statistical analysis plan (sap)

A Feasibility Study Exploring the Benefits of Sudarshan Kriya Yoga in Young People with Symptoms of Anxiety or Depression (BREATHE

statistical analysis plan

Quantitative measures investigating psychopathology will be analysed via linear mixed effects model with restricted maximum likelihood estimator. All models will include random intercepts for participants and fixed effects for time, age, and gender. Quantitative measures investigating acceptability and feasibility will be analysed via t-test midway through the study (Day 15) with pre- and post- comparisons for the CEQ assessment. All quantitative measures will be accompanied by effect size analysis and 95% confidence intervals. Descriptive statistics will calculate mean values and standard deviations for normally distributed data and medians with range or percentage for non-normal and categorical data. Descriptive analysis of adverse and serious adverse events will be examined, including estimation of any causal relationships they may have had with adapted SKY-CHP or SKY practice.

Qualitative interviews will be recorded and transcribed and analysed using Interpretive Phenomenological Analysis. This process will involve data familiarization, generation of initial codes, theme identification, refining of themes, and theme names. Qualitative data analysis will be led by research team member JG, with theme confirmation and statistical support from JDK and AR.

## Collection & pre-processing of EEG data

The EEG acquisition including preparation and clean-up will take approximately 60 minutes. EEG is a non-invasive, painless procedure. EEG recordings will take place in a sound-attenuated and electrically shielded room at Orygen. Subjects will be seated in a slightly reclined chair with a head rest and asked to fixate their gaze on a cross presented on a computer monitor 2m in front of them. It is essential that participants feel comfortable at all times to avoid excessive movement that can lead to artefacts in the data. Participants will wear a cap with 64 electrodes. Electrodes are small metal disks which detect electrical brain activity from the scalp. The participants’ hair will be gently parted with a blunt-ended needle to make space between the electrode and the skin which will then be filled with conductive gel. Electrophysiological data will be primarily collected and analysed by a trained research team. EEG resting states will involve recording cortical electrical activity while the participant is resting in a seated position with their eyes closed. Data will be continuously sampled at 1000 Hz from the 64 electrodes on a Quick-Cap (Compumedics Limited, Australia Corporate HQ). EEG data will be amplified using the SynAMPRT and recorded using the CURRY 8 software (Compumedics Limited, Australia Corporate HQ). Recordings will be referenced to an electrode positioned between Cz and Cpz and the ground electrode to be located on the nasion (nose) of the participant. Data will be acquired from electrodes located on left and right mastoids, to facilitate a linked mastoid reference in off-line analysis. Electro-ocular gram (EOG) and electromyography (EMG) placement will allow for online muscle artefact detection and removal. Preferable electrode impedance will be set at 5 kΩ. Changes in spectral power (total and during resting EEG will be examined to establish any changes following the intervention. Spectral power will be calculated by Fast Fourier Transforming 1-second epochs of the resting EEG data with no overlapping, using a Hamming window with 10% taper with a resolution of 0.978Hz in a range of 500Hz. The data will be averaged within the following frequency bands: delta (δ) (1 - 3 Hz), theta (θ) (4 – 8 Hz), alpha (α) (9 – 13 Hz), beta (β) (14 – 30 Hz), and gamma (γ) (30 – 100 Hz). Total power will be calculated as 0 – 100 Hz for each electrode and averaged across all sites, and relative power will be derived by dividing absolute values within each frequency band by total power.

## Collection & pre-processing of electrophysiological markers

The mismatch negativity (MMN) paradigm is elicited using an auditory oddball paradigm using the following parameters: standard sounds (90% of trials; 1000 Hz; 50 ms); frequency-deviant sounds (10% of trials; 1200 Hz; 50 ms). 3000 stimuli will be presented in a pseudo-randomized order (never two frequency-deviants in a row) with a stimulus onset asynchrony of 500 ms. All stimuli will be presented binaurally through professional headphones (Sennheiser HD 280 Pro) while participants sit in a sound-proof room. Total time taken to complete these tasks will be up to 60 minutes. Pre-processing of electrophysiological data will be band-pass filtered from 1 to 35 Hz and down sampled to 250 Hz. All data sets will be corrected for eye-blink and movement artefacts. The selected epochs will be averaged for each of the different stimuli and each of the participants separately. To parameterize AOD and MMN effects in response to deviant stimuli, difference waves are calculated between event-related potentials (ERPs) evoked by standard stimuli and those evoked by duration and frequency deviants, respectively. Peak amplitudes of the MMN are determined at the frontal electrode Fz and defined as the amplitude of the maximum negative deflection of the difference waves occurring within 80–200 ms after stimulus onset.

To determine any effects of SKY on EEG and ERP, a linear mixed-effects regression model will be conducted. In this model, time (pre/post) will serve as a categorical predictor variable, and EEG and ERP changes as the outcome variable. This design allows for all individuals, even those who have been lost to follow-up, to be included in the analysis. Possible confounding effects of factors such as age, gender, and concomitant medication will be evaluated. Statistical significance will be interpreted at the alpha (a) level of 0.05. Given the preliminary nature of these analyses, results will not be corrected for multiple testing.