Recommender's and Reviewer's comments = black
Authors' comments = blue

PCI-RR INVITATION TO REVISE

I have now received comments on this Stage 1 RR submission from two expert reviewers (R#1 has identified himself as Sam Westwood). Both reviewers are positive about the aims of the experiment, and have only relatively minor suggestions to make. In particular, I would draw your attention to SW's second point, which concerns the possible additional between-session variability introduced into your within-subjects design by practice effects. Before you embark on this experiment (with its present design), it would be worth carefully considering the possible impact of practice effects on your data and, crucially, on your ability to test your stated hypotheses at the desired level of power.

Your revision should be accompanied by a full responses-to-reviewers document, and a tracked version of the manuscript in which any changes made are highlighted.

Best wishes,

Rob McIntosh

PCI-RR recommender

Dear Dr. McIntosh,

We write with a revised manuscript following reviewer comments. We feel confident we have responded to both reviewers in full.

Following your feedback, we have paid particular attention to Dr. Westwood's comment on within-subject versus between-subject design strengths. Specifically, we think we should be able to replicate the tRNS effect found by Ghin et al. over hMT+ in a three session within-subjects design as their original finding was found in a four session within-subjects design (Experiment 1: four conditions: sham, tRNS, a-tDCS, and c-tDCS, all over hMT+). Therefore, the original study was able to show a stimulation effect in the context of any learning effects across session. We respond point-by-point to the reviewers' comments below.

Thank you for your consideration, we look forward to hearing from you.

Best,

Grace Edwards, Mica Carroll, and Chris Baker

Reviewer 1:

This is an interesting replication that will be a welcome contribution to the field - more basic research replications in tDCS are needed! Please see my revisions, which are mainly minor below.

We thank Dr. Samuel Westwood for the constructive feedback and address each comment below.

Major Revision

1. The authors should assess blinding integrity - i.e., were the subjects and experiments blinded successfully throughout the study. There are important questions raised about this by Horvath, Leamouth, and Cohen Kadosh with tDCS, and the same applies to tRNS. It might be enough to simply ask participants what group they think were assigned to, but assessing their confidence and the effect stimulation sensations had on their performance would be a more thorough assessment. But see work from the above three authors for more ideas! (One final thing, the saline solution NaCl concentration would be a useful detail to report as this can influence perception of the stimulation as well as the efficacy of stimulation.

Following the suggestion of Dr. Westwood, after every session we will ask the participants if they perceive any sensation (tingling, burning, or pain) under the electrodes during stimulation. Sensation experience should be a good measure for perceived impact of stimulation. These questions evoke a response about the sensation of the stimulation without engaging the participants too deeply in the expected behavioral outcome of each stimulation session. However, following the final session, we will ask participants to report if they received stimulation or not in that session, and how confident they are in their response. For the final session only, this report will also enable us to determine if participants can distinguish between sham, active stimulation targeted at hMT+, and active stimulation to the forehead in a between-subjects comparison. We have included these additional measures in the manuscript on p.14:

“Participants will be blinded to the type of stimulation applied in each session. To assess blinding integrity, we will ask participants if they perceive any sensation (tingling, burning or pain) under the electrodes after each session. Sensation experience should provide a measure for the perception of present or absent stimulation. Additionally, in the final session we will also ask participants to report if they received stimulation in that session and how confident they are in their report. This report will also enable us to determine if participants can distinguish between sham, active stimulation targeted at hMT+, and active stimulation to the forehead in a between-subjects comparison. These questions evoke a response about the perception of stimulation, reserving the direct question about stimulation presence to the end of the experiment and avoiding preconceptions about the presence of stimulation during the experimental protocol.”

We have also added the saline solution NaCl concentration (0.9%) to our manuscript (pg. 13).

2. I did not see any mention of the spacing between the three sessions? Only that the sessions are non-consecutive, but does mean a washout period of 24hours?

We apologize for the oversight and have added the following to the manuscript (pg. 14):

"There will be a minimum of 72 hours between each stimulation session for each participant."

Minor Revisions

3. It's probably worth thinking about the downsides of having a within-subjects design, and how this might hamper comparisons with Ghin. For example, presumably there will be test-retest (practice) effects, even after the first session, and therefore a significant effect of session. Given the small effect of TES has, practice effects might wipe out the tRNS effect. If you want to mitigate this, you could run a practice session (if Ghin did), but again this might mean participants eventually perform close to ceiling. If a significant effect of session is found, I forsee an exploratory analysis where only the groups are compared with the first session data only (turning the design into a between-subjects design), which might be - albeit underpowred - a better replication of Ghin since they used between-subjects. This is only a minor revision, but it's perhaps worth thinking about the impact this might have on your eventual interpretation. You're wise to counterbalance the order of sessions, but perhaps there is more to be considered at the analytical and write up stage now before your pre-register.

Ghin et al., 2018 found their effect of hf-tRNS over hMT+ on contralateral motion processing in experiment 1, which was a four session within-subjects design (including sham, a-tDCS, and c-tDCS). Ghin et al., would have likely experienced similar learning effects across sessions within Experiment 1, which leads us to believe that if the hf-tRNS effect is replicable, we should also be able to find the effect within any learning effects in a three session within-subjects design. Further, as suggested by Dr. Westwood, we have an initial screening assessment to determine if participants can perform the task to 70% accuracy, thereby enabling us to include them in the experiment. As most learning occurs from the first experience with the task to the second (Beglinger et al., 2005), by the time we introduce the stimulation sessions, learning effects should have less of an impact across session.

4. There are a number of differences from the original experiments in this replication. To make this transparent, I would recommend the otherwise present a table that includes key methodological details of the original and replication so that the reader can see where the replication is the same and different from the original. This is particular useful when one compares the protocol and the eligibility criteria between studies.

We have added the below table to our manuscript (pg. 17):

Difference	*Ghin et al.*	*Carroll et al.*
Experimental design	• Two independent experiments • Experiment 1: stimulation over hMT+ versus sham control • Experiment 2: stimulation over V1 versus sham, and stimulation over forehead versus sham	• Single experiment including all stimulation conditions within subject.

Number of participants	• Experiment 1: 16 participants • Experiment 2: 12 participants for each active and sham stimulation pairing	• 42 participants
Stimulation type	• Experiment 1: hf-tRNS, a-tDCS, c-tDCS, and sham • Experiment 2: hf-tRNS, and sham	• hf-tRNS and sham
Electrode placement	• Experiment 1: hMT+ and Cz • Experiment 2: left V1 and Cz; left forehead and Cz	• hMT+ and Cz • Left forehead and Cz
Electrode size	• hMT+, forehead & V1: 16 cm^2 • Cz: 60 cm^2	• hMT+ & Cz: 25 cm^2
Thresholding	• Performed on each of five blocks separately, and averaged at the end	• Performed across all five blocks. The final threshold estimated on data from all five blocks

Table 1: The methodological differences between the original Ghin et al., 2018 study and our proposed replication. Particular attention paid to Experiments 1 and 2 of the Ghin et al., 2018 study which we propose to replicate.

I sign all of my reviews

Samuel Westwood, PhD

Reviewer 2:

The study proposed by Carrol et al. sets to replicate the findings from Ghin et al. (2018), investigating the effects of tES on motion coherence threshold. In particular, the authors aim to replicate the modulatory effects of high-frequency tRNS applied over hMT+ on a global motion perception task. This study is also set to extend previous findings corroborating the effectiveness of tRNS on the visual system. Overall, the research questions and methodology were well exposed and detailed and I do not have any particular remark at this stage. Thus, I do only have minor comments.

We thank reviewer two for their constructive feedback and have addressed each point below.

A concept briefly mentioned on page 4 is the importance of assessing the flexibility of the stimulation procedures in producing similar outputs. The concept of “flexibility” might be an essential point in the study, and it could be made more explicit, maybe in the introduction or when defining the hypothesis. This is because the study proposed here presents some critical differences compared to Ghin et al. For example, the authors plan to apply the stimulation at 1.5mA (like in Ghin et al.). However, they also say that their electrode size will be 25cm2 (while

Ghin et al. had 16 and 60cm2 for the “active” and the “reference” electrode, respectively). The authors are probably aware that by changing the electric size (while maintaining the current intensity), the current density also changes along with the current injected into the cortex. Similarly, on page 11, the authors explain that they will run the MLP over 160 trials, unlike Ghin et al., in which the final threshold was averaged across five blocks. Again, I feel that this is an important difference between protocols. Thus, based on these differences, if the proposed hypotheses are confirmed, this will also provide indirect evidence into the versatility of tRNS in global motion perception that might be worth discussing.

Following feedback from both reviewers, we have included a table which highlights any differences between the original study from Ghin et al., and our replication (see response to reviewer one, comment four). We agree that if a replication were to occur, it would provide evidence of the versatility of tRNS montages. We also agree that a more explicit statement to the flexibility of the stimulation technique is warranted in the introduction, and we have added the below to pg. 4:

“For tRNS to be a widely applied method, the modulatory effect of tRNS should generalize despite slight alterations to the procedure.”

The paper from Giangregorio, 2022 (page 22) is not reported in the reference list.

We thank Reviewer two for noticing the missing reference and have added the below reference in the reference list:

Generoso Giangregorio (2022). Very fast Mutual Information betweentwo images. (https://www.mathworks.com/matlabcentral/fileexchange/36538-very-fast-mutual-information-betweentwo-images), MATLAB Central File Exchange. Retrieved September 7, 2022.

Although it is for the exploratory analysis, the fMRI session is not mentioned in the procedure section, which creates a bit of confusion while reading the proposal.

We have added the fMRI into the procedures for clarity (p.15):

“In addition to the stimulation sessions, participants will be brought in for an fMRI session to localize regions active for lateralized motion processing, including left hMT+ (Figure 3). These data will be used for our exploratory analysis comparing the activity maps of lateralized motion processing to anatomically defined stimulation sites.”

To my knowledge, the ROAST toolbox does not simulate random noise current (not sure about tACS either). If this is the case, I assume the simulation is based on direct current stimulation. Nevertheless, I am unaware of any toolbox that can simulate the electric field for tRNS (see also Ghin et al. 2018), and therefore I think this can be kept. However, I would still make explicit what type of stimulation is used for the simulation.

Following personal communication with Yu (Andy) Huang, one of the developers of the ROAST toolbox, we established ROAST was designed for quasi-static modelling when the frequency is lower than 1k Hz. This means the head is ohmic for the purpose of the simulation and is the same for direct or alternating current (Huang et al., 2019). We have added this detail to the manuscript (pg. 22).

References:

Beglinger, L. J., Gaydos, B., Tangphao-Daniels, O., Duff, K., Kareken, D. A., Crawford, J., Fastenau, P. S., & Siemers, E. R. (2005). Practice effects and the use of alternate forms in serial neuropsychological testing. *Archives of Clinical Neuropsychology*, *20*(4), 517–529. https://doi.org/10.1016/j.acn.2004.12.003

Generoso Giangregorio (2022). Very fast Mutual Information betweentwo images. (https://www.mathworks.com/matlabcentral/fileexchange/36538-very-fast-mutual-information-betweentwo-images), MATLAB Central File Exchange. Retrieved September 7, 2022.

Huang, Y., Datta, A., Bikson, M., & Parra, L. C. (2019). Realistic volumetric-approach to simulate transcranial electric stimulation—ROAST—a fully automated open-source pipeline. *Journal of Neural Engineering*, *16*(5), 056006. https://doi.org/10.1088/1741-2552/ab208d