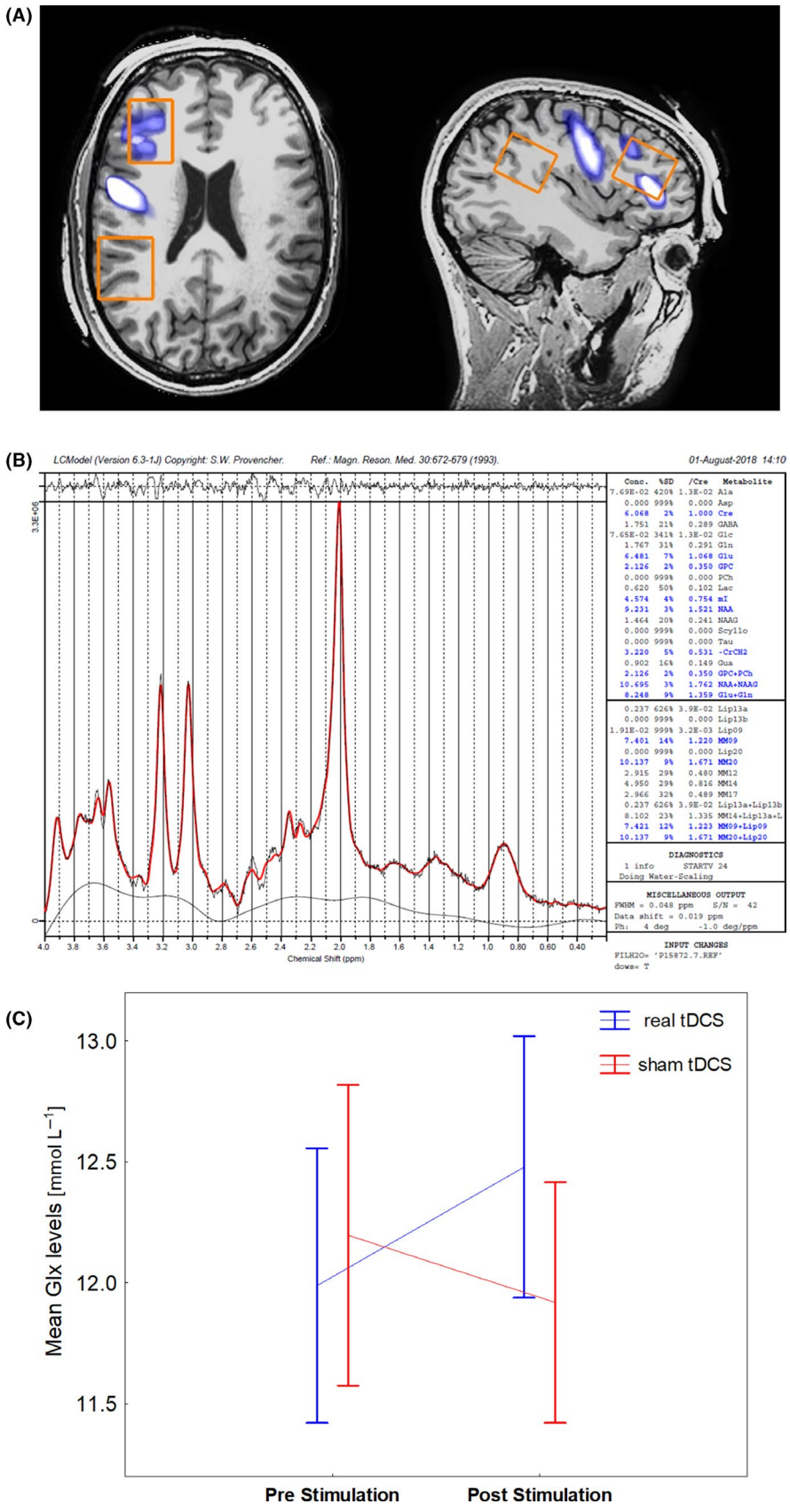

FIGURE 2 MR spectroscopy setup and results. Panel a) Voxel placement during MRS acquisition of the DLPC and TPC (sagittal and axial view) from one participant (in orange) and the simulated peak activation, threshold at 0.48 V/m for the illustration, as a group average (in blue). Panel b) Typical successfully acquired MRS spectrum as given by LCModel. The black line denotes the measured data, the red line the model. Concentration estimates for the different neurotransmitters are given in the right-hand box. Panel c) Trend towards increased Glx levels after real tDCS as compared to sham tDCS. Vertical bars denote 95% confidence intervals

were completed in the order as described below (see also Figure 1b). For all details regarding MR acquisition, quality control and hardware/software, please see Appendix S1.

2.2.1 | Structural MRI

After a localizer sequence, participants underwent a structural anatomical image 3D T1-weighted fast-spoiled gradient

sequence. The structural MR scan was carried out first for placing the voxels for the subsequent MRS and allowed electric field parameter simulations.

2.2.2 | MR spectroscopy

The structural scan was followed by two single-voxel point resolved spectroscopy (PRESS) sequences. Two voxels were placed, based on the T1 images, in the left DLPFC and the left TPC (Figure 2a). After the simultaneous tDCS/fMRI/dichotic listening sequence, MRS was performed again in both voxels. The voxel order was identical before and after the simultaneous tDCS/fMRI/dichotic listening sequence. In the second session, voxel order was reversed. Seventeen participants began with the left TPC, and 15 participants began with left DLPFC in the first session. The order was randomized, meaning, seven participants who began with the left TPC started with real and 10 started with sham. For the DLPFC, eight participants started with real and seven participants started with sham.

2.2.3 | tDCS

After MRS, the electrode cables were connected to the inner box and stimulation began. Codes were used to ensure double-blinding. tDCS lasted 20 min (+30 s ramp up and 30 s ramp down) at 2 mA (current density = 0.057 mA/cm) from an MR compatible DC-Stimulator Plus (neuroConn GmbH). Sham tDCS was delivered for 40 s, followed by very weak pulses of 110 μA lasting 15 ms, provided every 550 ms as an impedance check.

2.2.4 | Dichotic listening fMRI paradigm

During tDCS, participants completed a dichotic listening paradigm that was adapted to fMRI. It lasted 16 min and began 3.5 min after tDCS had started to ensure the left TPC and DLPFC had already been stimulated for a while (Figure 1b). In each dichotic listening trial, two out of six different syllables (/ba/,/da/,/ga/,/pa/,/ta/ and/ka/) are presented simultaneously, one to each ear. For example,/ba/ to the left ear and/ka/ to the right ear. Homonyms (e.g., /ba/-/ba/) were not included, leaving 30 possible syllable combinations. Participants completed these 30 trials twice, in three different conditions: In the non-forced condition, participants were instructed to verbally report the syllable they heard best and most clearly. In the forced-left and forced-right condition, they were instructed to specifically report the stimulus from the left and right ear, respectively. Verbal responses were scored and recorded during scanning as a measure of behavioral data.

The dichotic listening paradigm was carried out in a block design during fMRI acquisition, using an echo-planar imaging sequence. The paradigm had 270 volumes in total, distributed across 25 blocks (seven resting blocks + six non-forced + six forced-right + six forced-left). The block order was pseudo-randomized. Each block consisted of 10 trials, resulting in 180 dichotic listening volumes/trials and 70 resting volumes (Hugdahl & Andersson, 1986; Hugdahl et al., 2009; Thomsen, Rimol, Ersland, & Hugdahl, 2004). A silent gap, a delay until the following scan, was provided after each volume for presenting the stimuli and for recording the verbal responses from the dichotic listening task (van den Noort, Specht, Rimol, Ersland, & Hugdahl, 2008).

After the dichotic listening task, participants waited for 90 s in a quiet position until tDCS terminated. Then, the electrode cables were detached from the inner electrode box and the two remaining PRESS sequences were carried out. One participant with dyslexia was removed from the analysis including dichotic listening data because dyslexia might be associated with aberrant hemispheric asymmetry and/or performance in the forced attention conditions (Breznitz & Misra, 2003; Thomson, 1976).

2.2.5 | Adverse side effects

Side effects were measured with the tDCS Adverse Effects Questionnaire (Brunoni et al., 2011) after both sham and real tDCS sessions (Appendix S1).

2.3 | Data analysis

SPSS Statistics (version 25) and Statistica (version 13.3) were used for statistical analysis.

2.3.1 | Dichotic listening and fMRI

Correctly identified syllables were transformed into accuracy rates and subjected to a 2 × 3 × 2 repeated measures ANOVA with the within-participants variables *Stimulation* (real/sham), *Dichotic Listening Condition* (non-forced, forced-right, forced-left) and *Ear* (left/right). Similarly for the fMRI group analysis, individual contrast images were subjected to a 2 × 3 repeated measures ANOVA with *Stimulation* (real/sham) and *Dichotic Listening Condition* (non-forced, forced-right, forced-left). A mean contrast was estimated for illustrating the overall activation pattern across all conditions and for comparisons with earlier studies. This was supplemented with differential and interaction contrasts. For more details regarding preprocessing of

fMRI data in SPM12 (https://www.fil.ion.ucl.ac.uk/spm/), see Appendix S1. For dichotic listening and spectroscopy data, estimated marginal means are provided.

2.3.2 | MR spectroscopy

Water-scaled, tissue-content-adjusted Glx levels from LCModel (Appendix S1) were subjected to a 2 × 2 × 2 repeated measures ANOVA with the within-participants factors *Stimulation* (real/sham), *Time* (before/after tDCS) and *Brain area* (DLPFC/TPC). In four participants, MRS data from one voxel did not meet the data quality requirements. To retain the data from the other voxel with sufficient quality, we additionally calculated two *Time* x *Stimulation* ANOVAs separately for the left DLPFC ($n = 35$) and TPC ($n = 33$). For explorative reasons, we also ran the aforementioned 2 × 2 × 2 ANOVA with choline, creatine, myo-inositol and NAA levels.

2.3.3 | Simulation of electrical field during tDCS

Simulation (done in SimNIBS 2.1.2, Simulation of NIBS [non-invasive brain] stimulation [Version 2.1.2, Software] available from www.simnibs.org) of the tDCS electrical field in each participant was done based on their real tDCS session. To run the model, the electrodes in the simulation were placed over the real electrodes on the participants' head model. The simulated electrodes were 5 × 7 cm^2, like the real ones, with a 1 mm electrode thickness and 3 mm gel. The electric field strength (in [V/m]) and the focality (in cubic mm) of the stimulation were calculated for the entire cortex and the peak activation field (10 mm sphere). For field strength, 99% of the norm of the electric field and for focality the gray matter volume with an electric field greater or equal to 75% of the peak value are reported. Means and *SD* were calculated.

2.3.4 | Relationship between changes in Glx, dichotic listening accuracy, field strength, and focality

We computed a normality test for all variables and found some with non-normal distribution; hence, Spearman Rank correlations were computed between Glx and myo-inositol levels from before and after real stimulation as well as changes in Glx/myo-inositol levels (as calculated with Glx/myo-inositol$_{pre\text{-}tDCS}$ minus Glx/myo-inositol$_{post\text{-}tDCS}$), separately for DLPFC and TPC, with (a) field strength and focality from the simulation data and (b) the total number of correct responses in the non-forced, forced-right and forced-left condition. All measures were taken from the real tDCS session. As tDCS had no significant effect on fMRI data (see below), no correlations involving fMRI data were computed.

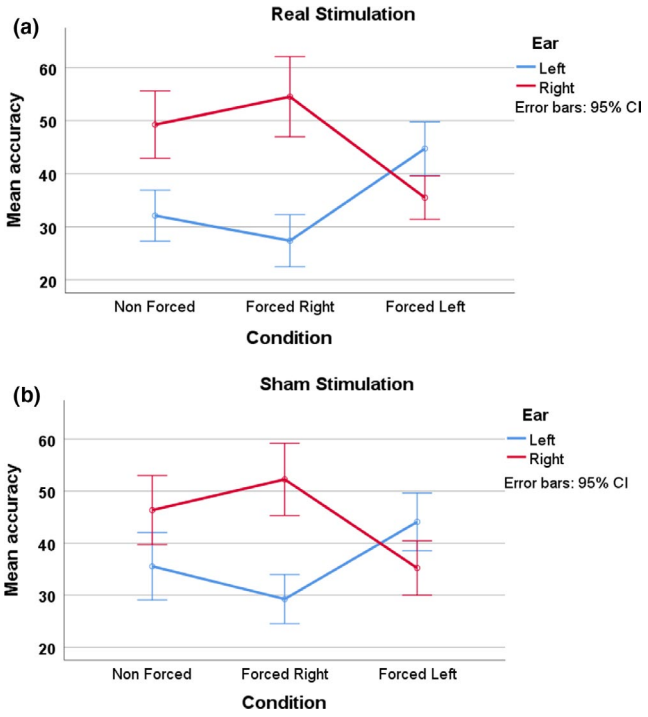

FIGURE 3 Mean accuracy scores (estimated marginal means) for dichotic listening. Panel a) Real tDCS stimulation. Panel b) sham stimulation.

3 | RESULTS

3.1 | Simultaneous Dichotic Listening and fMRI paradigm

3.1.1 | Dichotic listening behavior

The behavioral data revealed a significant main effect of Ear ($F_{(1,30)} = 5.63$, $p = .024$, $\eta_p 2 = .158$), showing that participants reported more syllables correctly from the right ($M = 45.51 \pm 13.86$) than the left ear ($M = 35.51 \pm 10.91$). There was also a significant Condition*Ear interaction ($F_{(1,30)} = 40.28$, $p < .0001$, $\eta_p^2 = .573$) with a substantial right ear advantage in the non-forced and forced-right condition, while a left ear advantage emerged in the forced-left condition (Figure 3). However, neither the main effect of Stimulation nor any interaction involving Stimulation reached significance (all $Fs \leq 1.64$, $ps \geq .203$, $\eta_p^2 s \leq .052$).

3.1.2 | fMRI

For the fMRI data, a mean contrast across all variables in the ANOVA (Figure 4a) showed activity in the auditory

cortex and the left DLPFC. Moreover, we found a main effect of Condition, showing two main significant clusters, one in the left cerebral white matter and precuneus (location in mm: $x = -10$ $y = -60$ $z = 52$, cluster level: #voxel = 1,739, p(FWE) < 0.001, peak: $F_{(2,186)} = 20.73$ p(FWE) < 0.001) and one in the right lingual gyrus and cerebellum exterior (location in mm: $x = 10$ $y = -64$ $z = -8$, cluster level: #voxel = 285, p(FWE) < 0.001, peak: $F_{(2,186)} = 21.71$, p(FWE) < 0.001; Figure 4b). There was also a significant cluster in the forced-right versus forced-left contrast in the right lingual gyrus and cerebellum exterior (location in mm: $x = 10$ $y = -64$ $z = -8$, cluster level: #voxel = 377, p(FWE) <0.001, peak: $T_{(1,186)} = 6.30$, p(FWE) < 0.001; Figure 4c). No significant suprathreshold clusters emerged for the main effect of Stimulation or the interaction between Conditions*Stimulation (all $Ts \leq 2.66$, $p_{\text{FWE-corr}} \geq 0.999$, $p_{\text{uncorr}} \geq 0.004$).

3.1.3 | MR spectroscopy

Glx showed a trend for a Stimulation*Time interaction ($F_{(1,31)} = 3.35$, $p = .077$, $\eta_p^2 = .098$). Glx levels were higher after tDCS than before when participants received real tDCS, while there was a very minor decrease during sham tDCS (Figure 2c). However, exploratory post hoc t-tests (unadjusted) did not find a significant difference between before and after real tDCS ($p = .109$) and sham tDCS ($p = .356$). As there was no significant three-way interaction ($F_{(1,31)} = 0.002$, $p = .961$, $\eta_p^2 < .001$), this Glx change did not differ between left TPC and DLPFC. Except for a main effect of *Brain area* ($F_{(1,31)} = 14.19$, $p = .001$, $\eta_p^2 = .314$), with higher Glx levels in the left DLPFC ($M = 11.45 \pm 1.47$) as compared to the left TPC ($M = 12.84 \pm 1.47$), no other main effect or interaction reached significance (all $F \leq 0.367$, $p \geq .549$, $\eta_p^2 \leq .012$). Likewise, there were no significant main effects or interactions in the 2 × 2 ANOVA for either left DLPFC or TPC (all $F \leq 2.013$, $p \geq .166$, $\eta_p^2 \leq .059$).

None of the other metabolites or parameters (choline, creatine and NAA) showed a significant main effect or interaction involving the factor Stimulation, except for myo-inositol, where a Stimulation*Time interaction emerged ($F_{(1,31)} = 4.59$, $p = .040$, $\eta_p^2 = .129$). Real tDCS led to an increase from $M = 5.34 \pm 0.53$ I.U. to $M = 5.47 \pm 0.57$ I.U., while there was a small decrease in sham tDCS from $M = 5.40 \pm 0.72$ I.U. to $M = 5.29 \pm 0.65$ I.U., uncorrected post hoc tests showed neither were significant. The difference between real and sham tDCS after stimulation was significant ($p = .032$).

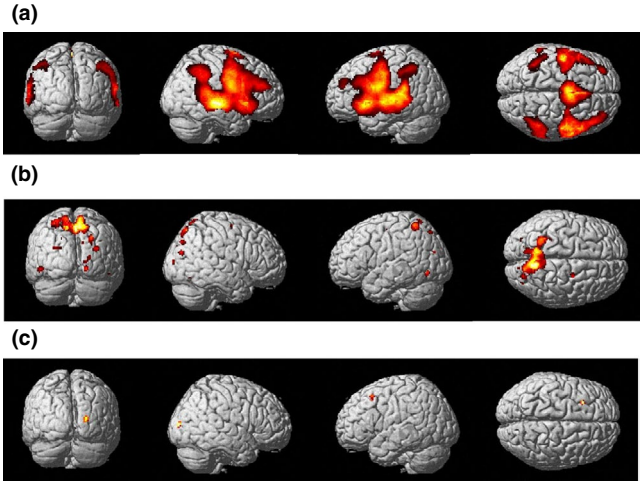

FIGURE 4 fMRI activity during dichotic listening. Panel a) Contrast across all variables in ANOVA. Panel b) Main effect dichotic listening condition. Panel c) Contrast forced-right vs. forced-left condition.

3.2 | Simulation of electrical field during tDCS

The simulated electric field strengths of all participants were strongest in the left central sulcus region and Broca's area, though with considerable inter-individual differences (Appendix S1). For the full cortex, the 99% peak field was $M = 0.65 \pm 0.096$ V/m and 75% focality was $M = 9{,}716 \pm 2045$ mm^2. For the Peak, the 99% peak field was $M = 0.77 \pm 0.144$ V/m and 75% focality was $M = 274 \pm 142$ mm^2.

3.3 | Correlations

Glx levels before/after tDCS as well as Glx changes between before/after did not correlate with either dichotic listening (all rs ≤ .345, ps ≥ .057) or simulated field strength and focality (all rs ≤ .234, ps ≥ .197). Similarly, myo-inositol levels before/after tDCS as well as myo-inositol changes did not correlate with dichotic listening (all rs ≤ −.271, ps ≥ .140). Myo-inositol changes did not correlate with simulated field strength and focality (all rs ≤ −.197, ps ≥ .280). There was one significant correlation, uncorrected for multiple testing, between focality of the simulated field and myo-inositol levels in the TPC before tDCS ($r \leq -.422$, $p \geq .016$), which would not withstand Bonferroni correction. All other correlations between myo-inositol levels before/after tDCS with focality and simulated field strength were not significant (all rs ≤ −.177, ps ≥ .332).

3.4 | Blinding and adverse side effects

When asked after the second session to indicate when they received real stimulation, 42% of participants responded

incorrectly (blinding data from one participant was missing). A binominal test found no statistically significant difference from 50% chance level ($p = .473$), implying that the blinding worked by and large. Results on adverse side effects are reported in the Appendix S1.

3.5 | Power analysis

A G*Power analysis (Faul, Erdfelder, Buchner, & Lang, 2009) suggests that to obtain a significant Time*Stimulation interaction with $n = 32$, one would need a medium effect size of $f = 0.26$ (with the settings: power = 0.80, $\alpha = .05$, number of groups = 1, number of measurements = 2, corr among rep measures = 0.5, nonsphericity correction = 1).

4 | DISCUSSION

The present study aimed to elucidate the underlying mechanisms of tDCS effects with a multimodal approach in areas beyond the rather well-researched primary motor/sensory cortex (Antal et al., 2012; Antonenko et al., 2019; Kim et al., 2014; Stagg et al., 2009). We expected a reduced right ear advantage/fewer correct responses in dichotic listening and increased Glx levels/functional activity in the TPC as well as reduced Glx levels/functional activity in the DLPFC during tDCS as compared to sham. However, we found no effects of tDCS on behavior and functional activity and only a trend towards a Glx increase after tDCS. There were only very weak correlations between Glx/myo-inositol levels and dichotic listening and simulated electrical field parameters, if any.

The mean contrast across all dichotic listening fMRI conditions replicated previous findings: Behaviorally, a right ear advantage arose that was modulated by instructions to focus attention on either the left or right ear stimulus (Bless et al., 2013; Hugdahl & Hammar, 1997). We further replicated increased functional activity in typical fronto-temporo-parietal language perception and attention areas (Noort et al., 2008; van den Kompus et al., 2012). Crucially, however, neither dichotic listening performance nor fMRI activity was significantly affected by tDCS. The negative behavioral performance results are in line with a previous study that did not find tDCS effects on dichotic listening after anodal and cathodal stimulation over the left auditory cortex (D'Anselmo, Prete, Tommasi, & Brancucci, 2015). tDCS effects on functional activity during dichotic listening have not been investigated before. In the primary motor cortex, tDCS also did not affect fMRI activity but led to reduced activity in the adjacent supplementary motor cortex (Antal et al., 2011). Finally, while our null findings do not support the hypertemporal/hypofrontal model (Hugdahl, 2015), they also do not invalidate it.

The weak increase in Glx after tDCS was independent of the electrode/brain area. This is inconsistent with findings showing increased Glx levels only after anodal stimulation (2 mA) of the right parietal cortex (Clark et al., 2011; Hunter et al., 2015) or decreased Glx levels only after cathodal tDCS (1 mA) of the motor cortex (Stagg et al., 2009). Our finding is in line, however, with other studies that failed to detect tDCS induced changes in Glx in the primary motor and sensorimotor cortex, posterior superior temporal gyrus and cerebellar cortex at both 1 and 2 mA (Antonenko et al., 2017; Dwyer et al., 2018; Jalali, Chowdhury, Wilson, Miall, & Galea, 2018; Kim et al., 2014; Zappasodi et al., 2017). Another study found increased Glx levels in the striatum during tDCS (1 mA) over the left and right DLPFC (Hone-Blanchet, Edden, & Fecteau, 2016). The inconsistent results are likely to arise from differences in stimulation intensity and electrode location, for instance. However, spurious findings with small samples also constitute a problem: some tDCS/spectroscopy studies have sample sizes around $n = 10$, which is plainly underpowered as recently demonstrated (Sanaei Nezhad et al., 2020).

The most parsimonious explanation for the weak dichotic listening behavioral, Glx and fMRI effects is that the electric current was too low to induce meaningful changes. However, there was a significant, electrode-independent increase of myo-inositol levels, in line with a previous study (Rango et al., 2008). Moreover, glutamate changes were reported in the sensorimotor cortex (Antonenko et al., 2019) and on motor learning in tDCS over primary motor cortex (Naros et al., 2016) with 1 mA—thus, in principal, lower electric field strength than in the present study. Finally, since the correlations between electric field parameters and dichotic listening performance as well as the MRS measures were either non-significant (or would become non-significant if adjusted for multiple testing), stronger electric field parameters might not have necessarily produced stronger tDCS effects.

Another possibility is test power. Our study has a reasonable sample size compared to previous studies. We cannot conclude that tDCS with the parameters described here has no effect at all. However, if it exists, the effect is likely to be small (at best medium) according to our power analysis.

A third possibility is that we failed to detect significant tDCS effects because the peak of the electric field was between the two electrodes, and not in the stimulated left DLPFC and TPC itself. While this could explain the lack of clear Glx-results, it is difficult to reconcile with the significant increase of myo-inositol levels in the left DLPFC and TPC, and with the fact that we did not observe any changes in functional activity in the left central sulcus/Broca's area.

Meta-analyses showed that cathodal tDCS in the DLPFC has little effect on cognitive tasks (Dedoncker, Brunoni,

Baeken, & Vanderhasselt, 2016) and that the cathode rarely induces inhibitory effects in cognitive tasks (Jacobson, Koslowsky, & Lavidor, 2012). In the auditory and posterior temporal cortex, performance in dichotic listening (D'Anselmo et al., 2015) as well as in reading and naming tasks (Westwood et al., 2017) was found to be unaffected by tDCS, and anodal stimulation of the posterior superior temporal gyrus did not change Glx levels (Dwyer et al., 2018). On the other hand, anodal tDCS over the temporo-parietal junction had behavioral effects on reality monitoring (Mondino, Poulet, Suaud-Chagny, & Brunelin, 2016) and tDCS over the DLPFC yields promising findings with respect to depression treatment (Mutz, Edgcumbe, Brunoni, & Fu, 2018; Palm, Hasan, Strube, & Padberg, 2016). Taken together, the DLPFC and TPC can evidently be affected by tDCS, but given the considerable body of null findings together with the present findings, it seems that at least the posterior temporal-parietal region might be less responsive to tDCS than, for instance, the primary sensory/motor cortex. This might make tDCS treatments targeting posterior temporal-parietal areas more challenging (e.g., in schizophrenia or tinnitus).

4.1 | Limitations

Since the structural MR scans were taken with the electrodes on, the electrodes were included in the modelling of the electric field parameters as part of the head. While this affects the thickness of skin and skull in the model, it does so for all participants and should not meaningfully affect the results of the simulation (G.B. Saturnino, A. Thielscher, personal communication, April 08, 2019). We placed the MRS voxels as closely under the electrodes as possible to measure tDCS effects. However, moving the voxel deeper into the brain, especially in the TPC, would have given better MRS measurements. We further avoided high CSF involvement by adding saturation bands, but this does not protect against signal loss from participants' movements.

Moreover, whilst the sample size is relatively large for a study of this nature, it is still small for correlational work and, finally, the effect of stimulation intensity needs further elaboration. A recent study (Samani, Agboada, Jamil, Kuo, & Nitsche, 2019) showed that at 2 mA the cathode might not have an inhibitory but excitatory effect—which is in fact in line with our increased Glx levels under the cathode. Thus, cathodal stimulation at 1 mA could have yielded different results.

In conclusion, we found at best weak effects of tDCS over the left DLPFC and TPC on behavior, glutamate, and functional activity. This is unlikely due to insufficient electric current but, together with other findings, could reflect that the stimulated regions, especially the left TPC, are less sensitive to tDCS than primary sensory/motor areas. Although such weak findings are naturally limited in terms of their scientific contribution, we still think they are relevant—especially in the field of brain stimulation, which due to its fast growth and popularity is sometimes subject to findings that raise replication issues: First, the present study further emphasizes that findings from the primary sensory/motor cortex cannot easily be generalized to other brain regions. Second, it demonstrates that multimodal approaches that combine behavioral with multiple neuroscientific assessments are feasible, in principle. Such studies are rare to date, but clearly have the potential to deepen our understanding the underlying mechanisms of tDCS.

ACKNOWLEDGEMENT

The current research has been funded by the Bergen Research Foundation (grant BFS2016REK03). We want to thank all our participants for their help and patience, Clovis Nugame and Thea Sveen for their help with the dichotic listening analysis, and Guliherme Saturnino and Axel Thielscher for their help with the simulation. The contribution of Kenneth Hugdahl was funded by a grant from the European Research Council (ERC), # 693124.

CONFLICT OF INTEREST

The authors Kenneth Hugdahl, Karsten Specht, and Alexander R. Craven have stock in NordicNeuroLab (NNL) AS, which produced MR accessories used during data acquisition. Otherwise there are no conflicts of interest.

AUTHOR CONTRIBUTIONS

Conception and design: MH, KH, IK and LM. Acquisition of data: MH, IK, AC, and LM. Analysis and interpretation, writing of article: MH, AC, KP and LM. Critical review of article and agreement to be accountable for all aspects of the work: MH, KP, KH, IK, AC, and LM.

DATA AVAILABILITY STATEMENT

Anonymized data will be made available to colleagues upon request to the corresponding author.

PEER REVIEW

The peer review history for this article is available at https://publons.com/publon/10.1111/ejn.14932

ORCID

Lynn Marquardt https://orcid.org/0000-0003-4247-0070

REFERENCES

Antal, A., Kovacs, G., Chaieb, L., Cziraki, C., Paulus, W., & Greenlee, M. W. (2012). Cathodal stimulation of human MT+ leads to elevated fMRI signal: A tDCS-fMRI study. *Restorative Neurology and Neuroscience*, *30*, 255–263. https://doi.org/10.3233/RNN-2012-110208

Antal, A., Polania, R., Schmidt-Samoa, C., Dechent, P., & Paulus, W. (2011). Transcranial direct current stimulation over the primary motor cortex during fMRI. *NeuroImage*, *55*, 590–596. https://doi.org/10.1016/j.neuroimage.2010.11.085

Antonenko, D., Schubert, F., Bohm, F., Ittermann, B., Aydin, S., Hayek, D., … Flöel, A. (2017). tDCS-induced modulation of GABA levels and resting-state functional connectivity in older adults. *Journal of Neuroscience*, *37*, 4065–4073. https://doi.org/10.1523/JNEUROSCI.0079-17.2017

Antonenko, D., Thielscher, A., Saturnino, G. B., Aydin, S., Ittermann, B., Grittner, U., & Flöel, A. (2019) Towards precise brain stimulation: Is electric field simulation related to neuromodulation? *Brain Stimulation*, *12*(5), 1159–1168.

Bless, J. J., Westerhausen, R., Arciuli, J., Kompus, K., Gudmundsen, M., & Hugdahl, K. (2013). "Right on all occasions?"–On the feasibility of laterality research using a smartphone dichotic listening application. *Frontiers in Psychology*, *4*, 42.

Breznitz, Z., & Misra, M. (2003). Speed of processing of the visual–orthographic and auditory–phonological systems in adult dyslexics: The contribution of "asynchrony" to word recognition deficits. *Brain and Language*, *85*, 486–502. https://doi.org/10.1016/S0093-934X(03)00071-3

Brunelin, J., Mondino, M., Gassab, L., Haesebaert, F., Gaha, L., Suaud-Chagny, M.-F., … Poulet, E. (2012). Examining transcranial direct-current stimulation (tDCS) as a treatment for hallucinations in schizophrenia. *American Journal of Psychiatry*, *169*(7), 719–724. https://doi.org/10.1176/appi.ajp.2012.11071091

Brunoni, A. R., Amadera, J., Berbel, B., Volz, M. S., Rizzerio, B. G., & Fregni, F. (2011). A systematic review on reporting and assessment of adverse effects associated with transcranial direct current stimulation. *International Journal of Neuropsychopharmacology*, *14*, 1133–1145. https://doi.org/10.1017/S1461145710001690

Buch, E. R., Santarnecchi, E., Antal, A., Born, J., Celnik, P. A., Classen, J., … Cohen, L. G. (2017). Effects of tDCS on motor learning and memory formation: A consensus and critical position paper. *Clinical Neurophysiology*, *128*, 589–603. https://doi.org/10.1016/j.clinph.2017.01.004

Clark, V. P., Coffman, B. A., Trumbo, M. C., & Gasparovic, C. (2011). Transcranial direct current stimulation (tDCS) produces localized and specific alterations in neurochemistry: A 1H magnetic resonance spectroscopy study. *Neuroscience Letters*, *500*, 67–71. https://doi.org/10.1016/j.neulet.2011.05.244

D'Anselmo, A., Prete, G., Tommasi, L., & Brancucci, A. (2015). The dichotic right ear advantage does not change with transcranial direct current stimulation (tDCS). *Brain Stimulation*, *8*, 1238–1240. https://doi.org/10.1016/j.brs.2015.09.007

Dedoncker, J., Brunoni, A. R., Baeken, C., & Vanderhasselt, M.-A. (2016). A systematic review and meta-analysis of the effects of transcranial direct current stimulation (tDCS) over the dorsolateral prefrontal cortex in healthy and neuropsychiatric samples: Influence of stimulation parameters. *Brain Stimulation*, *9*, 501–517. https://doi.org/10.1016/j.brs.2016.04.006

Ditye, T., Jacobson, L., Walsh, V., & Lavidor, M. (2012). Modulating behavioral inhibition by tDCS combined with cognitive training. *Experimental Brain Research*, *219*, 363–368. https://doi.org/10.1007/s00221-012-3098-4

Dwyer, G. E., Craven, A., Hirnstein, M., Kompus, K., Assmus, J., Ersland, L., … Grüner, R. (2018). No effects of anodal tDCS on local GABA and Glx levels in the left posterior superior temporal gyrus. *Frontiers in Neurology*, *9*, 1145. https://doi.org/10.3389/fneur.2018.01145

Faul, F., Erdfelder, E., Buchner, A., & Lang, A.-G. (2009). Statistical power analyses using G* Power 3.1: Tests for correlation and regression analyses. *Behavior Research Methods*, *41*, 1149–1160. https://doi.org/10.3758/BRM.41.4.1149

Hirnstein, M., Hugdahl, K., & Hausmann, M. (2014). How brain asymmetry relates to performance–a large-scale dichotic listening study. *Frontiers in Psychology*, *4*, 997. https://doi.org/10.3389/fpsyg.2013.00997

Hirnstein, M., Westerhausen, R., Korsnes, M. S., & Hugdahl, K. (2013). Sex differences in language asymmetry are age-dependent and small: A large-scale, consonant–vowel dichotic listening study with behavioral and fMRI data. *Cortex*, *49*, 1910–1921. https://doi.org/10.1016/j.cortex.2012.08.002

Hone-Blanchet, A., Edden, R. A., & Fecteau, S. (2016). Online effects of transcranial direct current stimulation in real time on human prefrontal and striatal metabolites. *Biological Psychiatry*, *80*, 432–438. https://doi.org/10.1016/j.biopsych.2015.11.008

Hugdahl, K. (2004). 12 dichotic listening in the study of auditory laterality. *The Asymmetrical Brain*, *441*. 441–444.

Hugdahl, K. (2009). "Hearing voices": Auditory hallucinations as failure of top-down control of bottom-up perceptual processes. *Scandinavian Journal of Psychology*, *50*, 553–560.

Hugdahl, K. (2015). Auditory hallucinations: A review of the ERC "VOICE" project. *World Journal of Psychiatry*, *5*, 193. https://doi.org/10.5498/wjp.v5.i2.193

Hugdahl, K., & Andersson, L. (1986). The "forced-attention paradigm" in dichotic listening to CV-syllables: A comparison between adults and children. *Cortex*, *22*, 417–432. https://doi.org/10.1016/S0010-9452(86)80005-3

Hugdahl, K., & Hammar, Å. (1997). Test-retest reliability for the consonant-vowel syllables dichotic listening paradigm. *Journal of Clinical and Experimental Neuropsychology*, *19*, 667–675. https://doi.org/10.1080/01688639708403752

Hugdahl, K., Nygård, M., Falkenberg, L. E., Kompus, K., Westerhausen, R., Kroken, R., … Løberg, E.-M. (2013). Failure of attention focus and cognitive control in schizophrenia patients with auditory verbal hallucinations: Evidence from dichotic listening. *Schizophrenia Research*, *147*, 301–309. https://doi.org/10.1016/j.schres.2013.04.005

Hugdahl, K., Westerhausen, R., Alho, K., Medvedev, S., Laine, M., & Hämäläinen, H. (2009). Attention and cognitive control: Unfolding the dichotic listening story. *Scandinavian Journal of Psychology*, *50*, 11–22. https://doi.org/10.1111/j.1467-9450.2008.00676.x

Hunter, M. A., Coffman, B. A., Gasparovic, C., Calhoun, V. D., Trumbo, M. C., & Clark, V. P. (2015). Baseline effects of transcranial direct current stimulation on glutamatergic neurotransmission and large-scale network connectivity. *Brain Research*, *1594*, 92–107. https://doi.org/10.1016/j.brainres.2014.09.066

Hunter, M. A., Coffman, B., Trumbo, M., & Clark, V. (2013). Tracking the neuroplastic changes associated with transcranial direct current stimulation: A push for multimodal imaging. *Frontiers in Human Neuroscience*, *7*, 495.

Jacobson, L., Koslowsky, M., & Lavidor, M. (2012). tDCS polarity effects in motor and cognitive domains: A meta-analytical review. *Experimental Brain Research*, *216*, 1–10. https://doi.org/10.1007/s00221-011-2891-9

Jalali, R., Chowdhury, A., Wilson, M., Miall, R. C., & Galea, J. M. (2018). Neural changes associated with cerebellar tDCS studied using MR spectroscopy. *Experimental Brain Research*, *236*, 997–1006. https://doi.org/10.1007/s00221-018-5170-1

Kim, S., Stephenson, M. C., Morris, P. G., & Jackson, S. R. (2014). tDCS-induced alterations in GABA concentration within primary motor cortex predict motor learning and motor memory: A 7 T magnetic resonance spectroscopy study. *NeuroImage*, *99*, 237–243. https://doi.org/10.1016/j.neuroimage.2014.05.070

Kompus, K., Specht, K., Ersland, L., Juvodden, H. T., van Wageningen, H., Hugdahl, K., & Westerhausen, R. (2012). A forced-attention dichotic listening fMRI study on 113 subjects. *Brain and Language*, *121*, 240–247. https://doi.org/10.1016/j.bandl.2012.03.004

Mondino, M., Poulet, E., Suaud-Chagny, M.-F., & Brunelin, J. (2016). Anodal tDCS targeting the left temporo-parietal junction disrupts verbal source-monitoring. *Neuropsychologia*, *89*, 478–484.

Mutz, J., Edgcumbe, D. R., Brunoni, A. R., & Fu, C. H. (2018). Efficacy and acceptability of non-invasive brain stimulation for the treatment of adult unipolar and bipolar depression: A systematic review and meta-analysis of randomised sham-controlled trials. *Neuroscience and Biobehavioral Reviews*, *92*, 291–303. https://doi.org/10.1016/j.neubiorev.2018.05.015

Naros, G., Geyer, M., Koch, S., Mayr, L., Ellinger, T., Grimm, F., & Gharabaghi, A. (2016). Enhanced motor learning with bilateral transcranial direct current stimulation: Impact of polarity or current flow direction? *Clinical Neurophysiology*, *127*, 2119–2126. https://doi.org/10.1016/j.clinph.2015.12.020

Ocklenburg, S., Westerhausen, R., Hirnstein, M., & Hugdahl, K. (2013). Auditory hallucinations and reduced language lateralization in schizophrenia: A meta-analysis of dichotic listening studies. *Journal of the International Neuropsychological Society*, *19*, 410–418. https://doi.org/10.1017/S1355617712001476

Palm, U., Hasan, A., Strube, W., & Padberg, F. (2016). tDCS for the treatment of depression: A comprehensive review. *European Archives of Psychiatry and Clinical Neuroscience*, *266*, 681–694. https://doi.org/10.1007/s00406-016-0674-9

Rango, M., Cogiamanian, F., Marceglia, S., Barberis, B., Arighi, A., Biondetti, P., & Priori, A. (2008). Myoinositol content in the human brain is modified by transcranial direct current stimulation in a matter of minutes: A 1H-MRS study. *Magnetic Resonance in Medicine*, *60*, 782–789.

Samani, M. M., Agboada, D., Jamil, A., Kuo, M.-F., & Nitsche, M. A. (2019). Titrating the neuroplastic effects of cathodal transcranial direct current stimulation (tDCS) over the primary motor cortex. *Cortex*, *119*, 350–361. https://doi.org/10.1016/j.cortex.2019.04.016

Sanaei Nezhad, F., Lea-Carnall, C. A., Anton, A., Jung, J., Michou, E., Williams, S. R., & Parkes, L. M. (2020). Number of subjects required in common study designs for functional GABA magnetic resonance spectroscopy in the human brain at 3 Tesla. *European Journal of Neuroscience*, *51*, 1784–1793. https://doi.org/10.1111/ejn.14618

Stagg, C. J., Best, J. G., Stephenson, M. C., O'Shea, J., Wylezinska, M., Kincses, Z. T., … Johansen-Berg, H. (2009). Polarity-sensitive modulation of cortical neurotransmitters by transcranial stimulation. *Journal of Neuroscience*, *29*, 5202–5206. https://doi.org/10.1523/JNEUROSCI.4432-08.2009

Thomsen, T., Rimol, L. M., Ersland, L., & Hugdahl, K. (2004). Dichotic listening reveals functional specificity in prefrontal cortex: An fMRI study. *NeuroImage*, *21*, 211–218. https://doi.org/10.1016/j.neuroimage.2003.08.039

Thomson, M. (1976). A comparison of laterality effects in dyslexics and controls using verbal dichotic listening tasks. *Neuropsychologia*, *14*, 243–246. https://doi.org/10.1016/0028-3932(76)90054-3

Tremblay, S., Beaulé, V., Proulx, S., Lafleur, L.-P., Doyon, J., Marjańska, M., & Théoret, H. (2014). The use of magnetic resonance spectroscopy as a tool for the measurement of bi-hemispheric transcranial electric stimulation effects on primary motor cortex metabolism. *Journal of Visualized Experiments*, *93*, 51631. https://doi.org/10.3791/51631

van den Noort, M., Specht, K., Rimol, L. M., Ersland, L., & Hugdahl, K. (2008). A new verbal reports fMRI dichotic listening paradigm for studies of hemispheric asymmetry. *NeuroImage*, *40*, 902–911. https://doi.org/10.1016/j.neuroimage.2007.11.051

Weber, M. J., Messing, S. B., Rao, H., Detre, J. A., & Thompson-Schill, S. L. (2014). Prefrontal transcranial direct current stimulation alters activation and connectivity in cortical and subcortical reward systems: A tDCS-fMRI study. *Human Brain Mapping*, *35*, 3673–3686. https://doi.org/10.1002/hbm.22429

Westerhausen, R., Kompus, K., & Hugdahl, K. (2014). Mapping hemispheric symmetries, relative asymmetries, and absolute asymmetries underlying the auditory laterality effect. *NeuroImage*, *84*, 962–970. https://doi.org/10.1016/j.neuroimage.2013.09.074

Westwood, S. J., Olson, A., Miall, R. C., Nappo, R., & Romani, C. (2017). Limits to tDCS effects in language: Failures to modulate word production in healthy participants with frontal or temporal tDCS. *Cortex*, *86*, 64–82. https://doi.org/10.1016/j.cortex.2016.10.016

Zappasodi, F., Musumeci, G., Navarra, R., Di Lazzaro, V., Caulo, M., & Uncini, A. (2017). Safety and effects on motor cortex excitability of five cathodal transcranial direct current stimulation sessions in 25 hours. *Neurophysiologie Clinique*, *48*(2), 77–87. https://doi.org/10.1016/j.neucli.2017.09.002

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

How to cite this article: Marquardt L, Kusztrits I, Craven AR, Hugdahl K, Specht K, Hirnstein M. A multimodal study of the effects of tDCS on dorsolateral prefrontal and temporo-parietal areas during dichotic listening. *Eur J Neurosci*. 2020;00:1–11. https://doi.org/10.1111/ejn.14932