Official Title: A Randomized, Double-Blind, Placebo-Controlled Pilot Study to Evaluate Non-invasive Brain Stimulation Effects on Orientation and Mobility Performance in Adults with Visual Impairments.

Date: 17th December, 2025

Statistical Analysis Plan

Primary objective: The primary objective of this study is to determine if orientation and mobility (O&M) performance, namely percentage preferred walking speed (PPWS) (%) of patients with constricted visual fields in both eyes improves after receiving active hf-tRNS.

***Secondary objectives*:**

1. To determine the effect of active vs. sham hf-tRNS on the change in the following:
    - The number of O&M errors.
    - Visual detection distance (VDD).
    - Visual identification distance (VID).
2. To assess if a range of covariates (age, eye condition/disease, use of a white cane) have any moderating effects.

Table of Contents

1. Planned Study Components

1.1. Completed number of participants:

a. Waterloo: 0

1.2. Independent variables:

a. Stimulation type (active vs. sham stimulation)
b. Test time: Assessments were conducted at three time points: baseline (before stimulation), post-stimulation (immediately after stimulation (2 minutes), and post-stimulation (30-minutes after stimulation).

For statistical analysis, test time will be transformed into one level to account for the large inter-subject variabilities in baseline measures:

i. Post stimulation - baseline

1.3. Covariates:

a. Age (will be dichotomized)[1]
b. Eye condition/disease/degree of peripheral field constriction (will be dichotomized)[1]
c. Use of a white cane for travel versus no white cane for travel (will be dichotomized)[1]

1.4. Analysis framework and primary outcome measures:

a. 2 (stimulation type: active and placebo) x 2 (test time: post 2, post 30) Repeated measures of ANOVA (RMANOVA): Outcome is change in PPWS, that is, post-stimulation measurements – baseline measurement.

1.5. Analysis framework and secondary outcome measures

1.5.1 Outcomes analysis

a. 2 (stimulation type: active and placebo) x 2 (test time: post 2, post 30) Repeated measures of ANOVA (RMANOVA) analysis for change (post-stimulation measurements – baseline measurement) in each of the following outcomes:
    - The number of O&M errors.
    - Visual detection distance (VDD).
    - Visual identification distance (VID).

** Interactions: An interaction between Stimulation type and test time indicates that active and sham hf-tRNS have different enhancement effects on O&M performance, but these effects are not consistent across all test times. Therefore, the planned contrast for such an interaction is to perform individual comparisons between active vs. sham hf-tRNS at each test time.*

1.5.2 Integration of covariates into analysis

a. 2 (Stim type) x 2 (test time) analysis with age as covariate: Primary outcome, PPWS and the above secondary outcome measures.

[1] The listed covariates will be dichotomized using a median split. The "low" group will include participants with a score at or below the median, while the "high" group will consist of participants with a score above the median.

b. 2 (Stim type) x 2 (test time) analysis with eye condition/disease/degree of peripheral field constriction as covariate: Primary outcome, PPWS and the above secondary outcome measures.
c. 2 (Stim type) x 2 (Test time) analysis with use of white cane for travel versus no white cane for travel as covariate: Primary outcome, PPWS and the above secondary outcome measures.

1.5.3 Sensitivity analysis for varying degrees of constricted visual fields in both eyes if the groups is heterogenous.

a. Bootstrapping approach: To address the limited sample size of the participants with constricted visual fields in both eyes and evaluate the hf-tRNS effect across the heterogenous group, a bootstrapping method will be employed. A total of 1,000 bootstrap samples will be generated by random sampling with replacements from the existing subject pool. For each resampled dataset, the differential outcome improvements between active vs. sham hf-tRNS will be computed for different degrees of constricted visual fields. The distribution of differences will be analyzed to estimate the confidence intervals of the degree of constricted visual field effects.
b. Subgroup analysis: The outcome improvements for active vs. sham hf-tRNS will be compared within the group. The results from this subgroup analysis will be compared to the overall outcome measures of the entire study population.

1.5.4 Sensitivity analysis for use of white cane for travel.

c. Bootstrapping approach: To address the limited sample size of the participants who use a white cane for travel and evaluate the hf-tRNS effect, a bootstrapping method will be employed. A total of 1,000 bootstrap samples will be generated by random sampling with replacements from the existing subject pool. For each resampled dataset, the differential outcome improvements between active vs. sham hf-tRNS will be computed for white cane users and nonusers. The distribution of differences will be analyzed to estimate the confidence intervals of the white cane effects.
d. Subgroup analysis: The outcome improvements for active vs. sham hf-tRNS will be compared within the white cane nonusers. The results from this subgroup analysis will be compared to the overall outcome measures of the entire study population.

1.6. Additional details:

a. Participants with incomplete data will be excluded from the analysis.
b. All statistical tests will be performed as RMANOVA tests, using a significance level of alpha = 0.05.

2. Analysis Procedure

2.1. Assess whether the assumptions for RM-ANOVA are violated.

a. Sphericity of variances: Use Mauchly's Test of Sphericity to check for significant violations of sphericity.
    i. If the sphericity assumption is violated, apply the Greenhouse-Geisser correction for all main effects and interactions.
b. Homogeneity of variances: Use Levene's test to check for significant violations of homogeneity.
    i. If the homogeneity assumption is violated, employ the Mann-Whitney U test to do the pairwise comparison between active and sham stimulation.
c. Normality of residuals: Use Kolmogorov-Smirnov test to check for significant deviations from normality.
    i. If the normality assumption is not violated, proceed with parametric analysis (*Section 2.2*).
    ii. If the normality assumption is violated, proceed with nonparametric analysis (*Section 2.3*).

2.2 Parametric statistics

2.2.1 Primary analysis:

a. Perform repeated-measure ANOVA with test time and stimulation type as a within-subject factors for PPWS.
    i. If there is a significant interaction, apply pairwise comparisons to examine differential effects of stimulation.

2.2.2 Secondary analysis:

a. Perform repeated-measures ANOVA with test time and stimulation type as a within-subject factors for the outcomes listed in *Section 1.5.1.*
    i. If there is a significant interaction, apply pairwise comparisons to examine differential effects of stimulation.

b. Performed repeated-measures ANCOVA with test time and stimulation type as a within-subject factors, incorporating each of the covariates listed in *Section 1.5.2.*
    i. If any listed covariate effect is significant or significantly interacts with another factor, perform separate repeated-measure ANOVAs for each level of the covariate.

c. Perform sensitivity analysis using Bootstrapping approach and subgroup analysis as described in *Sections 1.5.3* and *1.5.4.*

2.3 Nonparametric statistics

2.3.1 Primary analysis:

a. Perform Aligned Rank Transform (ART) for nonparametric ANOVA with test time and stimulation type as a within-subject factors for PPWS.

i. If there is a significant interaction, apply pairwise comparisons to examine differential effects of stimulation.

*2.3.2 **Secondary analysis:***

a. Perform ART nonparametric ANOVA with test time and stimulation type as a within-subject factors for the outcomes listed in *Section 1.5.1.*
    i. If there is a significant interaction, apply pairwise comparisons to examine differential effects of stimulation.

b. Perform ART nonparametric ANOVA with test time and stimulation type as a within-subject factors, incorporating each of the covariates listed in *Section 1.5.2*.
    i. If any listed covariate effect is significant or significantly interacts with another factor, perform separate ART nonparametric ANOVAs for each level of the covariate.

c. Perform sensitivity analysis using Bootstrapping approach and subgroup analysis as described in *Sections 1.5.3* and *1.5.4.*

3. Data and demographics summary table

A. A summary table will be generated and will include:

1. Age (mean with SD).
2. Sex (in percentage).
3. White cane users (in percentage).
4. Eye condition/disease (retinitis pigmentosa (RP), rod-cone dystrophy, or advanced glaucoma).
5. Best corrected visual acuity (distance and near).
6. Degree of constricted visual field in both eyes.
7. Binocular contrast sensitivity.
8. Baseline PPWS.
9. Baseline VDD.
10. Baseline VID.
11. Baseline time taken to complete each course section.
12. Baseline number of O&M errors.

B. Supplementary demographic table

A demographic table will be generated, one row per participant. The columns will include:

1. Age.
2. Sex.
3. White cane users.
4. Eye condition/disease (retinitis pigmentosa (RP), rod-cone dystrophy, or advanced glaucoma).
5. Best corrected visual acuity (distance and near).
6. Degree of constricted visual field in both eyes.
7. Binocular contrast sensitivity.
8. Baseline PPWS.
9. Baseline VDD.
10. Baseline VID.
11. Baseline time taken to complete each course section.

12. Baseline number of O&M errors.

Reference:

Silva, A. E., Lyu, A., Leat, S. J., Khan, S., Labreche, T., Chan, J. C. H., . . . Thompson, B. (2024). Statistical analysis plan for NCT04762368, Imporoving vision in adults with macular degenration, study 2: The effect of concurrent perceptual learning and brain sitmulation. Internet Version. Retrieved on 15th December, 2025 from: https://cdn.clinicaltrials.gov/large-docs/68/NCT04762368/SAP_003.pdf