Cover Page: Statistical Analysis Plan

Official Title of Study: Stress and the Sympathetic Nervous System in Adults with Depression

NCT04838262

Document Date: R21MH123928 Notice of Award (03/23/2021)

4.3 Statistical Design and Power

Data Analytical Plan: Two psychosocial stressor exposure indicators will be calculated: stressor frequency (i.e., percentage of interview days during which at least one stressor occurred) and total stress (i.e., total number of stressors reported across all interview days). The emotion item ratings will be averaged to obtain daily negative affect scores, and scores will be aggregated for the 8 interview days. Negative affective reactivity will be defined as the magnitude of the change in negative affect on days when stressors occurred compared to negative affect on non-stressor days. Negative affective reactivity scores will be calculated using two-level multi-level modeling, as previously described[17, 18]. Muscle SNA will be quantified as burst occurrence (burst frequency) and size (burst area) and will be assessed during an overall baseline and in response to each laboratory-applied stressor. Beat-to-beat sympathetic transduction to the vasculature and to BP will be determined via spike-triggered averaging[19-22]. Although daily stressor exposure and negative affective reactivity are the primary outcomes, exploratory analyses will expand our model of daily stress processes to examine the heterogeneity of the types of stressors experienced (stressor diversity[23]) in relation to sympathetic reactivity in adults with MDD, which will provide additional insight to stress system dysfunction in depression and form the basis of subsequent proposals.

Statistical Approach: To ensure robust and unbiased results, we will use defined protocols for classifying and testing subjects. For all aims, multiple experimental controls and orthogonal approaches are used, and relevant biological variables are factored into the research design and analyses. Data analysis will be blinded, when applicable, and performed by two independent investigators. Statistical significance will be accepted at $p<0.05$. Given our strict inclusion criteria, in order to achieve the required sample size of MDD patients, we will over-sample young adults with MDD; healthy non-depressed adults will be recruited and matched on a 1:1 basis, such that an equivalent number of participants will be enrolled and tested in each group.

Multiple linear regression will be used to assess the relation between the primary outcome variable (sympathetic reactivity; continuous response variable) and two predictors: daily stressor processes (exposure and negative affective reactivity; continuous variables) and MDD (dichotomous between-group variable). The interaction between daily stressor processes and group will also be included in the model, such that:

$$\textit{sympathetic reactivity} = \beta_0 + \beta_1 * \textit{MDD} + \beta_2 * \textit{daily stressor process} + \beta_3 * \textit{MDD} * \textit{daily stressor process} + e$$

In this model, β_0 is the intercept, $\beta_{1/2/3}$ are the regression coefficients for each variable (group, daily stressor process, and the interaction), and *e* is the random error, assumed to be normally distributed, with a mean of zero and constant variance. We hypothesize β_3 will be significant such that the effect of daily stressor exposure (Aim 1) and negative affective reactivity (Aim 2) on sympathetic stress reactivity will be stronger in adults with MDD. All models will be tested with and without person mean stressor exposure to account for potential group differences in the frequency of daily stressor exposure. Sensitivity analyses will examine the effect of depressive symptom severity (continuous variable) instead of using the dichotomous MDD variable.

Sample size calculations are based on a power analysis for the squared semi-partial correlation between the response variable and each predictor that remains after controlling for the effect of the other predictor. Our preliminary data indicate the model $R^2=0.40$, with the two squared semi-partial correlation coefficients as 0.30 (stressor exposure) and 0.16 (group). Since a squared semi-partial correlation coefficient as low as 0.15 can be considered clinically relevant (medium effect size), a total sample size of 55 is a conservative estimate to detect a significant partial correlation between the response variable and each predictor after controlling for the effect of the other predictor (80% power at $\alpha=0.05$; t-test). For negative affective reactivity, a sample size of 48 is a conservative estimate to detect a significant interaction effect (80% power at $\alpha=0.05$; t-test). To account for attrition and potential experimental failure, we will test a total of 80 participants (40/group).