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# **Effects of one session of theta or high alpha neurofeedback on EEG activity and working**

#### **memory**

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#### **Abstract**

Neurofeedback techniques provide participants immediate feedback on neuronal signals, enabling them to modulate their brain activity. This technique holds promise in unveiling brain-behavior relationship, and offers opportunities for neuroenhancement. Establishing causal relationships between modulated brain activity and behavioral improvements requires rigorous experimental designs, including appropriate control groups and large samples. Our primary objective was to examine whether a single neurofeedback session, designed to enhance working memory through the modulation of theta or high-alpha frequencies, elicits specific changes in electrophysiological and cognitive outcomes. Additionally, we explored predictors of successful neuromodulation. One hundred and one healthy adults were assigned to groups trained to increase frontal theta, parietal high alpha, or random frequencies (active control group). We measured resting-state EEG, working memory performance, and self-reported psychological states before and after one neurofeedback session. Although our analyses revealed improvements in electrophysiological and behavioral outcomes, these gains were not specific to the experimental groups. An increase in the frequency targeted by the training has been observed for the theta and high alpha groups, but training aimed at increasing randomly selected frequencies appears to induce more generalized neuromodulation compared to targeting a specific frequency. Among all the predictors of neuromodulation examined, resting theta and high alpha amplitudes predicted specifically the increase of those frequencies during the training. These results highlight the challenge of integrating a control group based on enhancing randomly selected frequency bands and suggest potential avenues for optimizing interventions (e.g., by including a control group trained in both up- and down-regulation).

*Keywords:* neurofeedback, working memory, theta, high alpha, active control group

#### **Introduction**

The brain consists of numerous neurons forming intricate networks. Within these networks, information transmission occurs through rhythmic electrical activity known as neural oscillations (Buzsaki & Draguhn, 2004; Engel et al., 2001; Weisz & Keil, 2022). Neural oscillations play a crucial role in various cognitive processes (Başar et al., 2001), including sensory processing (Haegens & Golumbic, 2018), attention (Clayton et al., 2015) and working memory (Chikhi et al., 2022; Pavlov & Kotchoubey, 2022). Given the foundational role of neural oscillations in cognitive processes, modulating specific brain activity parameters holds promise for improving cognition (Chiasson et al., 2023; Gruzelier, 2014; Viviani & Vallesi, 2021) and elucidating the causal relationship between these parameters and cognitive processes (Kvamme et al., 2022a; Ramot & Martin, 2022).

Neurofeedback is a brain-computer interface that trains participants to voluntarily modulate a specific parameter of their brain activity through real-time feedback on their activity (Enriquez-Geppert et al., 2017; Sitaram et al., 2017). Participants engage in a closed loop where their mental actions (i.e., changing mental states) modify the feedback signal, subsequently influencing their forthcoming mental actions (Birbaumer et al., 2013; Lubianiker et al., 2022). Through repetition and trial-and-error learning, positive feedbacks that reward the mental state corresponding to the targeted neuronal state (e.g., increase of theta frequency above a certain threshold) will promote the reoccurrence of this mental state (Birbaumer et al., 2013; Gaume et al., 2016; Sherlin et al., 2011). Repeated production of the rewarded neuronal state may lead to neuroplasticity, strengthening synaptic connections within the underlying neural network (Davelaar, 2018; Shibata et al., 2019). Thus, the neurofeedback technique could enable individuals to learn to self-regulate their brain activity autonomously, without the aid of an external device (e.g., neurostimulation techniques; Herrmann et al., 2016). This can be achieved not only within controlled laboratory or clinical settings but also extended to real-world environments (Bassett & Khambhati, 2017; Herrmann et al., 2016).

In the field of EEG-neurofeedback, numerous studies have sought to increase the amplitude of two specific brain frequencies to enhance cognition: theta and alpha frequencies (Pfeiffer et al., 2024; Yeh et al., 2021; Yeh et al., 2022). The frontal theta frequency (4-8 Hz), intricately linked with cognitive control (Cavanagh, & Frank, 2014; Senoussi et al., 2022), has been targeted to improve attention (Brandmeyer & Delorme, 2020; Kerick et al., 2023), episodic memory (Eschmann et al., 2020; Tseng et al., 2021), autobiographic memory (Shoji et al., 2017), memory consolidation (Reiner et al., 2014; Rozengurt et al., 2016; Rozengurt et al., 2017; Shtoots et al., 2021), motor performance (Eschmann et al., 2022), working memory (Li et al., 2019; Reis et al., 2016; Wang & Hsieh, 2013) and executive functions (Enriquez-Geppert et al., 2014; Eschmann & Mecklinger, 2022; Smit et al., 2023). The posterior alpha frequency (8-12 Hz) serves as a cortical inhibitor, suppressing irrelevant information (Klimesch et al., 2007; Jensen & Mazaheri, 2010; Van Diepen et al., 2019) and contributing to working memory processes (Chen et al., 2023; Riddle et al., 2020). This frequency, and in particular the higher range of this frequency (referred to as high alpha, 10-12 Hz), has been a target of neurofeedback training aiming to enhance mental rotation (Hanslmayr et al., 2005; Zoefel et al., 2011), episodic memory (Guez et al, 2015), attention (Navarro Gil et al., 2018), executive control (Nawaz et al., 2022) and working memory processes (Bobby, & Prakash, 2017; Chen & Sui, 2023; Dehghanpour et al., 2018; Escolano et al., 2012; Escolano et al., 2014; Esteves et al., 2021; Hsueh et al., 2016; Naas et al., 2019; Shen et al., 2023; Takabatake et al., 2021; Wei et al., 2017). While the training duration varies across studies (from one to twenty), some research has indicated successful neuromodulation of the trained frequencies within a single session (Eschmann et al., 2022; Escolano et al., 2012; Escolano et al., 2014; Reiner et al., 2014; Rozengurt et al., 2016; Rozengurt et al., 2017; Shtoots et al., 2021). Although single neurofeedback sessions may not suffice for achieving mastery over one's brain activity, they do induce notable modifications in cortical excitability (Ros et al., 2010) as well as neuroplastic changes in structural and functional connectivity (Marins et al., 2019; Ros et al., 2013;

Sampaio-Baptista et al., 2021). These observations suggest that brief neurofeedback training can transiently affect brain activity and behavioral outcomes.

These effects are particularly interesting given the existing dilemma within the field of cognitive training. Indeed, a choice often has to be made between a large sample size or conducting a larger number of training sessions, due to limited resources. The field of neurofeedback is not exempt from these constraints, and numerous protocols suffer from insufficient statistical power. This often results in an inflation of effect sizes and low reproducibility of results (Button et al., 2013; Szucs & Ioannidis, 2017; Thibault & Pedder, 2022). In fact, a recent systematic review highlighted that only 4% of neurofeedback studies had sample sizes exceeding 50 participants, with an average number of participants per group of *n* = 16.64 (Chiasson et al., 2023). Hence, it is crucial for the neurofeedback field to rely on larger samples to rigorously evaluate the effects of this technique. Additional concerns arise regarding the potential influence of non-specific factors on neurofeedback outcomes. Non-specific factors encompass various extraneous variables that are not directly related to the neurofeedback protocol but can significantly contribute to the observed effects (Ros et al., 2020). Such factors include expectancy effects, placebo effects, and unmeasured confounding variables (e.g., La Marca et al., 2018; Thibault et al., 2017). Inadequate control of these non-specific factors may compromise the validity and interpretability of neurofeedback research findings (Thibault et al., 2016). Lastly, the psychological, cognitive, and electrophysiological variables that determine the ability to modulate brain activity remain insufficiently identified (Haugg et al. 2021; Jeunet et al., 2018; Kadosh & Staunton, 2019; Weber et al., 2020). Identifying these variables would optimize participant recruitment, mitigate inefficiency issues (Alkoby et al., 2018), and enable the conduct of more powerful studies without increasing sample size (Thibault & Pedder, 2022).

This study aimed to investigate the specific effects of a single neurofeedback session on brain frequencies amplitude and working memory. We compared the performance of a group that was trained to increase the amplitude of frontal theta frequency with a group trained to increase the amplitude of posterior high alpha frequency during a single neurofeedback session. Theta oscillations facilitate cognitive control and item organization in working memory (e.g., Cooper et al., 2019; Deiber et al., 2007; Hanslmayr et al., 2019; Hsieh et al., 2011; Itthipuripat et al., 2013; Pesonen et al., 2007; Riddle et al., 2020; Roux & Uhlhaas, 2014; Sauseng et al., 2010, 2009, 2005), while alpha oscillations may inhibit taskirrelevant information (e.g., de Vries et al., 2020; Hsu & Hämäläinen, 2022; Pesonen et al., 2007; Sghirripa et al., 2021). We hypothesize that neurofeedback-based amplitude modulation of these frequencies will improve performance on working memory tasks. To better distinguish between the specific and nonspecific effects of the neurofeedback, we included an active control group that was trained to modulate randomly selected frequencies. By selecting random frequency bands, participants engage in a genuine neurofeedback task that targets their own neurophysiological signals. However, because they cannot learn to modulate a specific frequency, this approach minimizes potential interference with their cognitive processes. Additionally, unlike sham control groups (Kvamme et al., 2022b), providing real EEG feedback reduces the risk of inducing a sense of learned helplessness in participants (Aliño et al., 2016). We assessed brain frequency amplitude, performance on working memory tasks (including digit span, *n*-Back, and Corsi blocks), as well as self-reported psychological states related to emotional and attentional aspects before and after the training period. We also aimed to identify factors (psychological, cognitive or electrophysiological) that could predict neuromodulation success during the neurofeedback task.

#### **Method**

#### **Sample**

G-Power (Faul et al., 2007) was utilized to determine the sample size needed for an analysis of variance with repeated measures focusing on within-between interactions. For the investigation of the impact of training on working memory, where a comparison was made between pre- and post-training performances, we computed the required sample size based on the following parameters: effect size *f* =

.20, power level = .90, number of groups = 3, number of measures = 2, correlation between repeated measures = .50. An estimated total sample size of 84 participants was obtained. 70 participants were randomly assigned to either the Theta group, aiming to increase the amplitude of the theta frequency in the frontal cortex, or to the Control group, aiming to increase randomly selected frequencies within each block. Data from some participants had to be excluded (*N* = 8, 11%) due to low-quality EEG data (i.e., more than 20% of the signal was removed after artifact rejection,  $N = 7$ ) and one participant was too fatigued to complete the entire protocol and chose to discontinue the experiment. The data from the group trained to increase parietal high alpha amplitude were obtained through a prior identical experimental protocol where the research question aimed to investigate the effects of a strategy list on neuromodulation (Chikhi et al., 2023). A total of 101 participants were included in our analyses. The Theta group consisted of 28 participants (Mean age =  $19.4 \pm 1.81$ , 75% woman), the Control group comprised 34 participants (Mean age =  $21.41 \pm 8.28$ , 91% woman) and the High alpha group consisted of 39 participants (Mean age =  $23.28$ ) ± 5.96, 82% woman). The inclusion criteria were normal or corrected-to-normal vision, no psychiatric or neurological disorders, and no usage of medication. All participants were naïve to the neurofeedback technique.

#### **Neurofeedback task and procedure**

Each participant underwent a single experimental session lasting approximately 120 minutes (Figure 1) and received course credits for their participation. Prior to and after neurofeedback session, participants completed computerized tasks assessing working memory (digit span, spatial span and *n*-back task) from the Psychology Experiment Building Language (Mueller & Piper, 2014). Additionally, participants completed a shortened version of the Neurofeedback Evaluation & Training questionnaire (NExT questionnaire; Bismuth et al., 2020; Jaumard-Hakoun et al., 2017; Pillette et al., 2021) to evaluate their subjective state during the neurofeedback protocol. Neurofeedback training began by measuring the dimensions of the participants' heads to determine electrode locations. Next, conductive gel and paste

were applied to prepare the skin and improve signal quality at each electrode location. Once the correct impedance was achieved at each electrode, a baseline was measured by recording the resting-state EEG signal during 3 minutes. Participants were instructed to maintain their gaze on a black screen positioned in front of them. They were required to remain relaxed with their eyes open, refraining from blinking, making any bodily movements, or clenching their jaw.

After recording the baseline, the average amplitude of theta and high alpha frequencies were extracted to determine the threshold for activating positive feedback. In the Control group, participants received feedback based on a threshold that automatically adapted to the average amplitude of the targeted brain signal. The frequency bands used for the participants in the Control group were randomly generated 2 Hz bands for each block and each participant. The randomly selected frequencies included a range of 2Hz, situated between 1 and 30 Hz, except for the frequency bands targeted by the experimental groups (4-8Hz and 10-12Hz). Participants were always required to increase the amplitude of the selected frequency band. Irrespective of the group, all participants were informed that the gauge shown on the left side of the screen depicted the real-time amplitude of a specific brain frequency. When the gauge exceeded the threshold (as denoted by an orange bar), a screen animation (depicting a roller coaster) was triggered, accompanied by enjoyable music. The participants' goal was to 'keep the level of the gauge above the threshold for as long and as frequently as possible'. All participants were informed that the audiovisual feedback would convey information about their brain activity and aid in achieving a mental state that triggers the reward feedback. They were not provided with particular mental strategies for regulating their brain activity. Following the completion of the 10 training blocks, the resting-state EEG signal was once again recorded. Subsequently, the electrodes were removed, the participants' scalps were cleaned, and they proceeded to complete the NExT questionnaire and computerized working memory tasks once more.

## **Figure 1**

*Experimental protocol overview*



*Note*. Preceding and subsequent to the neurofeedback session, they engaged in computerized tasks designed to evaluate their working memory (Mueller & Piper, 2014). Additionally, they responded to a concise questionnaire aimed at assessing their psychological state (Bismuth et al., 2020; Jaumard-Hakoun et al., 2017). The neurofeedback protocol encompassed measuring a 3-minute resting-state EEG before and after the 10 neurofeedback training blocks. For the Theta and High alpha groups, the positive feedback activation threshold was determined based on the mean amplitude of theta and high alpha frequencies, respectively. Both groups were provided real-time feedback, presented as a gauge positioned on the left side of the screen. Exceeding the established threshold led to the activation of animation and music. The primary objective was to sustain the gauge level above this threshold. Following the 10 training blocks and the second resting-state EEG, participants undertook post-session working memory tasks and questionnaire.

#### **EEG acquisition and processing**

An encoder (ProComp T740M) was connected to a laptop via a fiber optic cable and USB interface for recording the EEG signal. The BioGraph Infiniti software (version 6.1, Thought Technology Ltd., Montreal, QC, Canada) was used to analyze the signal and convert the targeted brain activity into audiovisual feedback. The High alpha group was trained using an electrode positioned at Pz. For the Theta group, the electrode was positioned at Fz. In the Control group, the electrode was placed at the vertex (Cz), which is equidistant from Fz and Pz (Pimenta et al., 2018). Additionally, a reference electrode and a ground electrode were positioned on the right and left earlobes, respectively. The signal was sampled at 256 Hz, filtered using a bandpass filter (0.1 to 60 Hz), and a notch filter (50 Hz) to eliminate electrical interference. The software extracted the amplitude of the signal trained for each group (theta, high alpha, or random frequency) and converted it into audio-visual feedback. The main brain frequencies were recorded for offline analysis: delta (1-4 Hz), theta (4-8 Hz), low alpha (8-10 Hz), high alpha (10-12 Hz), beta (12-35 Hz), and gamma (35-64 Hz) using fast Fourier transform (FFT). Nuprep Skin Prep Gel (Weaver and Company, Aurora, USA) was used to clean the skin prior to electrode placement. Electrodes were fixed with Ten20 Conductive Paste (Weaver and Company, Aurora, USA). Impedance was assessed using an EEG-Z sensor and kept below 10 kΩ. An automatic artifact rejection threshold was set at 100 μV before visually inspecting the raw data and manually rejecting artifacts. Additional pre-processing and artifact correction steps could not be performed due to the software used. The feedback was presented as an animation illustrating a roller coaster, accompanied by calming music (Wang & Hsieh, 2013).

For the groups trained to increase the theta or high alpha amplitude, positive feedback (animation and music) was activated when the average amplitude of the trained frequency exceeded 80% of the baseline's average value. For the Control group, a fixed threshold could not be applied because the trained frequency varied between each block. Therefore, we employed an adaptive threshold based on the amplitude of the selected frequency. This threshold corresponded to +1% of the average signal amplitude, with a delay of 0.2 seconds. This adaptive positive threshold ensures that participants receive feedback that is contingent upon their own neural activity, reducing the risk of frustration and learned helplessness (Sorger et al., 2019).

#### **Working memory tasks**

To assess different facets of participants' working memory, three distinct working memory tasks were administered via the Psychology Experiment Building Language (Mueller & Piper, 2014). The tasks included the backward digit span (Wechsler, 1955), the *n*-back task with 1, 2, and 3-back conditions (Owen et al., 2005), and the backward spatial span task, also known as Corsi's blocks task (Corsi, 1972). The backward digit span task required participants to recall a series of randomly presented digits in reverse order (Richardson, 2007). To complete this task, participants utilized the number pad of a standard keyboard. Throughout the task, the length of the digit sequence gradually increased. The dependent variable was defined as the longest successfully recalled sequence. The *n*-back task involves presenting participants with a series of stimuli (e.g., a letter), presented one after another in a sequential manner. The task required participants to identify if the current stimulus matches the one presented '*n*' position earlier in the sequence. For example, during the 2-back condition, if the letter sequence displayed on the screen was F-N-B-N-B-F, participants were instructed to press the 'yes' key when the second 'N' and the second 'B' appeared. They had to press the 'no' key for all other letters. The *n*-back task included three

conditions: 1-back, 2-back, and 3-back. The dependent variable for each condition was the percentage of correct responses. The computerized Corsi block task comprised nine fixed squares displayed on a computer screen in a pseudo-random arrangement. These squares change color individually over a short period. In the backward condition, participants were instructed to click on the squares in the reverse order of their original sequence. As the experiment advanced, the length of the block sequence progressively increased. The dependent variable was the longest accurately recalled sequence's length (Mueller & Piper, 2014). The digit span backward task was used to engage the executive control component of working memory (Lezak et al., 2004). The Corsi blocks backward task was employed to involve visuospatial working memory processing (Kessels et al., 2008). The *n*-back task involves simultaneously retaining previously presented stimuli, processing the currently presented stimulus, and updating the contents of working memory (Lezak et al., 2004; Soveri et al, 2017). By using tasks that target various working memory processes, our aim was to distinguish the cognitive effects of theta and high alpha amplitude modulation.

# **Self-reported questionnaire**

To effectively assess the psychological state of participants before, during, and after neurofeedback, while minimizing the burden of the experimental procedure and cognitive load, we utilized a condensed version of the NExT questionnaire (Bismuth et al., 2020; Jaumard-Hakoun et al., 2017; Pillette et al., 2021). The questionnaire items were adapted from established and validated questionnaires (see Supplementary material in Chikhi et al., 2022 for item sources). Each participant was asked to rate their experiences using a 5-point Likert scale. The questionnaire covered five distinct dimensions: emotional state ('calm,' 'energetic,' 'happy,' 'relaxed,' and 'satisfied'), attentional state ('receptive' and 'focused'), cognitive load, motivation, and agency ('sense of control over the feedback signal' and 'predictability of the feedback signal'). The questionnaire was administered on two occasions. Firstly, it only consisted of the first two dimensions, assessing emotional and attentional state just before the neurofeedback session. This measurement aimed to evaluate the effect of self-reported psychological states before training on neuromodulation ability. Secondly, the questionnaire was administered immediately after the neurofeedback session and included all five dimensions. This second administration aimed to assess the impact of neurofeedback on participants' psychological state changes (via the first two dimensions). The remaining three dimensions were used to compare participants' psychological state during the neurofeedback task, including cognitive load, motivation, and sense of agency. This enabled an investigation into the potential influence of these variables on the participants' success in the neurofeedback task. For example, participants in the control group might exhibit a reduced sense of agency towards the neurofeedback device, indicating a weaker perception of control over the feedback signal, in contrast to the experimental groups. Such a disparity could potentially affect their motivation and consequently lead to a decline in performance both during the neurofeedback task and in the subsequent behavioral assessments.

#### **Statistical analysis**

Statistical analyses were performed with the RStudio environment (v.4.0.2; R Core Team, 2020) with the following packages: lme4 (v.1.1-27.1; Bates et al., 2015) and lmer (v. 3.1-3; Kuznetsova et al., 2017) for classical and mixed linear regression, multcomp (v. 1.4-17; Hothorn et al., 2008) and modelbased (v. 0.8.0; Makowski et al., 2020a) for post hoc contrasts, and report (v. 0.5.1; Makowski et al., 2020b), ggplot2 (v. 3.3.6; Wickham, 2016) and smplot2 (v. 0.1.0; Min & Zhou, 2021) for formatting and visualization of results. For repeated measures, mixed-effects linear models were estimated using the Restricted Maximum Likelihood (REML) method and fitted with a random intercept (1| Participant). Classical linear regression models were estimated using Ordinary Least Squares (OLS) method. P-values for Type III ANOVA F-tests were obtained using Kenward-Roger approximation for degrees of freedom (Kenward & Roger, 1997; Luke, 2017). In cases where an effect was statistically significant, we applied Tukey's post hoc contrasts with Holm correction for multiple comparisons (Schad et al., 2020). Only significant differences from post hoc comparisons were detailed. The significance threshold for all analyses was set at α = 0.05. Effect sizes were interpreted as follows:  $\eta_p^2$  = 0.01 as a small effect,  $\eta_p^2$  = 0.06 as a moderate effect, and  $\eta_p^2$  = 0.14 as a large effect.

#### **Results**

# **Effects of neurofeedback on targeted brain signal (Figure 2)**

# *Effect of neurofeedback on theta amplitude during training*

A mixed linear model was employed to predict the amplitude of theta frequency with Group (Theta, High alpha, Control) and Training Block (1 to 10) as fixed effects, and Participant as a random effect. The ANOVA revealed a significant and strong main effect of Group (*F*(2) = 14.14, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.22, 95% *CI* [0.11 - 1.00]). Post hoc analysis indicated that the average theta frequency amplitude was significantly lower in the High alpha group compared to the Theta group ( $\beta$  = -1.37, *SE* = 0.44,  $p$  = .004). The difference between the Theta group and the Control group was not statistically significant. The main effect of Training Block was also statistically significant and weak (*F*(9) = 2.10, *p* = 0.027; *η<sup>p</sup>* <sup>2</sup>= 0.02, 95% *CI* [0.001 - 1.00]), however, post hoc comparisons revealed no significant differences in amplitude between the blocks. The interaction between Group and Training Block was not statistically significant (*p* = .053).

The absence of global interaction among the three groups and the training blocks might mask divergent effects depending on the pair of groups under consideration. Thus, we conducted two complementary analyses investigating the Group x Training Block interaction, specifically comparing (1) Theta versus High alpha group and (2) Theta versus Control group. In the first mixed linear model, we predicted the amplitude of theta frequency with Group (Theta x High alpha) and Training Block as fixed effects, and Participant as a random effect. The ANOVA revealed a significant interaction between Group and Training Block (*F*(9) = 2.43, *p* = .010; *η<sup>p</sup>* <sup>2</sup>= 0.04, 95% *CI* [0.005, 1.00]), and subsequent post hoc analysis revealed that the average theta frequency amplitude was significantly lower in the High Alpha group compared to the Theta group across blocks 3 to 10 (*SE* = 0.44, all *p* < .01). The second mixed linear model, predicting the amplitude of theta frequency with Group (Theta x Control) and Training Block as fixed effects, and Participant as a random effect, indicated that the interaction between Group and Training Block was not statistically significant (*F*(9) = 1.17, *p* = .313; *η<sup>p</sup>* <sup>2</sup>= 0.02, 95% *CI* [0.00, 1.00]).

#### *Effect of neurofeedback on theta amplitude during resting-state*

A mixed linear model was performed to predict the amplitude of theta frequency recorded during resting-state EEG, considering Group and Resting-EEG (pre- and post-training EEG recording) as fixed effects and Participant as a random effect. The ANOVA revealed a statistically significant and moderate main effect of Group (*F*(2) = 5.64, *p* = .005; *η<sup>p</sup>* <sup>2</sup>= 0.10, 95% *CI* [0.02 – 1.00]). Post hoc analysis indicated that the average theta amplitude was significantly lower in the High alpha group compared to the Control group (β = -1.67, *ES* = .60, *p* = .02), while no significant difference was observed between the Theta group and the Control group ( $p = .52$ ) nor between the Theta group and the High alpha group ( $p = .09$ ). Additionally, the main effect of Resting-EEG was statistically significant and weak (*F*(1) = 4.82, *p* = 0.03; *η<sup>p</sup>* 2  $= 0.05$ , 95% *CI* [0.002 – 1.00]), with a slightly higher mean amplitude observed after training (*M* = 10.13 ± 2.86) compared to before (*M* = 9.94 ± 2.42). However, neither the overall interaction between the Group and Resting EEG reached significance ( $p = .11$ ), nor did the pairwise comparisons (Theta x High Alpha,  $p =$ .42; Theta x Control, *p* = .24). To assess the impact of resting EEG activity on the neurofeedback task, we applied a classical linear model to compare the average theta amplitude activity during the first restingstate among the groups. The ANOVA indicated that the main effect of Group was statistically significant and medium (*F*(2, 98) = 5.11, *p* = .008; *η<sup>p</sup>* <sup>2</sup> = 0.09, 95% *CI* [0.02, 1.00]). Post-hoc comparisons revealed that the mean theta amplitude was significantly lower in the High alpha group compared to the Control group (β = 1.67, *p* < .01). No significant differences were observed between the Theta group and the Control group ( $p = .48$ ) or between the Theta group and the High alpha group ( $p = .06$ ).

#### *Effect of neurofeedback on high alpha amplitude during training*

A mixed linear model was performed to predict the amplitude of high alpha frequency with Group and Training Block as fixed effects, and Participant as a random effect. The ANOVA revealed that the main

effect of Training Block was statistically significant yet weak (*F*(9) = 5.60, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.05, 95% *CI* [0.02, 1.00]). Post hoc analysis revealed no significant differences in amplitude between the blocks. Moreover, neither the main effect of Group nor the interaction between Group and Training Block reached statistical significance ( $p = .21$  and  $p = .08$ , respectively).

Again, the lack of global interaction between the three groups and the training blocks may mask differential effects depending on the pair of groups considered. We conducted two complementary analyses testing the Group x Block interaction when specifically comparing (1) Theta vs. High alpha group and (2) Theta vs. Control group. Using a mixed linear model, we predicted the amplitude of high alpha frequency with Group (High alpha and Theta) and Training Block as fixed effects, and Participant as a random effect. The ANOVA indicated a significant interaction between Group and Training Block (*F*(9) = 2.25,  $p = 0.018$ ;  $\eta_p^2 = 0.03$ , 95% *CI* [3.02e-03, 1.00]). However, post hoc analysis did not indicate any amplitude difference across blocks. The second mixed linear model, predicting the amplitude of high alpha frequency with Group (High alpha and Control) and Training Block as fixed effects, and Participant as a random effect, indicated that the interaction between Group and Block was not statistically significant (*F*(9) = 0.60, *p* = .795; *η<sup>p</sup>* <sup>2</sup>= 0.008, 95% *CI* [0.00, 1.00]).

#### *Effect of neurofeedback on high alpha amplitude during resting-state*

A mixed linear model was performed to predict the amplitude of high alpha frequency recorded during resting-state EEG, with Group and Resting-EEG as fixed effects and Participant as a random effect. The ANOVA revealed a statistically significant and robust main effect of Resting-EEG (*F*(1) = 22.37, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.19, 95% *CI* [0.08 - 1.00]), indicating a higher average amplitude after training (*M* = 6.18 ± 2.29) compared to before (*M* = 5.83 ± 2.17). However, the main effect of Group (*p* = .39) and the interaction between Group and Resting EEG (global: *p* = .99; Theta x High Alpha, *p* = .97; Theta x Control, *p* = .89) was not statistically significant. We also assessed the difference in EEG activity during the first resting-state using a classical linear model. The ANOVA indicated that the main effect of Group was not statistically significant  $(p = .36)$ .

#### **Figure 2**

*Evolution of theta and high alpha amplitude during neurofeedback and resting-states*



*Note*. **A.** Changes in theta amplitude during neurofeedback and resting-states. The first line graph shows that, after an initial decrease, there has been a gradual increase of theta amplitude in the Theta and Control group. The second graph represents the mean amplitude and individual data points of theta frequency amplitude during the resting state, measured just before and just after the neurofeedback session. The third and final plot is an interaction plot showing the differential effects of neurofeedback training on the theta amplitude of each group. **B.** Changes in high alpha amplitude during neurofeedback and resting-states. The first line graph shows that there has been an increase of high alpha amplitude in

the three groups. The second graph shows the mean amplitude of the high alpha frequency and the individual data points for each resting-state and the last graph is an interaction plot.

# **Effects of neurofeedback on all brain frequencies**

Figure 3 illustrate the effect of neurofeedback on all frequency bands. Statistical analysis of neurofeedback effects on other frequencies amplitude during training and resting-state are presented in Supplementary material (Section 1.1).

#### **Figure 3**

*Effect of neurofeedback on EEG frequency bands.*



*Note*. Forest plot illustrating the mean slope of frequency band amplitudes across the training, error bars represent standard errors.

#### **Effect of neurofeedback on working memory performance (Figure 4)**

#### *Effect of neurofeedback on backward digit span task performance*

We conducted a mixed linear model to predict the score on a backward digit span task, with Group and Cognitive Screening (pre-and post-training working memory measurement) as fixed effects and Participant as a random effect. The ANOVA revealed that the main effect of Cognitive Screening was statistically significant and strong (*F*(1) = 29.39, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.23, 95% *CI* [0.12 – 1.00]), with a higher

mean score after training ( $M = 6.71 \pm 1.62$ ) compared to before ( $M = 5.80 \pm 1.69$ ). However, the main effect of Group and the interaction between Group and Cognitive Screening were not statistically significant ( $p = .12$  and  $p = .18$ , respectively).

#### *Effect of neurofeedback on 1-back and 2-back tasks performance*

We conducted a mixed linear model to predict the score on the 1-back task, with Group and Cognitive Screening as fixed effects and Participant as a random effect. The ANOVA revealed that the main effect of Cognitive Screening was statistically significant and moderate (*F*(1) = 6.69, *p* = .011; *η<sup>p</sup>* <sup>2</sup>= 0.06, 95% *CI* [0.008 – 1.00]), with a higher mean score after training (*M* = 0.96 ± 0.08) compared to before (*M* =  $0.93 \pm 0.10$ ). The main effect of Group and the interaction between Group and Cognitive Screening were not statistically significant (*p* = .19 and *p* = .16, respectively). We then conducted a mixed linear model to predict the score on the 2-back task, with Group and Cognitive Screening as fixed effects and Participant as a random effect. The ANOVA revealed a statistically significant and robust main effect of Cognitive Screening (*F*(1) = 25.64, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.21, 95% *CI* [0.10 – 1.00]). Post-training scores (*M* = 0.93 ± 0.08) were significantly higher compared to pre-training scores ( $M = 0.88 \pm 0.09$ ). However, the main effect of Group and the interaction between Group and Cognitive Screening were not significant (*p* = .66 and *p* = .56).

#### *Effect of neurofeedback on 3-back and Corsi block tasks performance*

Further analyses were conducted to evaluate the effects of Group, Cognitive Screening, and their interaction on 3-back scores and Corsi block performance. Our mixed models and ANOVA results indicated that the main effects of Group (*p* = .70 and *p* = .08) and Cognitive Screening (*p* = .08 and *p* = .22), as well as the interaction between these variables ( $p = .53$  and  $p = .12$ ), were not statistically significant.

#### *Effect of brain activity modulation on working memory performance*

To assess the link between brain activity and cognitive performance, we evaluated the statistical relationship between the slope of theta and high alpha amplitude during neurofeedback and the changes

in working memory task performance (measured through the difference between post-training and pretraining scores). Five linear models were performed to predict the changes in performance across working memory tasks based on the modulation of the amplitudes of two frequencies targeted during training (slope of the amplitude evolution for theta and high alpha frequencies) and the Group variable as predictor variables. The ANOVA tests showed no significant main effects nor interactions (results are presented in the Supplementary material, Section 1.2).

#### **Figure 4**



*Changes in working memory tasks performance*

*Note*. Average performance and individual data points on working memory tasks, namely digit span, spatial span and *n*-back (1, 2 & 3-back). Descriptively, there was an average increase in performance on the digit span, 1-back and 2-back tasks. As this increase was observed in all three groups, we did not observe an interaction effect. The span block task presented less unequivocal results, with an average increase for the Theta group, a decrease for the Control group and stagnation of performance for the High

alpha group. Conversely, on the 3-back task, the Control group showed the greatest improvement in performance, followed by the High alpha group and then the Theta group, whose performance stagnated. However, as presented in the text, we observed no interaction effect between groups and those two behavioral measures.

#### **Effect of neurofeedback on subjective psychological state (Figure 5)**

#### *Calm score*

We conducted a mixed-effects linear model to predict the calm score using Group and Self-Reported Measure as fixed effects and Participant as a random effect. The ANOVA indicated that the main effect of the Self-Reported Measure was statistically significant and moderate (*F*(1) = 9.22, *p* = .003; *η<sup>p</sup>* <sup>2</sup>= 0.09, 95% *CI* [0.02 – 1.00]), with a higher average score after training (*M* = 3.19 ± 0.97) compared to before (*M* = 2.78 ± 1.05). The main effect of the Group and the interaction between Group and Self-Reported Measure were not significant ( $p = .92$  and  $p = .52$ , respectively).

#### *Energetic score*

We performed a mixed-effects linear model to predict the energy score using Group and Self-Reported Measure as fixed effects and Participant as a random effect. The ANOVA revealed that the main effect of the Self-Reported Measure was statistically significant and strong ( $F(1)$  = 41.51,  $p < .001$ ;  $\eta_p^2$  = 0.30, 95% *CI* [0.18 – 1.00]), with a lower average score after training (*M* = 1.16 ± 0.97) compared to before (*M* = 1.88 ± 0.98). The main effect of the Group and the interaction between Group and Self-Reported Measure were not significant ( $p = .19$  and  $p = .38$ ).

#### *Relaxed score*

We fitted a mixed-effects linear model to predict the relaxation score using Group and Self-Reported Measure as fixed effects and Participant as a random effect. The ANOVA demonstrated that the main effect of the Self-Reported Measure was statistically significant and strong (*F*(1) = 38.52, *p* < .001; *ηp* <sup>2</sup>= 0.28, 95% *CI* [0.16 – 1.00]), with a higher average score after training (*M* = 3.08 ± 0.94) compared to before (*M* = 2.25 ± 1.28). The main effect of the Group and the interaction between Group and Self-Reported Measure were not significant (*p* = .69 and *p* = .78).

#### *Satisfaction score*

We conducted a mixed linear model to predict the satisfaction score, with Group and Self-Reported Measure as fixed effects and Participant as a random effect. The ANOVA revealed a statistically significant and moderate main effect of the Self-Reported Measure ( $F(1)$  = 7.58,  $p$  = .007;  $\eta_p^2$  = 0.07, 95% *CI* [0.01 – 1.00]), indicating a higher average score after the training (*M* = 2.69 ± 0.98) compared to before (*M* = 2.42 ± 1.19). However, the interaction effect between Group and Self-Reported Measure was statistically significant and moderate (*F*(2) = 4.70, *p* = .011; *η<sup>p</sup>* <sup>2</sup>= 0.09, 95% *CI* [0.01, 1.00]). Post hoc analysis revealed that for the Control group, the average score was significantly higher after the training compared to before (β = 0.59, *SE* = 0.19, *p* = .03). The main effect of Group was not significant (*p* = .053).

#### *Happiness, Receptivity, and Concentration scores*

We conducted three mixed linear models to predict the happiness score, receptivity score, and concentration score, with Group and Self-Reported Measure as fixed effects and Participant as a random effect. The ANOVA indicated that the main effects of Group, Cognitive Screening, and the interaction between these two variables were not significant for these three items (results are presented in the Supplementary material, Section 1.3).

#### *Cognitive Load score*

We performed a linear model to predict the subjective cognitive load score during neurofeedback training, with Group as the predictor variable. The ANOVA revealed a statistically significant and strong main effect of Group (*F*(2, 98) = 38.19, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.44, 95% *CI* [0.32 – 1.00]). Post hoc analysis demonstrated that the mean cognitive load score was significantly lower in the High Alpha group compared to the Theta group (β = -1.92, *t*(98) = 7.78, *SE* = 0.25, *p* < .001) and the Control group (β = -1.63, *t*(98) = 7.01, *SE* = 0.23, *p* < .001).

#### *Flow, Mastery, and Coherence scores*

We employed three linear models to predict the subjective feeling of flow, mastery and coherence of the feedback signal during the neurofeedback training, with Group as the predictive variable. The ANOVAs revealed that there were no significant main effects of Group on any of the scores (*p* = .37, *p* = .86 and  $p = 0.39$ , respectively).

#### **Figure 5**



*Statistically significant changes in self-reported psychological state*

*Note*. Average score and individual data point of questionnaire scores measured before and after neurofeedback. In a similar way for the three groups, there was an increase in the calm and relaxation scores and a decrease in the energy score. In terms of satisfaction, the Control group showed the strongest increase, as evidenced by the significant interaction effect (see Results section), followed by the Theta group. The High alpha group showed a slight decrease. \*\*\* *p* < .001.

# **Predictors of neuromodulation**

To better understand the factors involved in neuromodulation, we utilized resting-state EEG activity, pre-training scores on working memory tasks and questionnaires to predict the neuromodulation of targeted signals through neurofeedback. A mixed linear model was employed to predict the slope of theta amplitude during neurofeedback using Group and resting-state brain frequencies (delta, theta, low alpha, high alpha, beta, and gamma) as predictor variables. ANOVA results revealed a statistically significant and strong main effect of resting-state theta amplitude (*F*(1, 92) = 14.06, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.13, 95% *CI* [0.04 - 1.00]). The other main effects were not significant (see Supplementary material, Section 1.4.a). A mixed linear model was utilized to predict the slope of high alpha amplitude during neurofeedback using Group and resting-state brain frequencies (delta, theta, low alpha, high alpha, beta, and gamma) as predictor variables. ANOVA indicated a statistically significant and moderate main effect of resting-state low alpha amplitude (*F*(1, 92) = 7.80, *p* = .006; *η<sup>p</sup>* <sup>2</sup>= 0.08, 95% *CI* [0.01 - 1.00]) and a strong main effect of resting-state high alpha amplitude (*F*(1, 92) = 21.97, *p* < .001; *η<sup>p</sup>* <sup>2</sup>= 0.19, 95% *CI* [0.09 - 1.00]). The other main effects were not significant (see Supplementary material, Section 1.4.b). The performance scores on working memory tasks and the questionnaire measured before neurofeedback did not predict the changes in the amplitude of the signals targeted by the training (results are presented in the Supplementary material, Section 1.4).

#### **Figure 6**



*Scatter plots of the two predictors of neuromodulation*

*Note*. We predicted the evolution of the amplitude (i.e., slope) of the signals targeted by neurofeedback (i.e., theta and high alpha) via psychological (self-reported questionnaire), cognitive (working memory performance) and electrophysiological (resting-state EEG) variables measured before neurofeedback. Only resting-state EEG of the trained signal predicted the slope of those signals (**A**: theta slope, **B**: high alpha slope). The two scatter plots show the slope of the trained signal as a function of the mean amplitude recorded in the resting-state. The points represent individual data, the line represents the fitted linear model and the shaded area the 95% confidence interval of the fitted values.

#### **Discussion**

In this study, we investigated the effect of a single neurofeedback session on brain oscillations amplitude, working memory task performance, and self-reported psychological states. We hypothesized that the neurofeedback's effects on brain activity would vary depending on whether the group was trained to enhance the amplitude of the theta frequency, the high alpha frequency, or random frequencies (active control group). This modulation of the targeted brain signal was expected to result in improved

performance in working memory tasks. To gain a deeper understanding of the self-regulation mechanism, we utilized resting-state EEG activity, working memory task performance, and self-reported psychological states to predict the neuromodulation of the trained signal. While our analyses did reveal electrophysiological and behavioral improvements, these gains were not exclusive to the trained signals. An increase in the frequency targeted by the training has been observed for the theta and high alpha groups, but no significant difference has been observed between these groups and the active control group. Among all the predictors of neuromodulation that have been examined, theta and high alpha amplitudes at rest have been shown to predict the ability to modulate the frequencies targeted by neurofeedback. Participants who showed the greatest increase in amplitude for the trained frequency during neurofeedback were those with the highest amplitudes during resting EEG measurements. Overall, these findings contribute to a better understanding of the factors influencing the outcomes of a neurofeedback training and highlight the challenges associated with using random frequencies training as an active control group.

# **Effect of neurofeedback on the amplitude of trained brain oscillations**

The main goal of this study was to investigate the effects of a neurofeedback session designed to enhance the amplitude of theta and high alpha frequencies. The first hypothesis was that engaging in neurofeedback to increase the amplitudes of theta and high alpha frequencies would indeed affect the amplitudes of the targeted frequencies.

#### *Theta frequency*

When comparing the three groups together, we observed a main effect of neurofeedback training on the average amplitude of theta frequency but no interaction effect. Nonetheless, the absence of a global interaction effect may mask specific differences among the groups. Indeed, we observed a significant interaction between the Theta and High Alpha groups, indicating an increase in theta amplitude in the former compared to the latter. This interaction effect is consistent with previous research that indicates an increase in theta amplitude during a single training session (Eschmann et al., 2021; Rozengurt et al., 2016; Rozengurt et al., 2017; Shtoots et al., 2021), and suggests that the theta training had specific effects compared to a group trained to modulate another frequency. However, we did not observe significant differences between the Theta group and our active Control group. In the previously cited studies, the active control group underwent training to enhance the amplitude of the beta frequency, resulting in slight decreases (Rozengurt et al., 2017), no changed (Rozengurt et al., 2016), or slight increases in theta amplitude (8%, Shtoots et al., 2021). Another study utilized neurofeedback to increase beta1 frequency (15-18 Hz) amplitude and showed a slight decrease in theta amplitude during the training (Pimenta et al., 2018). The lack of difference could be attributed to our control group, which used random frequencies instead of the beta frequency. According to our findings, training to increase multiple frequencies appears to induce a more generalized neuromodulation than training targeting a specific frequency. Thus, a more appropriate control group would be one trained either to decrease randomly selected frequencies or to increase a different and uncorrelated frequency (e.g., beta). Concerning the effect of training on resting brain activity, our findings revealed an increase in the amplitude of restingstate theta amplitude after the training. However, no significant differences were observed between the groups concerning changes in theta amplitude, whether through assessing the overall interaction among the three groups or through additional pairwise comparisons. This lack of interaction effect cannot be directly compared to the findings of earlier studies (Eschmann et al., 2021; Rozengurt et al., 2016; Rozengurt et al., 2017; Shtoots et al., 2021), as these previous studies solely assessed theta frequency amplitude at rest before the neurofeedback, without considering post-training measurements.

#### *High alpha frequency*

Consistent with previous studies (Escolano et al., 2012; Escolano et al., 2014), we observed that a single high alpha neurofeedback training session leads to an increase in the amplitude of the high alpha frequency both during training (compared to the theta group) and in subsequent resting states. However, we did not observe an interaction effect when comparing the high alpha group with the active control group, as the latter also exhibited an increase in high alpha frequency. Again, this suggests that training to increase different frequencies across neurofeedback training may result in nonspecific, broad effects on frequency amplitude.

#### *Other frequencies*

The analysis of the neurofeedback effect on the amplitude of other brain frequencies has shown an increase in delta frequency amplitude within the Theta and Control group, a reduction in beta frequency amplitude among the High alpha group, and a decrease in gamma frequency amplitude across all groups. Thus, our findings corroborate the absence of selectivity resulting from a single neurofeedback session on EEG activity (Dessy et al., 2020; Jurewicz et al., 2018; Pimenta et al., 2018). This outcome was expected within the Control group, which underwent training to regulate various brain frequencies, but not within the Theta and High alpha groups. A unique neurofeedback session does not appear to enable the modulation of brain activity exclusively within the frequency targeted by the neurofeedback device.

#### **Effect of training on working memory performance**

The second hypothesis of this study was that enhancing the amplitude of theta or high alpha frequencies through neurofeedback would improve performance on working memory tasks. While significant improvements were observed in performance on the digit span task and in two conditions of the *n*-back task (1-back and 2-back), these gains were observed across all groups. These improvements in performance across the three groups may be attributed to the neuromodulation observed in each of the groups. To test this hypothesis, we predicted the difference in working memory performance (posttraining minus pre-training) using the slope of the evolution of theta and high alpha amplitude during training. Similar to the study conducted by Eschmann and colleagues (2021), which evaluated the link between the modulation of theta amplitude and changes in performance on a 2-back task, no statistically significant relationship was observed. The absence of statistically significant relationships in both studies

underscores the possibility that improvements in behavioral tasks may not directly result from the targeted neurofeedback training, but rather from non-specific factors such as increased task familiarity or increased motivation. It must, however, be emphasized that our ability to make psychophysiological inferences is limited here given that we did not record brain activity during the behavioral tasks. Further research examining cerebral activity during working memory tasks may yield more robust and comprehensive explanations.

#### **Effect of training on self-reported questionnaire**

The analysis of the questionnaires assessing participants' emotional state and consciousness revealed significant changes. A substantial decrease in the sense of energy was observed, alongside an increase in feelings of calmness and relaxation, regardless of the experimental group. The absence of differences between the groups suggests that these effects were not limited to the trained frequency and may indicate an increase in participants' levels of fatigue. Despite having short breaks between each block, the 30-minute neurofeedback training demanded continuous vigilance in focusing on the feedback signal. This increase in fatigue could potentially explain the observed modulation of various brain frequencies namely, delta, theta, alpha, and beta. A recent meta-analysis emphasized the connection between fatigue and changes in amplitude within these frequencies (Tran et al., 2020). Interestingly, self-reported questionnaire responses indicated that both the Theta and Control groups experienced higher cognitive loads during training than the High alpha group. While this may support the established link between theta amplitude and cognitive load (Chikhi et al., 2022), we found no statistically significant correlation between theta activity and perceived cognitive load. Nonetheless, this result, along with similar motivation and agency scores across groups, suggests that participants of the Control group were equally engaged in the task as participants in the Theta and High alpha groups.

## **Towards specificity of EEG neurofeedback**

Here, a single neurofeedback session aimed at increasing one frequency amplitude (theta or high alpha), does not elicit specific modulation of brain activity when compared to a group trained to increase random frequencies. First, this absence of significant differences may be attributed to the brief duration of our training, which was limited to a single session. Although there is no consensus on the time required for learning to occur, theoretical models suggest that the repeated production of the neural state rewarded by feedback induces neuroplasticity, facilitating the re-establishment of the neural state targeted by the training (Birbaumer et al., 2013; Davelaar, 2018; Lubianiker et al., 2022; Niv, 2013; Ros, 2014; Shibata et al., 2019). Thus, a prolonged training duration could strengthen the neural connections associated with the specific targeted frequencies, leading to more durable and robust neural changes compared to a group trained to modulate random signals. Consequently, by prioritizing sample size over the number of training sessions, our findings do not preclude the possibility that mastering the self-regulation of theta or high alpha frequencies over an extended period could produce distinct effects compared to a group trained to increase multiple frequency bands.

However, brain frequencies may be inter-correlated (Canolty & Knight, 2010; Klimesch, 2018), to the extent that EEG-neurofeedback training leads to non-specific modulation in the amplitude of brain frequencies. In this respect, the use of conventional EEG frequency bands could be sub-optimal when targeting a specific parameter of brain activity to positively impact a specific cognitive component. Therefore, it may be crucial to find functionally relevant biomarkers that can be trained independently. Hence, recent studies have used innovative methods for analyzing EEG signals in neurofeedback protocols. For example, studies have used multivariate pattern analysis (Haynes & Rees, 2006; Taschereau-Dumouchel et al., 2021) to "decode" cognitive states using machine learning techniques, thereby targeting specific brain networks involved in the cognitive state to be modified (Bu et al., 2019; Faller et al., 2019; Keynan et al., 2019; Tuckute et al., 2021; Rana et al., 2020; Wang et al., 2021). Other

studies have applied micro-state analysis, which identifies short, stable, distinct patterns of electrical activity measured by EEG across the scalp (Férat et al., 2022; Khanna et al., 2015; Michel & Koenig, 2018), representing various cognitive processes (Tarailis et al., 2024). Currently, few studies have attempted to modulate these neural states via neurofeedback (Asai et al., 2022; Diaz Hernandez et al., 2016), and it is necessary to assess larger samples in well-controlled protocols to highlight the potential of these methods compared to the use of conventional brain frequencies.

#### **Predictive variables of neuromodulation of trained signals**

In this research, our second objective was to identify factors that could potentially predict the modulation of brain signals through the use of a neurofeedback. We found that neither working memory performance nor pre-training questionnaire scores could predict the neuromodulation of the trained signals. Instead, the sole predictor for both trained frequencies (theta and high alpha) was the average amplitude of the targeted frequency recorded during rest before the training. This effect, already observed for alpha frequency (Su et al., 2021; Nan et al., 2018; Wan et al., 2014), was replicated here for theta frequency. This effect is particularly intriguing due to the specificity of the result: neuromodulation during training is not predicted by other brain signals or variables recorded in our study. The sole exception to this specificity pertains to the high alpha frequency, where modulation is also predicted by the amplitude of the resting low alpha. This result may suggest that the two sub-bands of the alpha frequency are not totally independent, at least within the temporal scope of our study. This effect could also contribute to an explanation for the observed increase in theta amplitude in the Control group. Indeed, the Control group exhibited higher resting theta activity compared to the High Alpha group and demonstrated similar activity to the Theta group, reflecting the neuromodulation pattern observed during neurofeedback. Our study appears to validate the existing relationship between neuromodulation capacity and resting-state signal amplitude. Thus, it may be relevant to consider using this measure as an inclusion criterion to select a sample with a higher proportion of learners. This approach would enhance

the statistical power of studies without increasing the sample size (Thibault & Pedder, 2022). What remains to be elucidated, however, is the mechanism behind this statistical relationship. In cognitive training, a lower baseline level is usually associated with greater improvement in the trained variable (Roheger et al., 2020; Traut et al., 2021). In the case of neuromodulation, our findings suggest an inverse relationship: higher amplitude of the trained frequency seems to indicate a higher likelihood of neuronal synchronization in the targeted brain region. One hypothesis could be that the oscillatory activity level (i.e., amplitude) recorded at rest reflects the level of neuronal excitability in the targeted frequency (Ogata et al., 2019; Schutter & Hortensius, 2011). To further explore this hypothesis, experimental testing could stimulate brain activity immediately prior to neurofeedback session (Orendáčová & Kvašňák, 2021).

#### **Conclusion**

Our neurofeedback training led to electrophysiological and behavioral improvements in all groups, including the active control group. Although these results may suggest a lack of specific neurofeedback effect, identifying the non-specific factors contributing to neuromodulation remains complex due to the sensory, cognitive, and affective factors involved in the self-regulation task (Lubianiker et al., 2019). A factor that appears robustly associated with brain activity neuromodulation is the resting-state activity recorded before training. Still, longer-term protocols with sufficient statistical power are still necessary to draw solid conclusions about the mechanisms involved in EEG activity modulation by neurofeedback.

#### **Declarations**

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#### **Conflicts of interest/Competing interest**

The authors have no relevant financial or non-financial interests to disclose.

#### **Ethics approval**

This study was performed in line with the principles of the Declaration of Helsinki. Approval was

granted by the Ethics Committee of the University Paris Cité (IRB No. = 00012019-41).

#### **Consent to participate/Consent for publication**

Informed consent was obtained from all individual participants included in the study.

#### **Availability of data and code (Open Practices Statement)**

The data and R script are available on this OSF repository:

#### [https://osf.io/zs97e/?view\\_only=7d01849a74454cdfa42a09a92938cf64](https://osf.io/zs97e/?view_only=7d01849a74454cdfa42a09a92938cf64)

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