

Protocol and Statistical Analysis Plan

Title: Movement-Associated tVNS and Responsiveness Testing for Personalized Rehabilitation

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Study protocol:

There will be only one group. All participants will wear surface electrodes on the outer ear on both sides and a finger device that monitors heart rate. The attachment sites are the cymba concha and tragus for tVNS and the ear lobe for sham. There will be 6 types of stimulation (3 locations x 2 conditions): unilateral left tVNS, unilateral right tVNS, bilateral tVNS, unilateral left sham, unilateral right sham, and bilateral sham. The electrical current will have a pulse duration of 0.1 ms at 30 Hz with an intensity below the pain threshold or 5 mA, whichever is lower, for a duration of 0.5 s. The experimental stimulation intensity will be determined as just below the pain threshold. Participants will receive 16 blocks of unilateral left, unilateral right, and bilateral stimulations to the sham or tVNS electrodes at rest, during which their pupil size will be recorded. Each block will contain one trial for all combinations of stimulation conditions and locations (6 trials) in random order. The pupil response will be trigger-averaged in reference to the time of stimulation within each condition-location combination.

Statistical analysis plan:

The following dependent variables will be obtained. 1) the peak pupil diameter changes relative to the baseline value and 2) the mean pupil diameter changes across the time points from the time point identified as the rising half-peak (time point with a pupil diameter change closest to half the peak value) to the falling half-peak. These variables will be tested with a two-way analysis of variance (ANOVA) with repeated measures, with factors being stimulation type (tVNS and sham) and stimulation location (unilateral left, unilateral right, and bilateral), followed by a post-hoc test, if appropriate. The alpha value of 0.05 will be employed. This test will clarify whether tVNS is effective and the efficacy is greater with bilateral tVNS.