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Investigating the effect of Integral Meditation: the impact of an original mindfulness-based intervention on psychological and neuropsychological outcomes.

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ABSTRACT

INTRODUCTION: Research on mindfulness-based interventions (MBIs) has constantly reported beneficial effects on psychological well-being associated with mindfulness meditation training. On the other hand, neurophysiological changes associated with MBIs are less clear. The Integral Meditation (IM) is an original MBI developed by our research team and previously associated with positive effects on several psychological domains.

AIM: This study aimed to assess the effects of the IM training, on young adults. The assessment included psychological factors, such as dispositional mindfulness, interoceptive sensibility, interoceptive accuracy, state anxiety and trait anxiety, and neurophysiological measures derived from the electroencephalography (EEG) alpha frequency band activity. The associations between the changes in those two domains were also explored.

METHODS: Participants (N=89) were recruited in the city of Pavia, Italy and randomized in intervention and control groups. Participants completed four self-report measures, a laboratory task (Heartbeat Tracking Task) and underwent a conventional eye-closed resting-state EEG recording at the two timepoints. The intervention group participated to the IM intervention while the control group did not engage in any intervention. Statistical analyses were performed to determine the effectiveness of the intervention.

RESULTS: Significant changes were found in the intervention group for dispositional mindfulness and interoceptive sensibility, and their subdomains. Analysis of EEG alpha activity revealed that the effect of the intervention on the Individual Alpha Peak power changes to participants' individual characteristics. Exploratory analysis showed that the effect of the intervention on the alpha activity varies also among different brain regions, highlighting the role of the individual differences. Correlation analysis indicates several bidirectional associations between the changes in the psychological measures and the changes in EEG alpha activity.

CONCLUSIONS: This study on IM among healthy young adults revealed significant improvements in dispositional mindfulness and interoceptive sensibility, despite no discernible impact on anxiety. The findings emphasize the need for personalized mindfulness interventions, recognizing individual variations in response for optimal effectiveness. The study is affected by several limitations such as the reduced sample size, due to the high dropout. Future studies on IM should explore the effects in alpha sub-bands as well as in other EEG frequency bands.

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1. INTRODUCTION

1.1 Roots of Meditation

A timeless tradition that originated in ancient India continues to endure to our days, creating a bridge between ancient wisdom and contemporary investigation: this is meditation, a simultaneously simple and complex practice that has withstood the test of time and is now a subject of interest in the world of science.

There are various forms of meditation, primarily rooted in ancient religious and spiritual traditions, each involving different techniques. Individuals practicing meditation usually adopt specific methods, such as maintaining a particular posture, sustaining focused attention, and fostering an open mindset towards distractions [1]. Among these techniques, mindfulness stands out as one of the most popular. It consists of two main components: attention and acceptance. The attention aspect involves tuning into your experiences, focusing on the present moment by directing your awareness to your breath, thoughts, physical sensations, and emotions. Acceptance involves acknowledging and observing your feelings and sensations without judgment. [2].

The term "meditate" shares its roots with "medicine," both stemming from the Latin verb "medeor", meaning "to heal." This connection goes beyond etymology. The growing interest in meditation is grounded by observed and measured positive effects on health and well-being. Meditation yields physical, emotional, and cognitive benefits, making it applicable as a therapeutic approach for mental health disorders [3], as well as conditions like addiction [4] and chronic pain [5]. Moreover, it serves as a tool to enhance well-being, reduce stress, and improve emotional functioning in healthy populations [6], [7], [8]. Recent studies indicate that meditation induces neuroplastic changes in brain regions associated with attention, emotion regulation, and self-awareness and these findings have been observed in both long-term practitioners and in individuals undergoing short-term training programs [9].

Being an accessible, cost-effective, and efficient practice to learn, meditation training finds applications in various settings. As noted by Creswell [10], it is now being integrated into institutional environments such as schools, military, prisons, and workplaces. The question arises: why does meditation prove to exert beneficial effects across diverse populations and in such a wide array of contexts? To comprehend this, it is essential to highlight that the essence of meditation, which involves being aware of the present moment, contrasts with many aspects of our daily experiences [10]. In our day-to-day lives, we often unintentionally let our minds wander [11], operate on autopilot [12] or suppress unwanted experiences [13]. Moreover, the mindless state that frequently dominates our minds has been demonstrated to be undesirable. A study revealed that mind wandering occurs in almost half of people engaged in other activities, with at least 30% experiencing it during every activity, predicting subsequent unhappiness [11]. Conversely, the capacity to be mindful is linked to greater well-being in daily life [14].

1.2 Effects of meditation on psychological factors

Mindfulness meditation has been reported to produce diverse effects on behaviours related to psychological factors, contributing to increased well-being in various ways.

Dispositional mindfulness

One of the main aspects of mindfulness meditation is the cultivation of mindfulness. Using the words of Kabat-Zinn (2023), mindfulness is "paying attention in a particular way: on purpose, in the present moment, and nonjudgmentally". Mindfulness is conceptualized both as a momentary state, representing a transient condition, and as a trait, denoting a stable characteristic present in the general population even without formal mindfulness training [16]. The positive outcomes of mindfulness interventions on state mindfulness have prompted investigations into mindfulness as an inherent human trait [17]. Changes in trait mindfulness, also known as dispositional mindfulness, are linked to mindfulness training [18]. Various scales, such as the Five Facet Mindfulness Questionnaire (FFMQ) [19], have been developed to explore changes in different sub-dimensions of dispositional mindfulness.

The link between dispositional mindfulness and psychological well-being has been assessed in numerous studies. For example, a study reported beneficial effect of mindfulness meditation on dispositional mindfulness and well-being, together with a decrease in stress [20]. In another study, dispositional mindfulness was strongly associated with well-being and perceived health, and it was suggested being a protective factor against the negative influence of perceived stress on psychological well-being [21]. Those findings align with a comprehensive review that examined empirical literature across multiple methodologies, concluding that mindfulness and its cultivation foster adaptive psychological functioning. The convergence of findings from correlational studies, clinical interventions, and laboratory-based experiments all suggests a positive association between mindfulness and psychological health, substantiating the idea that training in mindfulness can bring positive psychological effects [22].

Interoception

Another construct that more recently is matter of interest in meditation research it is interoception.

Interoception can be defined as the perception of internal body states, including sensations from the gastrointestinal, respiratory, and cardiovascular systems. This process is crucial for the nervous system to sense, interpret, and integrate signals within the body, providing a real-time map of the internal landscape at conscious and unconscious levels. Interoceptive signalling is integral to reflexes, urges, feelings, drives, and adaptive responses, contributing to homeostatic functioning, body regulation, and survival. Dysfunctions in interoception are increasingly recognized as significant components of various mental health conditions, including anxiety [23].

Given the role that interoception has in mental health and the relevance this construct is gaining in neuropsychology, a three-dimension model of interoception has been proposed [24] to account for individual differences. In this model the dimension of interoception have been classified as: (1) *interoceptive accuracy* (objective accuracy in detecting internal bodily sensations), (2) *interoceptive*

sensibility (self-perceived dispositional tendency to be internally self-focused and interoceptively cognisant) and (3) *interoceptive awareness* (metacognitive awareness of interoceptive accuracy) [24].

In the literature of mindfulness meditation, interoception is frequently evaluated through self-report measures such as the Multidimensional Assessment of Interoceptive Awareness (MAIA) [25]. This assessment tool captures different facets of interoceptive awareness, including the capacity to observe, describe, and act with mindfulness towards bodily sensations. Another measure commonly used to measure interoception is the Heartbeat Tracking Task [26]. This task, also known as the heartbeat detection task, is a used to measure interoceptive accuracy.

There is evidence that meditation and Mindfulness-based Interventions (MBIs) improve interoceptive sensibility among different populations [27], [28], [29], [30]. Nevertheless, a consistent correlation between mindfulness and other facets of interoception, such as interoceptive accuracy, does not seem evident [31], [32]. A study that assessed the effect of a MBI on young healthy adults, found an increase in interoceptive sensibility, but not in interoceptive accuracy [33], confirming that these dimensions of interoception are indeed separated. Despite some studies indicate effects of contemplative training [34] and body scan intervention [35] on interoceptive accuracy, other studies do not support such hypotheses [36], [37], [38], [39].

Evidence of the beneficial effect of MBIs on interoception also comes from neuroimaging studies. A study among healthy adults found changes in the connectivity of the right insula, a brain area associated with interoception, and these structural changes were potentially associated with functional changes [40]; a systematic review of functional magnetic resonance imaging studies pointed out an increased insula reactivity after completion of a mindfulness intervention as the most consistent finding [41].

Dispositional mindfulness and interoception are closely related constructs, partially overlapped depending on their definitions, and both independently associated with enhanced psychological wellbeing [42]. Some authors claim that interoception constitutes the primary mechanism responsible for the benefits of the mindfulness training [25], [43], [44].

Anxiety

Another relevant construct that can be modulated by mindfulness training is anxiety and this relationship has been observed in several studies. According to the World Health Organization [45], an estimated 4% of the global population currently experience an anxiety disorder, with more women affected than men. The prevalence of anxiety disorders is associated with a considerable degree of impairment, high health-care utilization, and an enormous economic burden for society [46]. In this framework, it is important to promote and deliver treatments with demonstrated efficacy in alleviating and preventing anxiety symptoms, with mindfulness meditation standing out as one of those intervention.

A study among participants diagnosed with Generalized Anxiety Disorder found a beneficial effect of a mindfulness meditation compared to a stress management education course on anxiety symptoms [47]. In a meta-analytic review of 39 studies among samples with elevated baseline scores of anxiety and depression, mindfulness-based therapy exhibited a moderate level of effectiveness in alleviating anxiety and mood symptoms from pre- to post-treatment. It was notably more effective in ameliorating anxiety and mood symptoms among individuals diagnosed with anxiety and mood disorders [48]. In a study among voluntary participants, a beneficial effect of meditation on anxiety was reported for the treated but not the control group. These findings suggest that mindfulness meditation may have a beneficial effect on anxiety among non-clinical samples, and that it may be a promising intervention for reducing anxiety symptoms in the general population. In this context, a wide used self-report measured of anxiety is the State-Trait Anxiety Inventory [49].

As to depression, a study explored the relationship between mindfulness and symptoms of depression and anxiety in the general population, investigating emotional regulation mechanisms as potential mediators, and found that mindfulness was associated with decreased depression and anxiety by increasing reappraisal and reducing worry, rumination, and suppression. Worry and rumination were identified as potent mediators, suggesting that individuals with high dispositional mindfulness can efficiently regulate emotions and handle intrusive thoughts. The results support the idea that mindfulness, whether dispositional or enhanced through meditation, can enhance well-being by reducing depression and anxiety symptoms [50]. In a randomized controlled trial, it was reported that individuals with higher dispositional mindfulness had lower levels of stress, state anxiety and trait anxiety state and it was found a reduction in those symptoms after a MBI [51]. Another study also supported the inverse association between dispositional mindfulness and anxiety. [21]. A metaanalysis used longitudinal data to test if the dispositional mindfulness scores predict a subsequent reduction in anxiety and identified that the dispositional mindfulness facet of "Acting with Awareness" (see 2.2.1 Self-report measures, FFMQ) consistently predicted a reduction in anxiety symptoms over time. In contrast, other facets of dispositional mindfulness exhibited varied associations [52].

Divergent outcomes in studies on interoceptive sensibility and anxiety are tied to the conceptualization of interoceptive sensibility. The Multidimensional Assessment of Interoceptive Awareness (MAIA), reflecting a valence-neutral or positive interoceptive style [24], [53], [54], contrasts with measures like the Body Perception Questionnaire (BPQ) [55], which primarily captures a maladaptive awareness of aversive bodily sensations [56]. Studies utilizing BPQ reveal a positive correlation between anxiety and interoceptive sensibility [57], [58], while those employing MAIA, or its subsections often show negative or no associations with anxiety measures [59]. This underscores the importance of distinguishing between maladaptive and adaptive aspects of interoceptive sensibility [60].

1.3 Contemplative neuroscience

In 1993, during a meeting, the Dalai Lama asked the neuroscientist Richard Davidson why modern neuroscience has focused mostly on studying negative psychological factors, such anxiety and depression, while neglecting qualities related to meditation, like kindness and compassion. This encounter marked one of the first interactions between two worlds that, at the time, appeared separate and even contradictory. A decade later the ground was favourable for a new beginning while another notable event took place. The Mind and Life Institute's public dialogue, titled 'Investigating the Mind,' took place at the Massachusetts Institute of Technology (MIT) in 2003. The participants included the 14th Dalai Lama, Nobel Laureate scientist Daniel Kahneman, and Eric Lander, Director

of the MIT Centre for Genomic Research. Attended by 1,200 scientists and contemplatives, this conference marked the public birth of contemplative neuroscience in the USA.

Since then, a significant transformation has occurred, leading to the establishment of a new field known as contemplative neuroscience. This evolution was driven by notable findings using neuroscientific tools to explore the impacts and mechanisms of meditation and other contemplative functional practices. Techniques like magnetic resonance imaging (fMRI) and Electroencephalography (EEG) offer a distinctive opportunity to investigate meditation's effects and mechanisms, addressing challenges related to the inherent subjectivity of the meditation experience. While contemplative neuroscience has contributed substantially to the scientific debate on meditation, many questions remain unanswered, or partially, and both theoretical and methodological challenges continue to affect the precision of these results.

When approaching the field of contemplative neuroscience, scientists grapple with an initial challenge: the absence of unified definitions and theories. This complexity arises from the vast diversity of meditation techniques and practices, which some are categorized as mindfulness meditation. Moreover, the multifaceted impact of meditation training across various levels and domains of human functioning adds further complexity. Consequently, interpreting the effects of meditation becomes intricate, with often mixed results characterizing the findings.

In recent decades, among the different types of meditation, the mindfulness meditation has been the most extensively explored both in scientific research and clinical applications within health sciences, including psychology and medicine. Described as "nonjudgmental attention to experiences in the present moment" [61], mindfulness meditation involves key components like body relaxation, breathing exercises, mental imagery, and heightened awareness of the mind-body connection. It encompasses both focused attention and open-monitoring processes, where focused attention involves sustained concentration on a chosen object, breath, or body part, and open monitoring entails non-reactive observation of moment-to-moment experiences [62], [63]. Other popular meditation techniques include mantra meditation, which centers on repetitive mantras to calm, and focus the mind effortlessly) [64], and loving-kindness-compassion meditation, designed to enhance well-being and prevent negative emotions like anger or irritation [64], [65]. A notable theory that clarifies the mechanisms of mindfulness is the S-ART model, proposed by Vago and David [62], which takes in account the variability and describes mindfulness in a broader framework of perceptual, physiological, cognitive, emotional, and behavioural component processes. In the S-ART model, the practice of mindfulness leads to an increase in three cognitive dimensions which are meta selfawareness, self-regulation, and self-transcendence (Figure 1); the authors also identified six mechanisms that underlie the mindfulness practice: intention and motivation, attention regulation, emotion regulation, memory extinction and reconsolidation, prosociality, non-attachment and decentering. Finally, the S-ART model is justified and explained with respect to the neural networks of the self-processing that have been found involved in mindfulness.

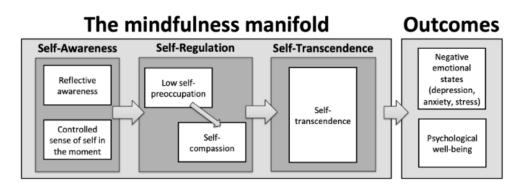


Figure 1. The S-ART Mindfulness Manifold. Obtained from Verhaeghen [66].

Seeking for a more profound comprehension of the mechanisms triggered by mindfulness in the brain we came to the exploration of mindfulness meditation effects through neuroimaging techniques, particularly Electroencephalography (EEG). EEG is a non-invasive neuroimaging technique that records electrical activity in the brain over time. It measures the synchronized electrical potentials generated by large populations of neurons, providing insights into the spatiotemporal dynamics of neural networks [67]. By placing electrodes on the scalp, EEG captures the summed electrical activity, offering valuable information about brain function and connectivity. Distinct EEG frequency bands, including delta, theta, alpha, beta, and gamma, correspond to different mental states (**Figure 2**). EEG is widely employed in studies investigating the neuropsychological effects of mindfulness and meditation and the alpha wave, a fundamental oscillation in the human brain, exhibits the most consistent alterations in EEG meditation investigations, offering reliable outcomes [68].

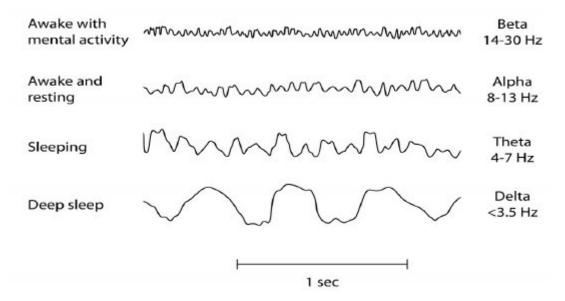


Figure 2: Different Human EEG Waves and Their Associated State of Consciousness. Obtained from Abhang [69].

The studies that have investigated the neurophysiological characteristics of meditation through EEG fall into three main categories: a) cross-sectional comparisons between expert meditators and novices/non-meditators, b) exploration of EEG changes during meditation sessions, and c) pretest-posttest designs assessing the impact of meditation practice on EEG patterns.

Regarding the first two categories, individuals with long meditation practice display increased theta and alpha power and a slower EEG activity compared to non-meditators [70]. Slowing in the alpha waves and an increased alpha power are the most reported changes during meditation compared to a resting state, for both experienced and novice meditators and across different meditation techniques, while another frequently reported result is an increase in theta power [68], [71]. Additionally, an increase in brainwave coherence, the synchronization between the electrical activities of different areas of the brain, is found in the alpha-theta span, both inter- and intra-hemispherical during meditation. Similar patterns are observed in meditators during rest and cognitive tasks. It should be noted that the results reported on oscillatory activity related to meditation techniques; indeed, different meditation techniques are related with activation of different regions of the brain and at different frequencies [72], [73], [74]. Alpha waves are generally associated with more relaxed and alert states of mind and alpha activity is inversely related to anxiety and similar association has been found for theta activity, while the changes in theta reported during meditation are less ambiguous compared to the results found in alpha waves [75].

Despite the increasing number of published studies which have used MBIs to promote mental health and wellbeing [76], there is a lack of pretest-posttest design randomized controlled studies which have used EEG to assess the changes following a meditation intervention especially among healthy novice meditators. The main results of such studies are primarily related to changes in alpha and theta activity.

Theta activity has been observed to rise in different types of meditation practices, and there is a positive association between the amount of training and experience in each practice and the presence of theta activity [77]. Frontal midline theta, which have been related with concentrative attentional engagement [78], are often reported to increase following meditation [77] and their increase following a meditation program have been indicated as the cause of increased brain plasticity and connectivity [79], [80].

A study [81] demonstrated, assessing physiological and neurophysiological parameters, that the Integrative body–mind training (IBMT), which combines aspects of mindfulness and meditation, can foster processes of self-regulation and attention in the central an autonomic nervous system. The authors reported in the treated vs control condition a power increase in the theta frequency band for frontal midline electrodes Fz, FCz, and Cz, which could reflect activity in the anterior cingulate cortex, which is related with self-regulation of cognition and emotion [82], [83]. Moreover, the intervention led to lower heart rates, chest respiratory rates, and skin conductance responses, as well as increased high-frequency Heart Rate Variability (HRV) and belly respiratory amplitude in the IBMT group. A comparison to the relaxation control group suggests that there have been improvements in the regulation of the autonomic nervous system, specifically indicating enhanced parasympathetic activity. Lastly, the authors reported a significant correlation between frontal-

midline theta and high-frequency HRV in the IBMT group, indicating enhanced central and autonomic nervous system interaction.

The default mode network (DMN) is a network of brain regions that is particularly active when the mind is at rest or not focused on the outside world. The DMN is strongly associated with mind-wandering [84], [85] and self-referential activity [86] DMN activity is negatively correlated with frontal and midline theta power [87], [88]. On the other hand, the posterior regions of the DMN have been found to be positively related to alpha power [89]. Berkovich-Ohana et al. [90] pointed out a discrepancy in the existing literature regarding the impact of meditation on DMN: some studies indicate that meditation training may reduce the DMN activity while others report an opposite effect of meditation-induced increased activity within the DMN, both in frontal [91] and in posterior regions [81].

In a study [92] reported a beneficial effect of a 4-week Mind–Body Training (MBT), the combination of mindfulness and movement-based meditation, on anxiety, trait anger, and functional connectivity (FC) within the default mode network (DMN) relative to the theta and alpha frequency bands. Those results provide further evidence for beneficial effect on MBIs on both psychological and neurophysiological aspects.

A study [93] reported that individuals with non-clinical high trait anxiety fail to synchronize DMN during resting-state, with a decreased theta connectivity between right medial prefrontal cortex and right posterior cingulate/retrosplenial cortex and decreased beta connectivity between right medial prefrontal cortex and right anterior cingulate cortex. The results also indicated that DMN lower functional connectivity was associated with higher trait anxiety.

In a study conducted among participants with mild to high levels of generalized anxiety [94], the authors employed a Virtual Reality (VR) meditation in contrast to a resting condition to explore the relation between EEG activity and self-report anxiety measures. The authors reported that anxiety scores were significantly inversely correlated with the relative alpha power, which is the proportion of alpha brainwaves relative to the total power of all frequencies. Moreover, at the sub-band level it was found that lower but not higher alpha frequencies supported this relationship. Conversely, higher but not lower beta frequency significantly predicted higher anxiety scores. Following one session of VR meditation, the intervention group showed reduction in Low-Beta/High-Beta power ratio, while no difference was found in anxiety measures. Trambaiolli et al. [95] tested if global EEG connectivity can predict anxiety symptoms and severity in depressive participants and healthy controls, found that the most relevant bands are alpha and beta, with the alpha2 band coherence presenting the strongest relationship with anxiety. In another study [96] there was found a positive relationship between the scores of states and trait anxiety and alpha relative power, while there was a negative relationship with delta relative power, independently of the cortical site being analysed. Additionally, the study discovered that the strength of the reciprocal relationship between alpha and delta oscillations, known as alpha-delta anticorrelation, was positively associated with trait anxiety.

A study [97] reported theta power decreasing across different sites of the scalp and a theta coherence increasing after a three-months transcendental-meditation practice, together with an overall state anxiety and cognitive worry symptoms decreased, while it was solely found a non-significant trend of increased alpha coherence in the frontal region. The authors suggested that the theta band is more sensitive to meditation practice than the alpha and that left hemisphere is more

modulated by meditation. The authors stated that theta power decrease following their intervention is in line with the high theta found in ADHD patient and with previous study on adolescents with ADHD, which reported decreased theta/beta power ratio after the training with the same type of meditation i.e., transcendental meditation [98]. Another study [99] found that following training, the MBSR group exhibited significant improvements in mindfulness and emotion regulation levels, which were positively correlated with the EEG power changes the theta, alpha, and low-beta frequency bands.

Brandmeyer et al. [77] suggest that changes in the power of lower frequency bands should be interpreted cautiously since many meditation EEG studies consistently observe bidirectional alterations in both in terms state and traits in the alpha and theta band.

Despite the field of contemplative neuroscience is growing rapidly, many authors highlight that contemplative neuroscience is still in its infancy and much more must be done. Longitudinal studies that trace the effects of specific meditation practices on neuroanatomy are needed to establish causality; functional studies with more appropriate control conditions and more control on lifestyle variables could help to understand the differences in the results reported in the literature; studies that investigate behavioural outcomes along with neural functioning and that involve clinical samples will be useful to test hypothesis derived from existing theories. Those are some of the challenges and directions that contemplative neuroscience should carry in the future.

1.4 Integral Meditation

Much of the early work on mindfulness interventions used a nonrandomized pretest post-test design; however, beginning in the early 2000s, there was a dramatic increase in randomized control trials that compare mindfulness interventions to treatment as usual, wait-list control, or active comparison interventions [10]. Among those interventions, called mindful based programs (MBPs), the most well-known is the mindfulness-based stress reduction (MBSR) program, aimed at providing participants with instructions and experiences in mindfulness practices, as well as guidance on integrating mindfulness into everyday life to facilitate increased well-being and reductions in psychological distress [100]. A study involving MBSR participants reports that the formal time spent in practicing mindfulness predicted increases in the self-reported tendency to be mindful in daily life, which, in turn, mediated reductions in stress and improvements in psychological functioning [101].

The MBSR program was created in the 70's by Jon Kabat-Zinn at University of Massachusetts and it was structured as an 8-week group format, during which participants meet weekly for a group session of 2.5 hours in addition to one daylong retreat that lasts 6 hours, for a total program duration of 26 hours. Even though the MBSR programs are particularly efficient targeting clinical population who suffers from psychological or medical stress-related condition, MBSR programs produce positive results among an array of clinical and nonclinical populations, including cancer patients, mixed illness populations, health care professionals, continuing education students, and college undergraduates [102]. Another example of MBP is the Mindfulness-Based Cognitive Therapy (MBCT), that is an approach to psychotherapy that was originally created to be a relapse-prevention treatment for individuals with major depressive disorder.

Other MBPs exist alongside MBSR and MBCT, also emphasizing the practice of mindfulness meditation as a central pedagogical component. Each of these programs can differ from the others in the structural features to fit the target population and context, such as the duration of each session of meditation and the length of the program. It should be noticed that despite MBPs were originally designed to be delivered in groups, the role of social and group processes has been often neglected in the literature compared to the attention given to individual outcomes [103].

Since the MBPs have become so popular, some fundamental questions have been addressed among the researchers to achieve a sustainable development of the field. A first question that needs an answer is about how to define a MBP since nowadays the term mindfulness could refer to a multitude of meaning and practices. A group of researchers discussed this topic in the paper "What defines mindfulness-based programs? The warp and the weft" [104]. Using a metaphor, they compare the textile manufacture to the 'fabric' of MBPs and they called as 'warp' the essential, constant, and integral threads that define an MBP regardless of population or context; those are the elements which make it a mindfulness-based program. Then they call 'weft' those specific features that characterized each adapted MBP to fit better for a particular population or context.

Taking the 'warp' elements as reference points we developed a program called "Integral Meditation" (IM) as a central part of the intervention. The IM incorporates different oriental meditation techniques (e.g., Vipassana, Tibetan, Transcendental, etc.), developed with the aim to be suitable for beginners, allowing them to reach psychophysical wellbeing quickly. This technique integrates mindfulness with compassion and kindness and this integration is manifested primarily in the relationship with oneself concerning internal peace, genuine well-being and self-esteem mediated by the ability of acceptance and openness to life. All in all, the goal of IM is to give a tool to maintain emotional balance, to develop the ability to face different relational situations, and skills to deal constructively with stressful and existential situations.

In previous research we tested the effect of IM on different samples. The IM program was firstly developed and tested in a pilot study started in 2017 [105] where we examined the impact the IM program, including lectures on neuroscience of meditation, on psychological indicators in a sample of 41 participants from the general population. Using scores from self-report questionnaires as outcomes, the results revealed positive effects on well-being, mindfulness, and emotion regulation. Specifically, there was a significant decline in global distress and improvements in subjective well-being. Mindfulness levels increased, especially in non-judging and acting with awareness domains. Emotion regulation demonstrated a decrease in expressive suppression and an increase in cognitive reappraisal. While this study brought promising results, strong limitations such as the absence of a control group and a small sample size were present.

In subsequent research [106], we investigated the causal impact of the IM on psychological factors among a large sample from the general population (N=334) using a randomized controlled trial. Results showed statistically significant improvements in the treated group, including decreased distress, enhanced subjective well-being, increased mindfulness, and improved satisfaction with life. While the study suggested a beneficial effects of the intervention on various aspects of psychological well-being, limitations included the voluntary-based enrolment, potential biases in self-report measures, and the possibility of respondent fatigue due to the length of the assessment.

Continuing to explore the effect of the IM, we tested the intervention in a non-clinical population of adult workers (N=42) [107]; in this case the IM was administered after working hours in a company's office room. While no significant effects were observed on distress, state anxiety, and positive and negative affective states, a positive trend of these outcomes emerged in the pre-post intervention in the treated group compared to the control group. On the other hand, significant changes were identified in specific mindfulness domains, particularly in the observing subscale of the FFMQ. The intervention exhibited a significant positive effect on self-compassion and enhanced interoception sensibility was evident, particularly in emotional awareness, self-regulation, body listening, and trusting domains of the MAIA. Moreover, the IM intervention effectively improved mental well-being and reduced perceived stress.

When the COVID-19 pandemic began, resulting in lockdowns across Italy, we adapted the IM to be delivered online via video call meetings and we tested its efficacy during this abnormal situation [108]. Participants (N=58) were categorized based on their trait anxiety levels, the IM intervention's impact varied depending on trait anxiety levels. Stratified analysis showed a significant treatment effect in the group with higher trait anxiety, indicating improvements in state anxiety and well-being. However, no significant treatment effect was observed in the lower trait anxiety group.

In another study conducted during the pandemic we examined the impact of the IM on sleep quality, quantity, emotional regulation, and mindfulness among individuals experiencing poor sleep (Fazia, et al., 2023). Our findings revealed a significant positive effect of the intervention in improving sleep quality and reducing insomnia symptoms. Notably, the intervention led to a decrease in the prevalence of insomnia conditions, with a substantial portion of participants reporting no poor sleep quality post-intervention. However, there were no significant changes in predisposition to arousability or sleep reactivity, possibly due to the stability of these traits. Sleep hygiene behaviours, did not show significant changes, suggesting that the intervention's benefits may have been more immediate (stress relief, relaxation) rather than altering habitual behaviours. Emotional regulation strategies and dispositional mindfulness showed limited effects, except for non-reactivity to inner experience. This suggests that the intervention efficiently addressed immediate sleep-related needs but may require more consistent practice to induce deeper shifts in emotional regulation and overall mindfulness levels.

In the latest research using IM intervention, the latter was tested among a large sample of medical students (N=362) and the intervention was delivered online [110]. The study revealed a significant positive effect of the intervention on various psychological indicators, including reduced perceived stress, improved mental well-being, emotional regulation, resilience, reduced mind wandering, enhanced attentional control, and decreased overall distress. A post-hoc analysis suggested that improvements in emotional regulation mediated the positive effects on psychological well-being.

The studies assessing the impact of the IM program across diverse populations and consistently demonstrated positive effects on various psychological domains. While the accumulated evidence underscores the beneficial impact of IM on psychological well-being, it is essential to acknowledge the reliance on self-report measures in these studies. Strengthening the robustness of these findings involves integrating additional objective measures, such as electroencephalography (EEG), to deepen the understanding of IM effects. These complementary techniques offer a more comprehensive grasp of the neurophysiological mechanisms driving observed improvements, affirming the positive

outcomes associated with the IM program. This integrative approach fosters a nuanced and holistic comprehension of the intervention's effectiveness, supporting its potential integration into broader mental health practices.

1.5 Hypotheses of the study

In order to test our a priori hypothesis of a causal beneficial effect of our intervention, the IM training, on psychological indicators as dispositional mindfulness, interoceptive sensibility and accuracy, state and trait anxiety and on neurophysiological indicators such as alpha frequency activity, we carried out a two-group pre-test-post-test experimental design in which participants were randomly assigned to the treated group, who underwent the intervention, and to the control group (waiting list), who did not meditate during the same period. The aim was to evaluate the between-subjects causal effect of this training on psychological indicators, measured through four self-report questionnaires: the Five Facet Mindfulness Questionnaire (FFMQ), the Multidimensional Assessment of Interoceptive Awareness (MAIA), the State-Trait Anxiety Inventory (STAIX-1 and STAIX-2), and through a laboratory task, the Heartbeat Tracking Task (HBTT), as well as on neurophysiological indicators such as Individual Alpha Peak (IAP) power, IAP frequency, and Alpha Relative Power. We tested the following hypotheses:

- 1. There are improvements in the behavioural measures of mindfulness, interoception, state and trait anxiety for the treated group compared to the control group following the intervention indicating more optimal psychological status.
- 2. Participants in the IM group compared to non-active controls showed changes in the neuroelectric activity of the brain related to the alpha frequency band.

Additionally, we will explore the correlation between changes in behavioral measures and those in alpha frequency band measures to determine if there is a correspondence between changes at the behavioral and neuropsychological levels.

2. MATERIAL AND METHODS

2.1 Participants

The target population for this study is young adults between 19 and 35 years of age. Since the laboratory session and the intervention were held in Pavia, a mid-small university student city in Lombardy, we focused the advertising campaign on the area of Pavia and nearby. We used digital and no-digital channel to reach out the young adults. We displayed posters outlining the research requirements and organization on the walls of various locations commonly frequented by young people, including university facilities, libraries, and bars. We utilized mailing lists, including those from the University bulletin, university course mailing lists, and student residences, to disseminate information about the current research to students. We also employed social media posts and targeted advertising to reach young adults in the Pavia area.

Given the high density of university students living in Pavia and the existence of meeting points and digital channels dedicated to students, it was easier to reach out university students more than the non-student young adults. Individuals who showed interest in joining the study and meet the inclusion criteria were contacted by email and received an informed consent and privacy policy, which they signed and agreed to participate in the study. Each eligible participant received a brochure that briefly describes the research design, objectives, and timeline of the study. The decision to join the meditation program was spontaneous and informed, which could indicate an inclination and interest in meditation topics.

The inclusion and exclusion criteria are listed below.

Inclusion criteria:

- Being a native Italian speaker or being able to understand Italian
- Age between 19 and 35 years
- Not suffering at the time of the study and/or not having suffered in the previous 3 years from any serious psychiatric illness or other major medical condition

Exclusion criteria:

- Having experienced at least one severe episode of anxiety, depression, or other psychological disorders (hypomanic, psychotic) in the past 6 months
- Suffer from or have suffered from seizures
- Being under drug treatment or having a drug addiction
- Being engaged in a meditation practice in the past year (at least one meditation session per week for more than two months)

These criteria were clearly stated during the recruitment call and were further checked by asking participants direct questions on these issues, thus, they were assessed solely based on the information reported by participants. Participants were asked to evaluate 'severe episodes of anxiety, depression, or other psychological disorders' based on the extent to which the episode disrupted their regular activities and functioning, the persistence of the episode, and whether it was caused by a specific negative event (e.g., grief) or occurred unexpectedly without a clear trigger. During the assessment of the inclusion and exclusion criteria, participants were invited to ask questions in case of any doubt.

Through the software G*Power 3.1 [111], a necessary sample size of 68 total subjects has been calculated to observe an effect size of f=0.3, with a probability of error α of 0.05 and a test power of 0.8. This calculation was performed considering a repeated measures ANOVA design, with two groups and two main measures. Given our previous experience with this type of research, we know that a high percentage of participants would dropout or not complete the questionnaires and/or EEG measures, we aimed to recruit a sample size 30% larger than calculated to counterbalance potential dropouts and exclusions.

2.2 Measures

2.2.1 Self-report measures

We selected four different self-report measures, or psychological questionnaires to measure participant specific behavioural aspects and psychological well-being. We were particularly interested in measuring the dispositional mindfulness, the anxiety and the interoceptive awareness,

and we selected four questionnaires to measure those psychological factors: FFMQ, STAI-X1, STAI-X2 and MAIA.

FFMQ

The FFMQ (Five Facet Mindfulness Questionnaire) is a 39-question multidimensional assessment instrument designed to measure a person's level of mindfulness [19]. Specifically, it aims to measure five interrelated components of mindfulness, which are:

- 1. Observing (noticing or attending to internal and external experiences),
- 2. Describing (labelling internal experiences with words),
- 3. Acting with awareness (attending to own activities of the moment and it can be contrasted with behaving mechanically while attention is focused elsewhere),
- 4. Non-judgment of inner experiences (taking a nonevaluative stance toward thoughts and feelings),
- 5. Non-reactivity to inner experience (the tendency to allow thoughts and feelings to come and go, without getting caught up in or carried away by them).

A higher overall score in the FFMQ scale as well as in its sub-scales reflects a higher level of mindfulness. The questionnaire showed good psychometric properties in both the English and Italian versions, which also shows a similar factorial structure compared to the original version [112].

STAI-X1 and X2

The STAI-X (State-Trait Anxiety Inventory) [49] questionnaire consists of two parts: the X1 scale assesses state anxiety to investigate how the patient feels at the time of the assessment while the X2 scale assesses trait anxiety reflecting how a person generally feels. Each consists of 20 questions and the answers are given in a 4-point Likert scale. Participants were asked to rank themselves with respect to certain statements on the STAI-X1 scale from "not at all" (1) to "very much so" (4), and on the STAI-X2 scale from: "almost never" (1) to "almost always" (4). The values obtained in each of the scales range from 20 to 80 points, with the range 20-40 described as low anxiety, 41-60 as moderate anxiety, and 61-80 as high anxiety. We used the STAI-X1 and STAI-X2 questionnaire as the dependent variable to measure the effect of treatment on each of them. The questionnaires have good psychometric properties in both the English and Italian versions [113].

MAIA

The MAIA (Multidimensional Assessment of Interoceptive Awareness) [25] is a multidimensional questionnaire with 32 questions in which answers are given on a scale of 1 to 5. This questionnaire investigates various aspects of both positive and negative interoception, that is, the perception of the internal state of one's body. Specifically, MAIA assess the interoceptive sensibility, the subjective perception, and beliefs about the internal focus and/or accuracy of an individual in perceiving

interoceptive signals. The MAIA scale is divided into eight sub-scales, conceptually organized into five dimensions:

- 1. awareness of bodily sensations (sub-scale Noticing),
- 2. emotional reaction and attention response to sensations (sub-scales *Non-distracting* and *Non-worrying*),
- 3. ability to regulate attention (sub-scale Attention regulation),
- 4. awareness of mind-body integration (sub-scales *Emotional awareness*, *Self-regulation*, and *Body listening*),
- 5. physical feelings of trust (sub-scale *Trusting*).

This questionnaire has good psychometric properties both in original and Italian version [114].

2.2.2 Heartbeat Tracking Task

The Heartbeat Tracking Task (HBT) [26] is a task to assess the participant's interoceptive accuracy. During the task, participants were focused on their heartbeat and tried to estimate their heartbeats. The HTT measures the accuracy of perceiving and counting heartbeats during a of period.

2.2.3 EEG recording

To assess neuropsychological changes associated with the mindfulness meditation intervention, we recorded participants' brain activity during an eye-closed resting state condition using electroencephalography (EEG). EEG is a non-invasive technique that measures electrical activity in the brain, providing valuable insights into neural dynamics and connectivity. The primary objective of EEG recording in this study is to investigate alterations in specific brainwave patterns, particularly focusing on alpha waves. Alpha frequency band, oscillating at a frequency of approximately 8 to 12 hertz, are associated with a relaxed and wakeful state of consciousness. Research suggests that changes in alpha wave activity may reflect shifts in attention, cognitive processing, and overall mental state. By concentrating on alpha frequency, we aim to discern whether the mindfulness meditation intervention induces observable modifications in participants' neural oscillations during the resting state. The EEG recording us to capture the participant's baseline neural activity. This neurophysiological approach complements the self-report measures and laboratory tasks employed in this study, providing a comprehensive understanding of the intervention's effects on both subjective experiences and neural correlates.

2.3 Procedure

2.3.1 Participants enrolment

The recruitment phase commenced in mid-September 2022 and concluded at the end of October 2022. Prospective participants expressing interest during the recruitment phase were provided with comprehensive details regarding the study's organization and eligibility criteria. Those who confirmed they enrolment in the study were assigned a unique ID and were randomly assigned to either the control or intervention group using a simple randomization method, which ensured

unbiased allocation of participants. The randomization procedure was done using the software R [115] using the functions 'set.seed' and 'split'. With the first, we ensured the reproducibility of the random number generation, while we used the latter to divide the participants into the control and intervention groups randomly.

A total of 89 participants were enrolled in the study. Subsequently, participants were contacted via email to receive additional information about their assigned group and the experimental procedures. Each participant received an email containing three essential documents: the study information and objectives, the Informed Consent form, and the Privacy Policy. Furthermore, participants were directed to a Google Form page where they had to enter their unique ID and completed the four self-report assessments outlined in the previous chapter. Additionally, the form included a sociodemographic questionnaire tailored to collect general information and details relevant to the study. Participants were also requested to indicate their preferred day and morning time for their laboratory visit to complete tasks and EEG measures before the starting of the intervention. Only those assigned to the intervention group received a calendar outlining the date, time, and location of the mindfulness meditation intervention. The control group employed in this study was an inactive control, selected due to resource constraints that hindered the implementation of a parallel intervention for comparison. Although the absence of an active control group may diminish the relevance of the results, this choice was motivated by practical considerations.

Participants in the control group were informed that they would not participate in the initial meditation training but would be placed on a waiting list, offering them the opportunity to engage in the meditation training after the study's conclusion. Importantly, participants in the control group were instructed not to engage in meditation practices during the study period. After the intervention, we sent another email to the participants asking them to complete the questionnaires again via Google Form. Participants were then instructed to schedule their post-intervention laboratory session.

2.3.2 Laboratory session

The laboratory measurements took place during the same time of the recruitment phase, and lasted a bit longer, until the starting of the intervention. Each participant came to the laboratory the day of the appointment to complete the EEG and HBTT measurement. A few days before the participant was scheduled to come to the laboratory, we sent them important information on how to reach the laboratory and a do's and don'ts list to ensure that participants would undergo the EEG recording in optimal condition. The do's and don'ts list included:

- The evening before the laboratory session, have a full night's sleep (at least 6 hours).
- Avoid alcohol or drug consumption the evening before or on the day itself.
- Try to not consume coffee, energy drinks or Coca-Cola within 2 hours before the laboratory session.
- Refrain from smoking at least 1 hour before.
- Wash your hair at least once in the 2 days prior to the recording and do not use conditioner or other leave-in products after the last wash.
- Have a light breakfast if you will be in the laboratory within the next 2 hours.

When participants arrived at the laboratory, they were asked to sign the Informed Consent and Privacy Policy that they had previously received via email; they were also invited to ask any questions they might have regarding the study.

EEG recording

The EEG signal was acquired using a Mitsar-EEG-201 BT amplifier with 21 EEG channels and 4 poly channels connected to a laptop via USB. Data from 19 EEG channels placed over the scalp following the International 10-20 system were recorded using waveguardTMconnect EEG caps to transmit the signal from participant's scalp to the amplifier (**Figure 3, Figure 4**). The data was acquired using WinEEG software [116] and stored on a dedicated laptop. We used linked ears as a reference electrode. Signals were digitalized at 500 samples per second with a low-frequency filter of 0.5, a high-frequency filter of 50 Hz and and a 50Hz notch filter to remove electrical main noise, and impedance levels $\leq 5 \text{ k}\Omega$. To ensure consistency across the procedure and setting, all the EEG recording were conducted by the same researcher, which has previously gained knowledge and practice about the EEG recording.

Upon arriving at the laboratory, participants were provided with brief information about the EEG recording procedure, including its overarching purpose in the study. They were then invited to sit comfortably on a chair, facing an empty white wall. Head circumference was measured to ensure the selection of the appropriate EEG cap size (Small: 47-51 cm, Medium: 51-56 cm, Large: 56-61 cm). All participants underwent a conventional EEG registration, which included 5 minutes of eyeclosed resting state condition recording and 5 minutes of eye-opened resting state condition recording. During these sessions, participants were instructed to be in a sitting position, mentally and physically relaxed, and awake. To enhance data quality, participants were advised to get a full night of sleep and abstain from alcohol or drugs the day before the recording. Prior to the recording, participants were educated about potential artifacts caused by certain movements (eye movements, blinks, etc.) and were encouraged to minimize these movements without experiencing distress or discomfort. Participants were told to request a pause in the recording if they were feeling excessively discomfort or encountered any issue. Between the eye-closed and eye-opened conditions, participants were given a minute to rest. During this brief interval, the researcher ensured that everything was fine with the participants and checked that the impedance level remained below the designated threshold.



Figure 3. EEG Equipment: a Mitsar-EEG-201 BT Amplifier and waveguard[™] connect EEG Caps.

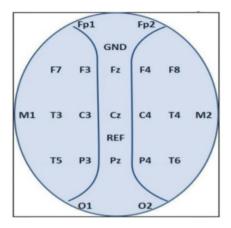


Figure 4. EEG Cap Layout with Electrodes Placement Following the International 10-20 System.

Heartbeat Tracking Task

After the completion of the EEG recording, participants engaged in the Heartbeat Tracking Task (HBTT). In this task, participants were instructed to sit comfortably with their eyes closed, arms resting on their legs, and count their own heartbeats over specific time intervals. Simultaneously, the experimenter measured the participants' actual heartbeats using two electrodes placed on their wrist, following the standard bipolar limb lead II configuration. These electrodes were connected to the poly channels of the same amplifier utilized for the EEG recording. Using WinEEG software [116], the researcher observed the electrocardiogram (ECG) of each participant to accurately count the heartbeats. The HBTT comprised six trials, each with varying time intervals (25s, 30s, 35s, 40s, 45s, and 50s). To mitigate order effects, the presentation of these time intervals was randomized for each participant. After each trial, participants were prompted to report the number of heartbeats they perceived. Once ready, they initiated the next trial. The experimenter indicated the start of each trial, and an auditory cue indicated the end of each trial.

2.3.3 Intervention

Participants in the intervention group were provided the opportunity to join a mindfulness meditation training program. The training commenced in mid-November 2022 and concluded in mid-February 2023, spanning a total of three months. The program comprised a series of 12 classes distributed throughout this period. While it was not mandatory for participants to complete the entire course, we strongly encouraged their active participation in as many meditation classes as possible.

The IM training that our research team developed and tested in previous studies [106], [107], [108], [109], [110], [117] integrates meditation practice with the social components involved during the meditation classes.

Those social aspects refer to: (a) participating in a gathering of people that share the same intention (i.e., to meditate) that may influence the motivation and the feelings that one has toward the activity and (b) to the environmental differences, such as the possibility of socializing, of group meditation compared to individual meditation that may generate psychological effects distinct from those that arise from individual meditation practice.

In the following we first discuss and characterize our meditation training in respect to a classical MBP. The main substantial difference between our intervention approach and MBPs is that MBPs such as MBSR and MBCT have been used to address health issues, while ours is aimed to promote psychological well-being and personal growth in a non-clinical population. Nevertheless, there are many common features between MBPs and our intervention which we discuss in here according to the metaphor of "wrap" and "weft" used to represent the fabric of MBP [104].

Our program, we named IM is spiritual in nature and shares the following "wrap" features with MBP:

- it uses mindfulness practices as a vehicle for a systematic training of the mind in the service of developing greater awareness of self and others and it is informed by theories and practice that draw from contemplative traditions, while leaving behind their religious, esoteric, and mystical elements;
- t is underpinned by a model of human experience that addresses the causes of human distress and the pathways to relieve it;
- it develops a new relationship with experience characterized by focusing on present moment, and on decentring (i.e., by considering thoughts and feelings as mental events which come and go in the mind as clouds in the sky). The training enables the participants to make a radical shift to their thoughts, feelings, and body sensations, as well as to outer circumstances;
- supports the development of greater attentional, emotional, and behavioural self-regulation, as well as positive qualities such as compassion, wisdom and equanimity by cultivating an internal climate of friendliness towards experience whether it be pleasant or unpleasant;
- the training develops familiarity with and understanding of the mind and body and appreciation that attention can be regulated, fine-tuned, and optimized through training. Rather like physical training, the training progresses developmentally and sequentially throughout the program.

The "weft" feature of our program resides in:

- a) using imagery to enhance concentration, leading to a different state of consciousness;;
- b) being scheduled over 12 weekly guided sessions lasting 60 min each. The 12 sessions are structured into 4 cycles, each cycle comprises three subsequent meditation sessions. Each cycle is focused on a specific ability as reported below:

Cycle 1: aware diaphragmatic breathing, maintaining the posture

Cycle 2: body scan and awareness of body sensations

Cycle 3: emotions and thoughts feeling and releasing

Cycle 4: imagery activity to change the state of consciousness

Each cycle also incorporates the abilities acquired in the preceding cycles, resulting in an evolution across cycles.

The last cycle, i.e., Cycle 4, also comprises all the abilities acquired in the preceding cycles and is hence structured into the following six final phases:

- Maintaining posture during meditation, focusing on an aware diaphragmatic breathing, body scan and awareness of sensations to induce body relaxation leading to an inner emotional calm that is tightly linked to the calm of the physical body; (abilities acquired in Cycle 1 and Cycle 2). Duration 10 min.
- Feeling and examination of emotion and thoughts; (abilities acquired in Cycle 3).
 Duration 5 min.
- 3) Visualization of images (e.g., a sphere of light), colours, relaxing landscapes (e.g., a garden). Visualization powers concentration and calms negative and disturbing dominant emotions. This is not yet a meditation state, but a state leading to the true meditation experience; (abilities acquired in Cycle 4). Duration 20 min.
- 4) A time of silence to enjoy the new state of consciousness. After phase iv) the person is free of body sensations, emotions, and mental processes and ready to expand his/her own consciousness and open fully him/herself with trust to the union with the higher self and the unified universal field, thus entering a fine state of meditation to enjoy his/her own personal experience; (abilities acquired in Cycle 4). Duration 15 min.
- 5) Reconnection to the feeling of the body sensations; (abilities acquired in Cycle 4). Duration 5 min.
- 6) Awareness of the new mental and physical state (well-being, happiness, peacefulness, serenity etc.). (abilities acquired in Cycle 4). Duration 5 min.

Additional elements of the program:

- 7) Sharing the experience: at the end of each meditation class, the trainer asks the participants how they feel to allow them to freely share their feelings and impressions about the meditation experience.
- 8) Practice at home: participants were strongly encouraged to engage in meditation practice on their own. To facilitate this, they were given two audio files featuring guided meditations. The first audio, an 18-minute guided meditation, was distributed during the

third class. The second, lasting 25 minutes, was distributed during the eighth class. Both sessions involved a repetition of the exercises learned in previous classes. was distributed during the eighth class. Both sessions involved a repetition of the exercises learned in previous classes.

The meditation classes were held in a lecture hall at the University of Pavia (**Figure 5**), where participants meditated together seated on chairs. We consider the effect of meditate in group as part of our intervention. The classes were attended by several participants at the same time, and they had the opportunity to talk to each other and socialize before and after the class.

The meditation trainer had the competences and the experience for teaching meditation, and he used them to facilitate the beginners to acquire the technique and feel some benefits quickly.



Figure 5. Photo During an Integral Meditation Class

3. STATISTICAL ANALYSIS

3.1 Data management, pre-processing, and descriptive analysis

In this section we are going to describe the procedure used to clean, transform, and pre-process the data before to conduct the hypotheses testing analysis. The description of the procedure is divided into three categories according to the type of measurements employed in this study: self-report questionnaire, the interoceptive accuracy task and the EEG recording.

3.1.1 Sociodemographic information

We downloaded from Google Form the excel sheet containing the sociodemographic information of the participants. This information was imported in the statistical software R [115] which was used for all the subsequent analysis except for the EEG preprocessing. Each sociodemographic variable's

name was recoded with a short unique name and the answers were recoded from text type answers to discrete numbers, indicating the different categories. In some cases, the categories were reorganized to include different type of answers if enough similar (for example, the answers to 'Degree field' were rearranged into 'Scientific' and 'Humanistic' categories to include different type of degree field under the same category).

To describe the general characteristic sample of this study, we calculated descriptive statistics for the sociodemographic variables. Subsequently, we compared the two groups to observe if there were any relevant differences among the two groups (control and intervention).

For the sociodemographic variables, we calculated:

- a) the mean and standard deviation for each group in case of numeric variables
- b) the frequency for each group in case of categorical variables

and then the two groups were compared with the following procedure:

- a) in case of numeric variables, at first, we checked the distribution and the variance among the two groups. To assess the normality of the data, we employed the Shapiro-Wilk test. Subsequently, to assess the homogeneity of variances between groups, a variance test was performed. Based on the results of the normality test and variance test, the appropriate statistical test was selected. In case the data met the assumptions of normality and homogeneity of variances, a parametric test (t-test) was employed. When the assumption of equal variances was violated a Welch's t-test was performed. When both the assumptions were violated, indicating non-normally distributed and heteroscedastic data, a non-parametric test (Wilcoxon test) was chosen.
- b) in case of categorical variables, we used chi-squared test when the expected frequency in each cell was greater than or equal to 5 or Fisher's exact test when expected frequencies were less than 5 in any cell.

3.1.2 Self-report measures

The participants' answers to the questionnaires were downloaded from Google Form and saved in an excel file. The two excel files containing the t0 and t1 participants' answers for the questionnaires were cleaned from empty and non-complete entries and rearranged to facilitate the analysis. After this preliminary step, the data were opened with the software R to be processed before the analysis. Each column of the dataset containing a single questionnaire item was recoded with a unique name indicating the name of the questionnaire and the number of the item (for example, the first item of the FFMQ was renamed as 'FFMQ_1'). Where the participants' answers to the questionnaire were expressed as a text type answer, we recoded the answers to the corresponding numerical values. To reach the final score of each questionnaire, it is necessary to proceed with the inversion of the coding of some items, as indicated by instruction of the questionnaire, and the final calculation of the mean or sum of the scores of each individual item after recoding.

For STAIX1 we have reversed the following items: 1, 2, 5, 8, 10, 11, 15, 16, 19. To find the final score, we summed the scores of each individual item. For STAIX2 we have reversed the following items: 1, 6, 7, 10, 13, 16, 19 and we found the final score by summing the scores of all items. For

FFMQ questionnaire, we have reversed these items: 3, 5, 8, 10, 12, 13, 14, 16, 17, 18, 22, 23, 25, 28, 30, 34, 35, 38, 39. To find the scores for each subscale we summed the scores of the items belonging to each subscale. Finally, we added up all 39 items to find the total score.

In MAIA questionnaire only items 5, 6, 7, 8, 9 were reversed. Like what we did with the FFMQ questionnaire, we found the individual score of each subscale as the mean of the items belonging to it, and eventually the total score calculating the mean across all the items and subscales.

We finally checked the internal consistency of each questionnaire. Cronbach's alpha is a statistical index widely used nowadays to measure the internal consistency or reliability of a questionnaire consisting of quantitative questions (items) on Likert scales [118].

When we talk about the internal consistency of a questionnaire, we are talking about analysing whether the answers given to this questionnaire are consistent with each other, that is, they follow a consistent thread among them, relating to each other. If the items are highly correlated with each other, there is a high internal consistency and thus a good measure of reliability; a low correlation, on the other hand, is a warning of low reliability or low internal consistency.

The formula for calculating Cronbach's alpha is as follows:

$$\alpha = K \cdot \frac{\overline{r}}{1 + (K-1) \cdot \overline{r}}$$

where:

- K=number of items
- \bar{r} =mean of correlations calculated before.

This coefficient is always between 0 and 1 and if it is close to 1, it means that the internal consistency is high.

In our case, we calculated the Cronbach's alpha for each questionnaire and its subscale at both times (t0 and t1) using the 'alpha' function from the package 'psych' [119] in the software R. In this case the whole sample was included in the calculation of the internal consistency.

To describe the scores of the two groups in the self-report assessment, we calculate the mean and the standard deviation of for each scale and subscales in the two timepoint. To compare the two groups, we followed the same strategy as described above for the numeric sociodemographic variables.

3.1.3 Heartbeat Tracking Task

We used the Heartbeat Tracking Task (HBTT) [26] to assess the participant's interoceptive accuracy. The data collected from every participant in the laboratory were saved in an excel file and opened in R; to derive a scores of interoceptive accuracy, the raw data from the six different trials were analysed with the following formula, derived from the literature [53]:

$$1 - \frac{|\sum BR - \sum BC|}{\sum BR}$$

Where BR corresponds to the actual beats recorded during each trial and BC corresponds to the beats counted by the subject in each trial. The scores obtained are usually distributed between 0 and 1, where 1 refers to perfect performance on the task; negative values are rare but can be obtained when the difference between BR and BC is very high. Afterward, for each timepoint we calculated the interoceptive accuracy mean and the standard deviation of the groups and the scores at t0 of the two groups were compared using a Student's t-test.

3.1.4 EEG

After recording the EEG data, we took several steps to primarily clean the data through a preprocessing procedure, a crucial step to remove noise and artifacts, allowing for a clearer interpretation of the underlying neural signals. After preprocessing the EEG signal, we extracted from the data the relevant feature to perform the group analysis. Besides we recorded EEG signal during both eye-closed and eye-opened conditions, we only analysed the eye-closed condition data for this study, so from now on we are referring only to such data while describing the EEG analysis.

Preprocessing and Fast Fourier Transformation

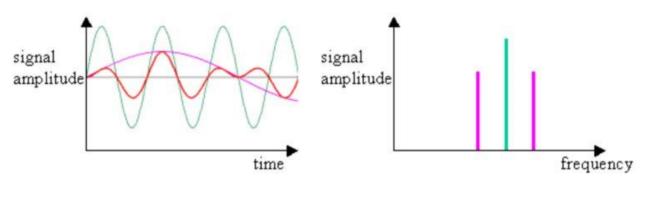
The original EEG raw data, exported in EDF format from WinEEG software [116], underwent a meticulous preprocessing pipeline to ensure the reliability of subsequent analyses. Initially, the EDF files were scrutinized in R to verify data consistency across participants at the two distinct timepoints t0 and t1. Following this verification, the files were imported into MATLAB for further processing, utilizing the FieldTrip toolbox [120], a specialized MATLAB toolkit designed for EEG analysis. For each file, a temporal adjustment was made, commencing from the 10th second, effectively eliminating potentially noisier initial segments of the recording. In subsequent stages, the dataset underwent band-pass frequency filtering (0.1 to 50 Hz) and a referencing step wherein EEG brain channels were referenced to EEG ears channels to enhance signal clarity. This referencing adjustment is strategically designed to optimize the interpretation of EEG signals by mitigating the impact of common noise sources. Specifically, electrodes proximal to the earlobes were employed as reference points to bolster the signal-to-noise ratio. This approach effectively diminishes interference from artifacts, consequently refining the spatial localization of EEG signals.

Notably, channels Fp1 and Fp2, positioned closer to the eyes, were designated as Electrooculogram (EOG) channels for the identification and removal of artifacts associated with eye muscular activity. Independent Component Analysis (ICA) was then applied to isolate components correlated with eye EOGs from the remaining channels. The selection of components related with eye movements was guided by visual inspection of signal activity over time and topographical representations. Following this, a meticulous visual inspection of individual channel signals, focusing on statistics such as variance, facilitated the identification and removal of channels exhibiting excessive noise. To further refine the EEG data an automatic artifact suppression method was employed. We first calculated thresholds that serve as reference points for identifying potential anomalies or artifacts within the EEG data. To determine these thresholds, considering the distribution of EEG values, we derived two crucial values: the maximum threshold and its corresponding minimum threshold. The maximum threshold represents the upper limit beyond which EEG values are considered abnormal.

It was computed by adding two times the standard deviation to the mean EEG value, reflecting the expected range of normal EEG activity. Conversely, the minimum threshold was determined by negating the maximum threshold, defining the lower boundary of normal EEG values. Once these thresholds were established, they provided criteria for identifying potential artifacts within the EEG data. Any EEG values falling outside the range defined by the minimum and maximum thresholds were flagged as potential artifacts, indicating abnormal signal activity. In some cases, adjustments were be made to ensure that the calculated thresholds align with expected EEG signal characteristics. Additionally, padding was applied to the artifact time-windows to ensure that the surrounding EEG data remains intact and unaffected by the removal process. Following artifact removal, the EEG data were subjected to temporal interpolation to address any gaps introduced by the removal of artifacts. This interpolation technique filled the missing points with estimated values based on the surrounding EEG data, effectively restoring the continuity of the EEG signal. The interpolation process ensures that most of the EEG trials were retained, minimizing data loss while effectively suppressing artifacts.

Following artifact suppression, a subsequent phase of pre-processing involved a refined frequency filtering to ensure that the EEG signals persisted within the specified frequency range. This step is essential to preserve the relevant neural information contained within the EEG data. The chosen frequency band (0.1 to 50 Hz) encompasses the spectrum of brain activity typically observed in EEG recordings. Frequencies below 0.1 Hz are often associated with non-neural low-frequency drift from the continuous EEG data, while frequencies above 50 Hz are typically considered noise or muscle artifacts. By confining the EEG signals to this specific range, the analysis focuses on the neural oscillations and activities relevant to cognitive processes, enhancing the interpretability of subsequent findings. Furthermore, during this frequency filtering stage, any linear trend, such as gradual shifts in amplitude over time, in the data was systematically removed. The last pre-processing step before frequency analysis was downsampling the EEG data to 250 Hz. Downsampling reduces the number of data points, saving computational resources and speeding up subsequent analyses. This balance ensures efficient processing of large datasets while preserving crucial temporal dynamics in the EEG signals. Overall, these pre-processing procedures listed above contributed to create a high-quality EEG dataset, free from artifacts and noise.

Each subject's pre-processed data was shortened to 305 seconds to ensure consistency across the data and underwent a segmentation into 10-second epochs with a 50% time overlapping. This choice aimed at ensuring a good balance in the resolution between time and frequency for resting-state EEG data. The decision considered obtaining a frequency bin of 0.1 seconds each, enhancing the precision to identify the alpha peak. The subsequent step included Fast Fourier Transform (FFT) analysis. FFT is a mathematical algorithm employed in EEG data analysis to transform time-domain EEG signals into their frequency-domain representation. This transformation is crucial for understanding the distribution of signal power across different frequency bands and involves the decomposition of the EEG signal into its constituent sinusoidal components (**Figure 6**).



Time domain

Frequency domain

Figure 6. Representation of Fast Fourier Transformation (FFT). The complex signal (red) is made up of simple sine waves (pink and green). A FFT expresses these sine waves in the frequency domain, where high amplitudes at certain frequencies become apparent. Image credit: National Instruments.

This process is instrumental in aiding the identification of specific features like the alpha peak, providing a comprehensive insight into the intricate frequency dynamics of the neural activity captured in the EEG data. In our study, the FFT focused on frequencies from 0.5 Hz to 30 Hz, capturing the relevant frequency range associated with EEG activity and providing insights into the dynamics of neural oscillations during resting-state conditions. FFT outcomes were organized into a novel data structure, encompassing amplitude spectrum, power spectrum, and associated frequency information. Individual power spectral density (PSD) plots were generated, illustrating EEG signal characteristics. These plots display normalize power across distinct frequency bands and alpha peak frequencies were identified on the plots and marked, providing a detailed visualization of frequency dynamics within the specified frequency range. In **Figure 7** two examples (one for intervention group and one for control group) of PSD plots are depicted, showing the pre and post distribution of the normalized power across the range of frequency 0-30 Hz. In **Figure 8** the PSD plots as a mean of each group are represented.

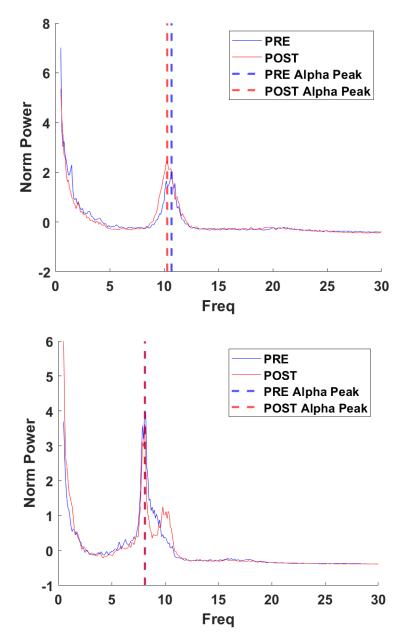


Figure 7: Pre and Post Eye Closed Resting-state EEG Power Spectral Density Plots. The plots represent an example from a subject of the intervention group (up) and a subject of the control group (down). In the plots is possible to observe the power spectral density for the frequencies across the range 0-30 Hz. A dashed line indicates the Individual alpha peak for the pre and post intervention data.

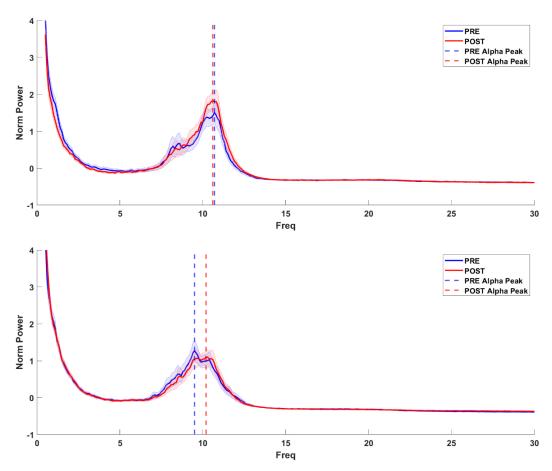


Figure 8: Mean Power Spectral Density (PSD) Plots. The plot contrasts, separately for the intervention group (up) and for the control group (down), the group's average PSD at baseline (t0) with the post-intervention (t1), represented by two distinct lines. Shaded error bars depict the standard error around each mean, providing a visual representation of the variability within the group. Each line signifies the mean PSD of the group at the respective time point, offering a clear visualization of the spectral changes induced by the intervention across the 0-30 Hz frequency range. Dashed lines denote the Individual alpha peak for both pre- and post-intervention data.

Individual Alpha Peak Frequency and Power

The IAP calculation is centred on the frequency range associated with alpha oscillations, specifically defined between 8 to 12 Hz. For each subject, the determination of the global IAP frequency involved identifying the frequency at which the normalized mean power across all EEG channels reached its maximum within the specified alpha frequency range. This global IAP serves as a collective metric, encapsulating the alpha oscillatory activity across the entire dataset for the subject. Simultaneously, IAP frequencies were extracted for each EEG channel by isolating and normalizing channel-specific power spectrum data. The frequency at which maximum power occurred for each channel was recorded as the channel IAP frequency, while the normalized power at the global and channel IAP frequency was documented as the global and channel IAP power. The results, encompassing global and channel-by-channel IAP frequencies and powers for each subject and structured for further analysis.

Alpha Relative Power

The extraction of Alpha Relative Power involved a systematic analysis of EEG power spectrum data, quantifying the power contributions of the distinct frequency bands. The predefined frequency bands, including Delta (0.2-4 Hz), Theta (4-8 Hz), Alpha (8-12 Hz), Beta (12-30 Hz), and potentially more as needed, served as key markers for neurophysiological activity. For each subject and EEG channel, the mean power was calculated within these frequency bands, providing insights into the distribution of power across different oscillatory ranges. The total absolute power for each channel was determined by summing the mean powers across all frequency bands. Subsequently, relative power indices were computed, representing the percentage contribution of the relative dominance of specific frequency bands within individual channels. Additionally, the total relative power across all channels and frequencies was computed, offering a global perspective on the overall power distribution. The results, in this case the Alpha Relative Power channel-specific and global for each subject, were organized into a structured format.

3.2 Group analysis and Partial correlation analysis

The final dataset included:

- The scores of the questionnaires and their subscales
- The interoceptive accuracy score
- The IAP frequency, as a global index and as a channel-by-channel index
- The IAP power, as a global index and as a channel-by-channel index
- The Alpha relative rower, as a global index and as a channel-by-channel index

The final dataset was imported on the software R and a series of analysis were performed to test the hypotheses of the study.

3.2.1 Effects of the intervention on the outcomes

The primary focus of group analysis was to understand the effects of our intervention on psychological and neuropsychological factors, here measured as questionnaire scores (FFMQ, MAIA, STAIX-1, STAIX-2), performance in the HBBT task, and key EEG alpha activity indices, including IAP power, IAP frequency, and alpha relative power.

To delineate the intervention's effect, we constructed linear regression models where the postintervention measurements (t1) served as the dependent variable (y). The intervention factor, distinguishing between the intervention and control groups, and baseline scores at t0 were included as independent variables. Additionally, we incorporated relevant covariates ('Sex' and 'Previous meditation experience') to account for potential confounding effects. In our analysis, we introduced weights into linear regression models to address variations in class attendance within the intervention group. The weights were calculated by dividing each subject's number of class attendances by the maximum attendance, resulting in a weight range of 0 to 1. This approach prioritizes subjects with higher attendance, acknowledging our a priori assumption that increased participation enhances the detectability of intervention effects. Subjects with maximum attendance received a weight of 1, emphasizing their influence in the models, while those with fewer attendances were proportionally weighted. All the subjects in the control group were assigned a weight of 1. This strategy enables the statistical models to effectively capture the potential impact of class attendance on observed outcomes.

In an initial exploration, we assessed whether the intervention's effect varied across different levels of baseline scores through fitting linear regression models with interaction terms. The interaction term quantified the nuanced impact of the intervention at varying baseline score levels, with significance determined by p-values (≤ 0.05). If interactions were not statistically significant, thus indicating uniform intervention effects across baseline levels, we refined the models to include the main effect of the intervention factor only.

The subsequent analysis was concentrated on the main effect only, unveiling the direct impact of the intervention on the dependent variables. The refined linear regression models included the intervention factor, baseline scores, and covariates, with the coefficient of interest β representing the intervention effect adjusted for baseline scores and other covariates. The value of the β coefficient, coupled with its associated p-values, provided insights into the extent of extent of change and its statistical significance induced by the intervention on behavioural scores and EEG indexes. This multifaceted statistical approach allowed us to discern not only the overall impact of the intervention but also its nuanced interactions with baseline scores.

In order to account for multiple comparisons and control the false discovery rate (FDR) in our statistical analysis, we employed the Benjamini-Hochberg method [121] using the function 'p.adjust' in the software R to adjust simultaneously all the p-values obtained in this study. This method is widely utilized in hypothesis testing scenarios where multiple comparisons are conducted simultaneously, such as in our study where we evaluated multiple p-values. The primary purpose of multiple testing correction is to mitigate the increased risk of falsely rejecting null hypotheses (Type I errors) that arises when conducting numerous statistical tests.

The Benjamini-Hochberg method operates by adjusting the individual p-values obtained from each test to control the FDR at a predetermined threshold level. Specifically, the method involves sorting the p-values in ascending order and calculating corresponding critical values based on their rank and the chosen FDR threshold. Subsequently, the method compares each p-value to its corresponding critical value. P-values that are smaller than their critical values are deemed statistically significant, while those exceeding the critical values are considered non-significant.

By applying the Benjamini-Hochberg method with a specified FDR threshold of 0.10 in our analysis, we ensured that no more than 10% of the statistically significant results are likely to be false positives. This correction approach strikes a balance between controlling the overall error rate and maintaining statistical power, thereby enhancing the reliability and validity of our findings. It is worth noting that the adjusted p-values obtained through this method provide a more conservative assessment of statistical significance compared to unadjusted p-values, reflecting a stricter criterion for declaring significance.

3.2.2 Exploratory analysis: Effects of the intervention across different brain regions

We performed an exploratory analysis to examine the impact of the intervention across diverse brain regions and varying baseline values, Linear Mixed Models (LMM) were employed [122]. The data from the 17 EEG channels were categorized into three regions representing the left hemisphere, right hemisphere, and midline electrodes. Electrodes within each region were aggregated as follows:

Left Hemisphere Electrodes: F3, F7, C3, T3, T5, P3, O1

Right Hemisphere Electrodes: F4, F8, C4, T4, T6, P4, O2

Midline Electrodes: Fz, Pz, Cz

In this analytical approach, each subject contributed multiple observations corresponding to the number of electrodes within the specific brain region of interest. Within each LMM, the post-test value of the EEG alpha activity index (IAP power, IAP frequency, or alpha relative power) represent the dependent variable, while the intervention factor, baseline values, and their interaction were treated as independent variables. The variables 'Sex' and 'Previous meditation experience' were included as covariates in the LMMs, and a random intercept for subjects (1|subject) was incorporated to address intra-subject variability arising from repeated measurements across different electrodes. This comprehensive approach allows us to capture and assess the specific changes induced by the intervention across different brain regions and alpha activity indices.

3.2.3 Partial correlation between behavioral Measures and EEG alpha activity

The Partial Correlation Analysis explored the nuanced relationships between changes in behavioural measures and alterations in IAP power, IAP frequency, and alpha relative power. We run two different Partial Correlation Analysis, one for each group. The analysis accounted for covariates such as sex and previous meditation experiences, utilizing the Pearson method to examine associations between pre-post intervention changes (Δ score) in behavioural variables and corresponding changes in alpha activity. The changes in behavioural scores have been calculated to make a positive Δ score value correspond to a positive change (increased dispositional mindfulness, increase interoceptive sensibility, decreased state and trait anxiety). A significance threshold of R > 0.4 was set to focus on relatively strong correlations.

Positive correlations suggest concurrent changes in self-report measures and alpha activity, while negative correlations indicate potential inverse relationships. The use of Δ scores provides a more immediate understanding of the intervention's impact on both behavioural and neuroelectric indices.

4. RESULTS

4.1 Sociodemographic and Descriptive statistics

A total of 89 subjects were initially enrolled in the study and randomly assigned to either the intervention group (N=46) or the control group (N=43). In the intervention group, 16 subjects were subsequently excluded, with 2 not meeting the inclusion criteria and 14 voluntarily dropping out. In the control group, 10 subjects were subsequently excluded, with 3 not meeting the inclusion criteria

and 7 voluntarily dropping out. A total of 63 subjects were eligible for the analysis (N=30 in the intervention group, N=33 in the control group). A total of 11 subjects (N=7 in the intervention group, N=4 in the control group) did not complete any of the post-intervention measures and were excluded from the analysis. Two subjects (one in the intervention group and one in the control group) were excluded because of bad quality of the EEG data in at least one timepoint. The final sample consisted in 50 subjects (22 subjects in the intervention group and 28 subjects in the control group) (**Figure 9**) with a mean age of 22 years old, ranging from 18 to 30 years old. Around the 60% of the total sample were female, with similar gender distribution among the two group. The compliance with the intervention was relatively poor, with 24% of the subjects in the intervention group which attended no more than the half of the meditation classes, and only 28% of the subjects attended at least 75% of the meditation classes.



CONSORT 2010 Flow Diagram

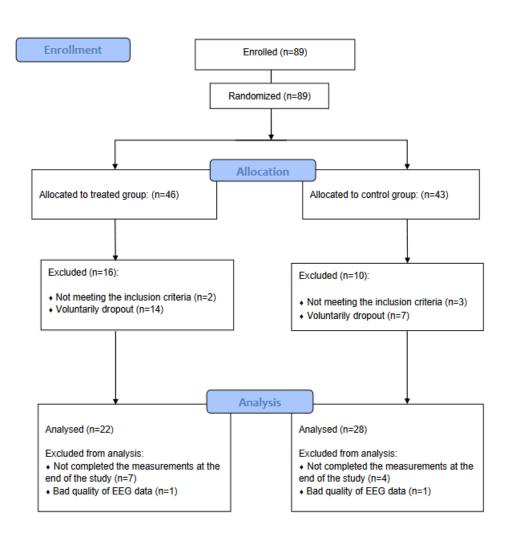


Figure 9: Participant Flow Diagram

In **Table 1** is presented an overview of sociodemographic variables collected at baseline (t0) for both the intervention and control groups. Age is presented as a mean with the standard deviation, while other categorical variables are presented as frequencies and percentages within each group. To assess differences in sociodemographic variables between the two groups, statistical comparisons were performed. Since the continuous variable 'Age' was not normally distributed, the Wilcoxon test was employed to compare the two groups. For the categorical variables, the Chisquared test was utilized when the expected frequencies in each cell were greater than five while for the variables with expected frequencies less than five, the Fisher's exact test was employed.

Variables	Mean (SD) controls	Mean (SD)	p-value ^a
		intervention	
Age	21.6 (2.7)	22.4 (2.8)	0.27
	N (%) controls	N (%) intervention	
Sex			
Female	17 (61%)	14 (63%)	0.83
Male	11 (39%)	8 (37%)	
Nationality			
Italian	28 (100%)	20 (91%)	0.47
Non-Italian	0 (0%)	2 (9%)	
Dominant hand			
Right	25 (89%)	18 (82%)	0.68
Left	3 (11%)	4 (18%)	
Student			
Yes	25 (89%)	20 (91%)	1
No	3 (11%)	2 (9%)	
Degree field			
Humanistic	5 (18%)	5 (23%)	0.74
Scientific	19 (68%)	15 (68%)	
Na	4 (14%)	3 (14%)	
Compliance with exams			
Yes	22 (79%)	15 (68%)	0.61
No, and it is a problem	1 (4%)	2 (9%)	
No, and it is not a problem	2 (7%)	3 (14%)	
Na	3 (11%)	2 (9%)	
Work			
No	21 (75%)	15 (68%)	0.80
Few days a week	4 (14%)	4 (18%)	
Every day (part-time)	0 (%)	1 (4%)	
Every day (full-time)	3 (11%)	2 (9%)	
Housing situation			
I live with my partner	1 (4%)	3 (14%)	0.28
I live with my family	7 (25%)	2 (9%)	
I live alone	4 (14%)	1 (4%)	
I live in a shared flat	10 (36%)	12 (54%)	
I live in a university college	6 (21%)	4 (18%)	

 Table 1. Baseline characteristics of the analyzed sample (controls=28, intervention=22)

Sport activities			
Very few/No	8 (29%)	6 (27%)	0.99
Yes, but irregularly	11 (39%)	9 (41%)	
Yes, regularly	9 (32%)	7 (32%)	
Member of a cultural/sportive			
association			
No	14 (50%)	9 (41%)	0.52
Yes	14 (50%)	13 (59%)	
Books read in a year			
0-1	6 (21%)	4 (18%)	0.70
2-5	14 (50%)	9 (41%)	
5+	8 (29%)	9 (41%)	
Previous meditation experience			
No	17 (61%)	10 (45%)	0.28
Yes	11 (39%)	12 (54%)	
Diet			
None in particular	22 (79%)	16 (73%)	0.41
Vegetarian / vegan / nutritionist	6 (21%)	4 (18%)	
Restricted because of intolerances	0 (0%)	2 (9%)	
Sexual life			
Mostly unsatisfying	12 (43%)	8 (36%)	0.73
Mostly satisfying	13 (46%)	12 (54%)	
I prefer not to answer	3 (11%)	2 (9%)	
Regular sleep			
Mostly no	7 (25%)	7 (32%)	0.59
Mostly yes	21 (75%)	15 (68%)	
Frequent use of alcohol			
No	22 (79%)	17 (77%)	0.91
Yes	6 (21%)	5 (23%)	
Smoker			
No	21 (75%)	16 (73%)	0.86
Yes	7 (25%)	6 (27%)	

^a p-value for between groups comparison.

In **Table 2**, descriptive statistics for each variable at baseline (t0) and post-intervention (t1) are presented separately for the control and intervention groups. To assess the assumption of normality and equal variance distribution at baseline, Shapiro-Wilk tests and variance tests were conducted for each variable. Subsequently, when both normality and equal variance assumptions were met, a two-sample t-test was employed to compare means between the control and intervention groups. In the cases where the normality assumption was violated, the non-parametric Wilcoxon rank-sum test was employed. In the Table 2 are reported the p-value of the comparison between treated and control at the baseline levels. The assessment revealed no statistically significant differences between the control and intervention groups at baseline, indicating that the two groups are sampled from the same population. In the Table 2 are also reported for each the questionnaire (FFMQ, MAIA, STAI X-1 and STAI X-2) and their respective subscales the internal consistency values, measured using Cronbach's alpha, at two time points—baseline (t0) and post-intervention (t1). Cronbach's alpha is a statistic that assesses the internal consistency of a set of items or variables within a measurement scale. It quantifies the extent to which the items in a scale are correlated,

providing an indication of the scale's overall reliability. Higher values of Cronbach's alpha, closer to 1, suggest greater reliability and consistency among the items in the scale. Conversely, lower values may indicate a poor internal consistency. A Cronbach's alpha of 0.70 and above is generally considered good. The Cronbach's alpha of the questionnaires and their subscales used in this study range generally from acceptable to very good, except for the subscales of MAIA: *Not-distracting,* which has poor internal consistency values both in t0 (0.41) and t1 (0.35), and *Self-regulation* which has a poor internal consistency value only in t0 (0.55). Anyway, it should be considered that the Cronbach's alpha values is strongly affected by the number of item and both those two MAIA's subscales has low number of items (item 3 and 4 respectively).

Table 2. Mean, Standard Deviation, and Internal Consistency (Cronbach's Alpha) Values for Questionnaires and Subscales in Two Groups Across Both Time Points (t0 and t1). The table also reports the p-value of the t-test, or the Wilcoxon rank-sum test if the assumption of normality is violated, to compare the baseline (t0) measures between intervention and control group.

Measures	Mean (SD) controls t0	Mean (SD) intervention t0	p-value	Mean (SD) controls t1	Mean (SD) intervention t1	Internal consistency t0	Internal consistency t1
FFMQ							
All Items	119.7 (15.9)	118.3 (20.1)	0.79	121.6 (19.6)	124.6 (19.7)	0.88	0.91
Observing	25.8 (5)	25.6 (5.2)	0.90	26.9 (4.9)	28.5 (5.24)	0.69	0.76
Describing	26.1 (6.4)	25.8 (6.2)	0.47	26.2 (7.8)	25.7 (6.4)	0.91	0.94
Acting with awareness	25.1 (6.4)	24 (4.9)	0.55	24.1 (5.9)	23.9 (5.6)	0.87	0.87
Non judging of inner experience	24.2 (7.3)	24.5 (7.8)	0.87	24.6 (8.5)	25.4 (7.7)	0.92	0.94
Non reactivity to inner experience	18.1 (4.3)	19.1 (4.8)	0.55	19.3 (4.5)	21.1 (4)	0.81	0.86
MAIA							
All Items	2.52 (0.5)	2.52 (0.62)	0.97	2.56 (0.57)	2.92 (0.51)	0.85	0.88
Noticing	3.07 (0.88)	2.98 (0.98)	0.73	3 (0.89)	3.35 (0.84)	0.65	0.62
Not-distracting	2.52 (0.75)	2.49 (0.89)	0.91	2.49 (0.74)	2.34 (0.79)	0.41	0.35
Non-worrying	2.13 (1.16)	2.57 (1.06)	0.19	2.24 (1.27)	2.68 (0.94)	0.76	0.82
Attention regulation	2.42 (0.89)	2.34(1)	0.77	2.42 (0.82)	2.79 (0.82)	0.86	0.83
Emotional awareness	3.21 (1.01)	3.06 (1.16)	0.64	3.18 (0.94)	3.29 (1.15)	0.82	0.86
Self-regulation	2.03 (0.77)	2.27 (0.81)	0.30	2.03 (0.95)	3.19 (0.79)	0.55	0.83
Body listening	2.02 (0.99)	1.94 (1.03)	0.61	2.18 (1.19)	2.55 (1.18)	0.78	0.80
Trusting	2.35 (1.27)	2.27 (0.93)	0.82	2.78 (1.43)	2.85 (1.07)	0.87	0.91
STAI X-1							
All Items	41.4 (10.1)	40.9 (7.3)	0.99	42 (10.3)	39 (8)	0.91	0.92
STAI X-2							
All Items	46.3 (9.7)	46.2 (9.8)	0.98	45.8 (10.7)	45.2 (9.7)	0.88	0.91
HBTT							
All Items	0.58 (0.21)	0.54 (0.23)	0.54	0.58 (0.23)	0.61 (0.29)	-	-
Individual alpha peak power*							
Global	2.69 (1.7)	2.95 (1.51)	0.50	2.63 (1.7)	3.06 (1.24)	-	-
Individual alpha peak frequency							
Global	9.93 (1.1)	10 (1.09)	0.81	10.07 (0.85)	10.07 (0.95)	-	-
Alpha relative power							
Global	37.71 (20.5)	36.8 (20.35)	0.87	34.1 (17.4)	42 (20.8)	-	-

4.2 Hypotheses testing

To explore the effect of our intervention, we conducted a series of hypotheses testing analysis on the behavioural measures, including questionnaires (FFMQ, MAIA, STAIX-1, STAIX-2) and the HBBT task, and on the EEG alpha activity indexes, including IAP power, IAP frequency and alpha relative power. In our statistical models, the behavioural and EEG indexes measured after the intervention (t1) represent the *y term* (dependent variable). The intervention factor (intervention group and control group) and the baseline (t0) scores of the dependent variable represent the *x term* in the models (independent variables). Relevant covariates 'Sex' and 'Previous meditation experiences' have been added to the model to adjust the effect of the independent variable on the dependent variable.

At first, we tested if the effect of the intervention changes across the different level of the baseline scores of the dependent variable (section 4.2.2, Table 3), using linear regression models with interaction term. The formula is the following:

 $y = \beta 0 + \beta 1$ intervention + $\beta 2$ baseline score + $\beta 3$ (intervention × baseline score)+ $\sum \beta$ covariates + ϵ

whereas the $\beta 0$ is the intercept, $\beta 1$ and $\beta 2$ represent the regression coefficient of the independent variables, $\beta 3$ represent the coefficient for the interaction between the dependent variables, $\sum \beta$ covariates represent the coefficient for each of the covariate included in the model and ϵ represents the error term. The $\beta 3$ quantifies the effect of the intervention at different levels of the baseline score, and particularly the changes of the intervention effect for each unitary increase in the baseline score. In this case, we report the results of the model only if the p-values for the interaction is significant (≤ 0.05).

Where the interaction between the two independent variables was not significant, we removed this interaction from the linear models, to investigate the main effect of the intervention factor (section 4.2.3, Table 4):

 $y = \beta 0 + \beta 1$ intervention $+ \beta 2$ baseline score $+ \sum \beta$ covariates $+\epsilon$

In this case, we report the values of $\beta 1$ and its relative p-values for each model we tested. The $\beta 1$ represent the effect of the intervention on the dependent variable, adjusting for the baseline score and the other covariates. The $\beta 1$ quantifies the amount of change that the intervention induced on the behavioural scores or on the EEG indexes.

In the following sections, we will discuss the significant findings based on the assessment of unadjusted p-values, using a threshold of 0.05. Both adjusted and unadjusted p-values are presented in the respective tables, with the adjusted values calculated using the Benjamini-Hochberg method to control the false discovery rate (FDR). We set the FDR threshold at 0.10 for the adjusted p-values. Results that fall below this threshold are considered statistically significant. The significance of the results remains consistent whether assessed with unadjusted or adjusted p-values.

4.2.1 Analysis of Group Differences Using Linear Regression Model with Interaction Term

In **Table 3** are reported only the significant results of linear regression analyses conducted to assess the intervention effect at different levels of baseline (t0) scores, adjusting for relevant covariates, which are sex and previous meditation experiences. The models included an interaction term between the intervention factor and the baseline score, allowing for an examination of how the intervention effect varied with individuals' pre-intervention levels. The models were appropriately weighted for the number of attended meditation classes, considering compliance with the intervention among subjects in the intervention group.

The examination of the global value of IAP power revealed a significant intervention*t0 scores interaction effect (β =-0.40, p=0.02), suggesting that the impact of the intervention on alpha peak power is not uniform across all participants; instead, it varies according to the individuals' baseline (t0) scores. A negative coefficient β indicates that the effect of the intervention is reduced along with a baseline score increase. Participants with higher levels of baseline score in the IAP power had have a smaller intervention effect.

In addition, R squared values were calculated for each model. R squared, also known as the coefficient of determination, represents the proportion of variance in the dependent variable that is explained by the independent variables included in the model. A higher R squared value indicates a better fit of the model to the observed data, suggesting that the included variables collectively account for a larger proportion of the variability in the outcome. The presentation of R squared values provides insight into the overall explanatory power of the models and their ability to capture the variance in the dependent variables, enhancing the interpretation of the reported findings. The dependent variables tested using linear regression model with interaction term which did not have a significant interaction effect have been tested with the same model but removing the interaction term, and the results are reported in the next section.

Table 3. Analysis of Group Differences Using Linear Regression Model with Interaction Term. Each measure, representing the dependent variable in the linear regression model, is reported in the first column. The β regression coefficient for the interaction between the intervention factor and the baseline (t0) score is reported in the second column. This coefficient indicates the magnitude and direction of the linear relationship between the interaction of the two factors with the dependent variable. The corresponding p-value indicates the statistical significance of this relationship is reported in the third column. The adjusted p-value obtained with the Benjamini-Hochberg method for the false discovery rate fixing a false discovery rate equal to 0.10 is reported in fourth column. R-squared values are reported in the last column, indicating the goodness of fit of the model.

Measure	β intervention*t0 score	p-value	adjusted p- value	R-squared
Individual alpha peak power				
Global	-0.40	0.02	0.07	0.74
In this table are reported only the models in which the p-value of the interaction was statistically				

In this table are reported only the models in which the p-value of the interaction was statistically significant ($p \le 0.05$). All models are adjusted for sex, for previous meditation experience, baseline level of the variable (t0 scores) and group. The models are weighted for the number of meditation classes attended by each subject.

4.2.2 Analysis of Group Differences Using Linear Regression Model without Interaction Term

In **Table 4**, the outcomes of our linear regression analyses examining the impact of intervention factor on behavioural and EEG measures, adjusting for sex and previous meditation experiences, are reported. The β regression coefficient indicates the magnitude and direction of the linear relationship between the intervention factor and each dependent variable, while the p-value indicates the significance of the linear relation between the dependent variable and the intervention, with a threshold for significance set to ≤ 0.05 . The models were appropriately weighted for the number of attended meditation classes, considering compliance with the intervention among subjects in the intervention group.

For behavioural measures assessed through self-report scales and the HBTT, significant intervention effects were observed in the total scores of FFMQ and MAIA, as well as their specific sub-domains. Notably, a statistically significant relation with the intervention factor was found for the FFMQ global score (β =7.06, p=0.03) and the *Non-reactivity to Inner Experience subscale* (β =2.2, p=0.005), indicating a beneficial intervention effect in the dispositional mindfulness domain. Similarly, the MAIA measure revealed a statistically significant association of the intervention factor with the global score (β =0.39, p=0.001), *Attention Regulation score* (β =0.45, p=0.02), and *Self-regulation score* (β =1.09, p<0.001); these results signifies that there was an increase in the interoceptive sensibility in the intervention group. No statistically significant intervention effects were detected for STAI X-1, STAI X-2 measures, and HBTT.

The examination of global IAP Frequency revealed a minimal effect of the intervention, with a negligible coefficient of -0.01 (p=0.95), indicating no substantial post-intervention shift in the IAP frequency. Although the relation between the intervention factor and the alpha relative power (β =7.77, p=0.13) did not reach conventional levels of statistical significance, this result suggests a potential increase in alpha relative power post-intervention.

The R squared values are also reported for each of the model we tested.

Table 4. Analysis of Group Differences Using Linear Regression Model without Interaction Term. Each measure, representing the dependent variable in the linear regression model, is reported in the first column. The β regression coefficient for the intervention factor is reported in the second column. This coefficient indicates the magnitude and direction of the linear relationship between the intervention and the dependent variable. The corresponding p-value indicates the statistical significance of this relationship is reported in the third column. The adjusted p-value obtained with the Benjamini-Hochberg method for the false discovery rate fixing a false discovery rate equal to 0.10 is reported in fourth column. The R-squared values are reported in the last column, indicating the goodness of fit of the model.

Measure	β intervention	p-value	adjusted p- value	R-squared
FFMQ				
All Items	7.06	0.03	0.08	0.71
Observing	1.93	0.07	0.18	0.61
Describing	1.67	0.21	0.33	0.7
Acting with awareness	0.93	0.43	0.53	0.65
Non judging of inner experience	0.78	0.64	0.77	0.6
Non reactivity to inner experience	2.2	0.005	0.03	0.61
MAIA				
All Items	0.39	0.001	0.01	0.6

Noticing	0.33	0.13	0.27	0.43
Non-distracting	0.01	0.95	0.95	0.15
Non-worrying	0.51	0.1	0.24	0.38
Attention regulation	0.45	0.02	0.06	0.53
Emotional awareness	0.01	0.75	0.78	0.27
Self-regulation	1.09	< 0.001	< 0.001	0.65
Body listening	0.36	0.2	0.33	0.41
Trusting	0.25	0.39	0.53	0.44
STALX-1				
All Items	-2.93	0.31	0.53	0.13
STAI X-2				
All Items	-2.17	0.28	0.42	0.6
HBTT				
Total score	0.02	0.73	0.78	0.42
Individual alpha peak frequency				
Global	-0.01	0.95	0.95	0.51
Alpha relative power				
Global	7.77	0.13	0.27	0.34

*P-values ≤ 0.05 are considered as statistically significant. All models are adjusted for sex, for previous meditation experience, baseline level of the variable (t0 scores) and group. The models are weighted for the number of meditation classes attended by each subject.

4.2.3 Analysis of Group Differences Using Linear Mixed Model with Interaction Term across Different Brain Regions with Pooled Electrodes Data

To test the effect of the intervention at different levels of baseline values among different brain regions, we conducted an exploratory analysis using Linear Mixed Models [122]. The data of the 17 EEG channel have been pooled into three brain regions, representing the two hemispheres and the midline, according to the following classification:

- Left hemisphere electrodes: F3, F7, C3, T3, T5, P3, O1;
- Right hemisphere electrodes: F4, F8, C4, T4, T6, P4, O2;
- Midline electrodes: Fz, Pz, Cz.

For each subject, the data from each of the electrodes represent an observation, therefore each subject will have as many observations as the number of the electrodes in the brain region of interest. In each model, we included as dependent variable the post-intervention values of EEG alpha activity index of interest (IAP power, IAP frequency, alpha relative power). The intervention factor and the baseline values were used as the independent variables, also including an interaction term between the intervention factor and the baseline score. Each model has been adjusted for relevant covariates, which are sex, previous meditation experiences and dominant hand. A random intercept for subjects in the form of 1|subject had been used to adjust the models for intra-subject variability produced by the repeated measurements (different electrodes) on the same subject. In Table 5 the results of the analysis across different brain regions are reported. As for the Left Hemisphere, we found a significant effect of the interaction between intervention and baseline values for the IAP power (β =-0.18, p=0.003), indicating that the effect of the intervention varies at different levels of the baseline IAP power levels, with an increase in the baseline IAP power associated with a reduction of the intervention effect. In the Right Hemisphere, we found a significant effect of the interaction between intervention and baseline values for the IAP power (β =-0.16, p=0.01) and for the IAP frequency (β =0.31, p=0.001). These findings indicate that an increase in baseline IAP power is associated with a reduction in the intervention effect on postintervention IAP power. Conversely, an increase in baseline IAP frequency is associated with an amplification of the intervention effect on post-intervention IAP frequency. Lastly, among the electrodes placed on the Midline, we observed a significant effect of the interaction between intervention and baseline values for the alpha relative power ($\beta = 0.36$, p=0.01). These findings indicate that an increase in baseline alpha relative power is associated with an increase in the intervention effect on post-intervention alpha relative power among the electrodes placed on the Midline.

Table 5. Analysis of Group Differences Using Linear Mixed Model with Interaction Term across Different Brain Regions with Pooled Electrodes Data. The data from all the electrodes were pooled in three different brain regions according to the hemispherical division, indicated in the first column. Each post-intervention measure, representing the dependent variable in the linear mixed model, is reported in the second column. The β regression coefficient for the interaction between the intervention factor and the baseline (t0) score is reported in the third column. This coefficient indicates the magnitude and direction of the linear relationship between the interaction of the two factors with the dependent variable. The corresponding p-value indicates the statistical significance of this relationship is reported in the third column. The adjusted p-value obtained with the Benjamini-Hochberg method for the false discovery rate fixing a false discovery rate equal to 0.10 is reported in fourth column.

Brain region	Measure	β intervention *t0 score	p-value	adjusted p-value
Left hemisphere	Individual alpha peak power	-0.18	0.003	0.02
Right hemisphere	Individual alpha peak power	-0.16	0.01	0.05
Midline	Individual alpha peak power	-0.04	0.71	0.79
Left hemisphere	Individual alpha peak frequency	-0.18	0.06	0.15
Right hemisphere	Individual alpha peak frequency	0.31	0.001	0.01
Midline	Individual alpha peak frequency	-0.18	0.17	0.32
Left hemisphere	Alpha relative power	0.11	0.18	0.32
Right hemisphere	Alpha relative power	-0.04	0.59	0.74
Midline	Alpha relative power	0.36	0.01	0.06

*P-values≤0.05 are considered as statistically significant All models are adjusted for sex, for previous meditation experience, dominant hand, baseline level of the variable (pre-test value) and group. For each model, a random intercept for subjects in the form of 1|subject had been used to adjust the models for intra-subject variability produced by the data from multiple electrodes measured within the same subject.

4.3 Correlation analysis

4.3.1 Partial Correlations Between Behavioural Measures and EEG Indexes among the Intervention Group.

In **Table 6** we present the results for the intervention group of partial correlations assessing the relationships between changes in behavioural measures and alterations in Individual IAP power, IAP frequency and alpha relative power as a global index and for each electrode within the intervention group. The partial correlation analysis accounts for the covariates sex and previous meditation experiences, and the partial correlation with Pearson method is used to explore associations between the pre-post intervention changes (Δ score) in behavioural variables and corresponding changes in alpha activity. The correlation coefficient R quantifies the strength and direction of the relationship between variables. A positive coefficient indicates a direct association, while a negative coefficient suggests an inverse association. In the Table 6 we reported only the correlation with a R coefficient >0.4 between behavioural and EEG measures. A threshold of 0.4 ensures that only relatively strong correlations are reported in the table.

Global and by channel Individual Alpha Peak Power: The partial correlation analysis investigating the relationship between the FFMQ questionnaire and IAP power revealed relatively strong associations. A substantial positive correlation was observed between changes in the FFMQ total score and corresponding fluctuations in IAP power at electrode F3 (R=0.43), C4 (R=0.41) and T3 (R=0.42). As regard to the FFMQ subscales, we found: a positive correlation between Observing and T6 (R=0.50); positive correlations between Non Judging of Inner Experience and C3 (R=0.45) and T6 (R=0.50) and negative correlation with Fz (R=-0.40); positive correlations between Non Reactivity of Inner Experience and F3 (R=0.42) and Pz (R=0.42) and a negative correlation with Fz (R=-0.42). From the correlation analysis emerged also that changes in different subscales of the MAIA are associated with changes in the IAP power in several electrodes. We found a relatively strong positive correlation between the pre-post difference between the whole MAIA scale and the electrodes P3 (R=0.45) and P4 (R=0.5). Also, the changes in the MAIA subdomains were found relatively strongly associated with changes in several electrodes. The Non-distracting subscale has a positive correlation with the P3 (R=0.57) and with the electrode P4 (R=0.50). We found relatively strong correlation between Attention Regulation subscale and the global IAP power (R=0.43) and with the F3 (R=0.42), F4 (R=0.44), P3 (R=0.46) P4 (R=0.59) and Pz (R=0.46), between the Self-Regulation subscale and the IAP power in F4 (R=0.41), T3 (R=0.46) and T6 (0.53). Notably, a negative correlation has been found between MAIA subscales Body Listening and Trusting with the electrode Cz (R=-0.52 and -0.50 respectively). These significant correlations highlight the nuanced associations between specific aspects of mindfulness, as measured by the FFMO and MAIA questionnaires, and alpha peak power at distinct EEG electrodes. Positive correlations suggest a concurrent increase in the pre-post changes for the self-report measures and the pre-post changes for the IAFF while negative correlations indicate a potential inverse relationship. Lastly, the changes in STAI X-2 scales correlates negatively with changes in C4 (R=-0.47). The full correlation matrix is reported in Figure 10.

Global and by channel Individual Alpha Peak Frequency: The partial correlation analysis investigating the relationship between the changes in FFMQ questionnaire and the changes in IAP frequency revealed relatively strong associations. The changes total FFMQ scale has a positive correlation with the shift in IAP frequency in the electrode P3 (R=0.50). The *Observing* subscale has negative correlation with the electrodes Cz (R=-0.47) and O2 (R=-0.54). The *Describing* subscale has a positive correlation with the electrode P3 (R=0.46), the subscale *Acting with awareness* with F7 (R=0.41) and T6 (R=0.42) and the subscale *Non reactivity to inner experience* with Fz (0.47). The differences in the MAIA global score and in its subscale *Body Listening* and *Trusting* both positively correlating with the global IAP frequency (R=0.44 and 0.5 respectively) and with several electrodes. Notably, the changes in the scores relative to the interoceptive accuracy, measured with the HBTT, negatively correlate with the changes in the global IAP frequency (R=-60) and with multiple electrodes, particularly those placed over the temporal and occipital regions of the scalp. The full correlation matrix is reported in **Figure 11**.

Global and by channel Alpha Relative Power: The major findings from the partial correlation analysis indicate a notably robust correlation between the shift in alpha relative power and changes in FFMQ and MAIA scores. Substantial negative correlations were identified between the FFMQ total score and Cz (R=-0.47), T3 (R=-0.41), T4 (R=-0.43) and O2 (R=-0.41). The changes in MAIA total score have a positive correlation with F3 (R=0.40), the subscale *Noticing* with Pz (R=0.43) and T6 (R=0.48), the subscale *Attention regulation* with F3 (R=0.51), F4 (R=0.51) and T3 (0.42). Lastly, the MAIA subscale *Non-worrying* has negative correlation with P3 (R=-0.62) and O1 (R=-0.48). The full correlation matrix is reported in **Figure 12**.

Table 6. Partial Correlations Between Behavioral Measures and EEG Indexes among the Intervention Group. This table presents the results of partial correlation analyses conducted within the intervention group, examining the association between changes in behavioral measures and alterations in alpha band activity (Δ score) following the intervention. The partial correlations were adjusted for covariates including 'sex' and 'previous meditation experiences.' The strength and direction of the correlations are quantified by the R coefficient.

Measure	Individual alpha peak power	R coefficient
(A score)	(Δ score)	
FFMQ		
All items	F3	0.43
All items	C4	0.41
All items	Т3	0.42
Observing	Τ6	0.50
Non judging of inner experience	Fz	-0.40
Non judging of inner experience	C3	0.45
Non judging of inner experience	Τ6	0.50
Non reactivity to inner experience	F3	0.42
Non reactivity to inner experience	Fz	-0.42
Non reactivity to inner experience	Pz	0.42
MAIA		
All Items	Р3	0.45
All Items	P4	0.50
Non-distracting	Р3	0.57
Non-distracting	P4	0.50
Attention regulation	Global	0.43
Attention regulation	F3	0.42
Attention regulation	F4	0.44
Attention regulation	Р3	0.46
Attention regulation	P4	0.59
Attention regulation	Pz	0.46
Self-regulation	F4	0.41
Self-regulation	Т3	0.46
Self-regulation	T6	0.53
Body listening	Cz	-0.52
Trusting	Cz	-0.50
STAIX2		
All items	C4	-0.47
Measure	Individual alpha peak frequency	R coefficient
(Δ score)	(Δ score)	
FFMQ	· · · · · · · · · · · · · · · · · · ·	
All Items	Р3	0.50
Observing	Cz	-0.47
Observing	02	-0.54
Describing	Р3	0.46

Acting with awareness	F7	0.41
Acting with awareness	T6	0.42
Non reactivity to inner experience	Fz	0.47
MAIA		
All Items	O2	0.49
Emotional awareness	O2	0.44
Body listening	Global	0.44
Body listening	Fz	0.65
Body listening	Cz	0.52
Body listening	P4	0.44
Body listening	Pz	0.43
Body listening	Т3	0.56
Body listening	T4	0.51
Body listening	O1	0.44
Body listening	02	0.77
Trusting	Global	0.50
Trusting	F8	0.43
Trusting	C3	0.63
Trusting	Cz	0.43
Trusting	P3	0.43
Trusting	P4	0.50
Trusting	Pz	0.63
Trusting	T3	0.42
Trusting	T4	0.56
Trusting	T5	0.48
Trusting	T6	0.46
Trusting	01	0.46
Trusting	O2	0.61
HBTT		
Total score	Global	-0.60
Total score	Pz	-0.65
Total score	T3	-0.42
Total score	T4	-0.45
Total score	T5	-0.55
Total score	01	-0.50
Total score	O2	-0.60
Measure	Alpha relative power	R coefficient
$(\Delta \text{ score})$	(Δ score)	K coefficient
FFMQ		
All items	Cz	-0.47
All items	T3	-0.41
All items	T4	-0.43
All items	02	-0.41
MAIA	02	0.71
All items	F3	0.40
Noticing	Pz	0.43
Noticing	T6	0.43
Non-worrying	P3	-0.62
Non-worrying	01	-0.48
Attention regulation	F3	-0.48 0.51
Attention regulation	F3 F4	0.51
Attention regulation	T3	0.42
raumon regulation	15	0.42

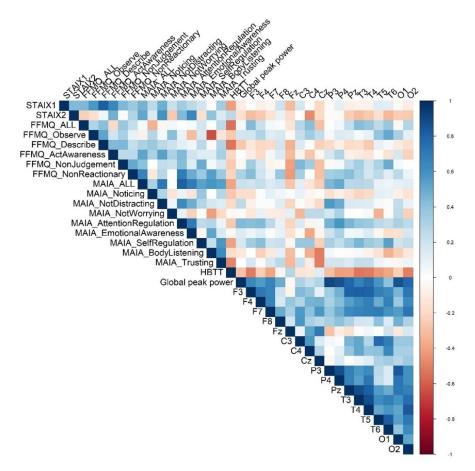


Figure 10. Partial Correlation Matrix: Behavioral Variables and IAP Power in the Intervention Group.

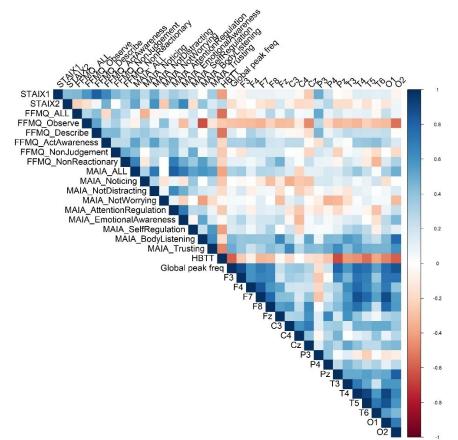


Figure 11. Partial Correlation Matrix: Behavioral Variables and IAP Frequency in the Intervention Group.

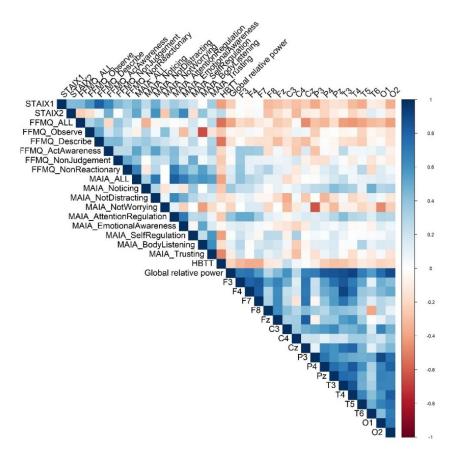


Figure 12. Partial Correlation Matrix: Behavioral Variables and Alpha Relative Power in the Intervention Group.

4.3.2 Partial Correlations Between Behavioural Measures and EEG Indexes among the Control Group.

In **Table 7** we present the results for the control group of partial correlations assessing the relationships between changes in behavioural measures and alterations in Individual IAP power, IAP frequency and alpha relative power as a global index and for each electrode within the intervention group. The partial correlation analysis accounts for the covariates sex and previous meditation experiences, and the partial correlation with Pearson method is used to explore associations between the pre-post intervention changes (Δ score) in behavioural variables and corresponding changes in alpha activity. The correlation coefficient R quantifies the strength and direction of the relationship between variables. A positive coefficient indicates a direct association, while a negative coefficient suggests an inverse association. In the Table 5 we reported only the correlation with a R coefficient > 0.4 between behavioural and EEG measures. A threshold of 0.4 ensures that only relatively strong correlations are reported in the table.

Global and by channel Individual Alpha Peak Power: The partial correlation analysis investigating the relationship between the FFMQ questionnaire and IAP power revealed relatively

strong associations. Negative correlations were observed between changes in the FFMQ total score and corresponding fluctuations in IAP power at electrode P3 (R=-0.42), O1 (R=-0.49) and O2 (R=-0.57). As regard to the FFMQ subscales, we found a positive correlation between *Observing* and the Cz (R=0.41), Pz (R=0.55), T3 (R=0.42) and T4 (R=0.48). From the correlation analysis emerged also that changes in different subscales of the MAIA are negatively associated with changes in the IAP power in several electrodes. We found a relatively strong negative correlation between the prepost difference between the MAIA subdomains *Non-distracting* and electrodes F8 (R=-0.52) and C3 (R=-0.51), between *Emotional awareness* and electrodes P3 (R=-0.57), and P4 (R=-0.50), between *Self-regulation* and electrodes Pz (R=-0.45), T3 (R=-0.41) and O1 (R=-0.49), between *Body listening* and electrodes Pz (R=-0.47), T4 (R=-0.50) and O1 (R=-0.49). The full correlation matrix is reported in **Figure 13**.

Global and by channel Individual Alpha Peak Frequency: The partial correlation analysis investigating the relationship between the changes in FFMQ questionnaire and the changes in IAP frequency revealed relatively strong associations. The changes in the FFMQ sub-domain *Describing* have a positive correlation with the electrode F3 (R=0.42), F7 (R=0.47) and F8 (R=0.54) and Cz (R=0.49) while *Non judging of inner experience* negatively correlates with Pz (R=-0.51). The differences in the MAIA scores showed bidirectional correlations with the changes in the IAP frequency; the total score correlate positively with P3 (R=0.49) while negatively with P4 (R=-0.42) and O1 (R=-0.70). The subscale *Noticing* correlates positively with C3 (R=0.50) and T3 (R=0.45); T3 correlates positively also with *Attention regulation*. On the other hand, negative correlations have been found between *Not-distracting* and P3 (R=-0.44), *Non-worrying* and C3 (R=-0.41), *Emotional awareness* and F8 (R=-0.44) and *Trusting* and O1 (R=-0.56). Positive correlations have been found also between the anxiety scores and IAP frequency, in particular STAI X-1 with F8 (R=0.41) and STAI X-2 with P3 (R=0.50). The full correlation matrix is reported in **Figure 14**.

Global and by channel Alpha Relative Power: The partial correlation analysis indicates several negative correlations between the shift in alpha relative power and changes in FFMQ, MAIA and STAI X-1 scores. Substantial negative correlations were identified between the FFMQ total score and F4 (R=-0.40), F7 (R=-0.47), T6 (R=-0.44) and P3 (R=-0.42). The changes in MAIA *Noticing* were found to negatively correlate with P3 (R=-0.43) and P4 (R=-0.45), while the subscale *Body listening* showed negative correlations with the global alpha relative power (R=-0.43) and with several electrodes. Lastly, STAI X-1 changes negatively correlated with T4 (R=-0.42), T5 (R=-0.40), O1 (R=-0.43) and O2 (R=-0.40). The full correlation matrix is reported in **Figure 15**.

Table 7. Partial Correlations Between Behavioral Measures and EEG Indexes among the Control Group. This table presents the results of partial correlation analyses conducted within the intervention group, examining the association between changes in behavioral measures and alterations in alpha band activity (Δ score) following the intervention. The partial correlations were adjusted for covariates including 'sex' and 'previous meditation experiences'. The strength and direction of the correlations are quantified by the R coefficient.

Measure	Individual alpha peak power	R coefficient
(Δ score)	(A score)	
FFMQ	D 2	0.42
All items	P3	-0.42
All items	01	-0.49
All items	02	-0.57
Observing	Cz	0.41
Observing	Pz	0.55
Observing	T3	0.42
Observing	T4	0.48
MAIA	F 0	0.52
Non-distracting	F8	-0.52
Non-distracting	C3	-0.51
Emotional awareness	P3	-0.57
Emotional awareness	P4	-0.50
Self-regulation	Pz	-0.45
Self-regulation	T3	-0.41
Self-regulation	01	-0.49
Body listening	Pz	-0.49
Body listening	T3	-0.47
Body listening	T4	-0.50
Body listening	01	-0.49
Measure	Individual alpha peak frequency	R coefficient
(Δ score)	(A score)	
FFMQ		
Describing	F3	0.42
Describing	F7	0.47
Describing	F8	0.54
Describing	Cz	0.49
Non judging of inner experience	Pz	-0.51
MAIA		
All Items	P4	-0.42
All Items	P3	0.49
All Items	O1	-0.70
Noticing	C3	0.50
Noticing	Т3	0.45
Not-distracting	P3	-0.44
Non-worrying	C3	-0.41
Attention regulation	Т3	0.45
Emotional awareness	F8	-0.44
Trusting	O1	-0.56
STAI X1		
Total score	F8	0.41
STAI X2		
Total score	Р3	0.50
Measure	Alpha relative power	R coefficient
(A score)	(A score)	
FFMQ	\$\$	
All items	F4	-0.40
All items	F7	-0.47
All items	Τ6	-0.44
Describing	Р3	-0.42
MAIA		
Noticing	Р3	-0.43
Noticing	P4	-0.45
Body listening	Global	-0.43
Body listening	F3	-0.43
Body listening	F4	-0.48
Body listening	P4	-0.48
, <u>0</u>		

Body listening	Pz	-0.42
Body listening	Τ4	-0.48
Body listening	Т6	-0.46
Body listening	O1	-0.47
STAI X1		
Total score	Τ4	-0.42
Total score	Т5	-0.40
Total score	O1	-0.43
Total score	O2	-0.40

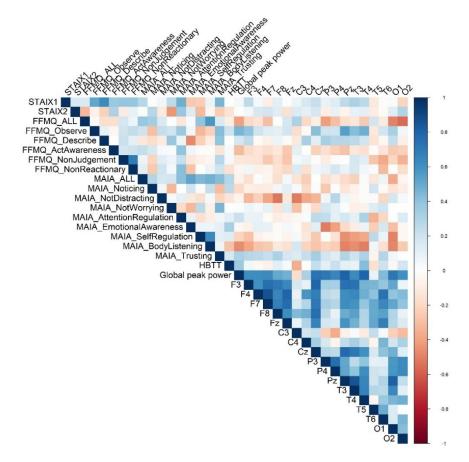


Figure 13. Partial Correlation Matrix: Behavioural Variables and IAP Power in the Control Group.

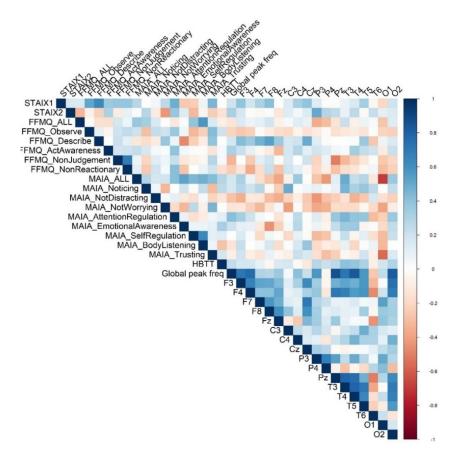


Figure 14. Partial Correlation Matrix: Behavioral Variables and IAP Frequency in the Control Group.

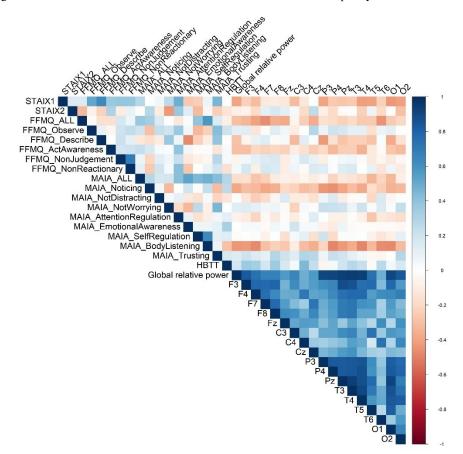


Figure 15. Partial Correlation Matrix: Behavioral Variables and Alpha Relative Power in the Control Group.

5. DISCUSSION

The IM is a novel Mindfulness-Based Intervention developed by our research team in 2017 and tested in a pilot study among healthy adults [117]. Subsequent research endeavours have systematically probed the efficacy of IM across diverse populations, revealing positive outcomes in areas such as well-being, mindfulness, emotion regulation, interoceptive sensibility, state anxiety and others [106], [107], [108], [109], [110]. These investigations, marked by evolving methodologies, collectively contribute to a growing body of evidence affirming the beneficial effects of IM on psychological health.

In this study, we set out to investigate the effectiveness of IM, our mindfulness intervention, among healthy young adults. We recruited voluntary participants in the city of Pavia, Italy. Employing a two-group pretest-posttest experimental design, participants were randomly assigned to either IM intervention group or a waiting list control group. Our a-priori hypotheses were that those in the IM group would demonstrate improvements in mindfulness, interoception, and anxiety compared to the control group. Moreover, we hypothesized that the intervention group would show distinctive changes in brain activity related to the alpha frequency band. In this study we focused on the alpha band activity, given its frequently association with mindfulness, and we examined various measures of alpha activity including the IAP frequency, the IAP power and the alpha relative power. Alpha band amplitude and alpha peak amplitude both assess the magnitude of alpha activity. However, alpha band amplitude is usually employed within a specified alpha frequency range, whereas alpha peak amplitude focuses on spectral features around individual alpha peaks [123].

Another major measure of alpha peak activity is the alpha peak frequency that is the discrete frequency with the highest power value in the alpha oscillation range. The alpha peak frequency is thought to be linked to brain metabolism and serves as an indicator of cognitive preparedness, reflecting the brain's ability for optimal cognitive performance [124]. Both the amplitude and frequency of alpha peak activity exhibit individual variability and have demonstrated associations with diverse cognitive functions and states across studies [124], [125], [126].

The results of the analysis indicated significant enhancements in various psychological domains, particularly dispositional mindfulness and interoceptive sensibility. Notably, dispositional mindfulness, as assessed through the FFMQ, and its Non-reactivity to Inner Experience subscale, showed an increase in the intervention group after the intervention. This means that participants who practiced the IM demonstrated a general increased tendency to be mindful in daily life and a heightened ability to observe internal thoughts and feelings without reacting to them. This is in line with the current evidence from three meta-analyses showing that the scores of self-reported mindfulness measures can noticeably increase in response to mindfulness training received [18], [127], [128].

The increase we observed in interoceptive sensibility scores, measured with the MAIA questionnaire, along with improvements in Attention Regulation and Self-regulation subscales, underscores the influence of IM training on individuals' positive aspects of internal awareness and regulation processes. The enhanced attention regulation suggests that participants who underwent

the mindfulness intervention displayed an improved capacity to sustain and control attention to body sensations. Similarly, the positive shift in the Self-regulation score indicates an increased ability to regulate distress by attention to body sensations. The overall heightened interoceptive sensibility indicates improved perception about one's own internal focus and accuracy in perceiving bodily sensations and this aligns with the core principles of mindfulness, which emphasize presentmoment awareness. Those results are in accordance with the literature regarding effects of mindfulness-based training on interoceptive sensibility [27], [28], [29], [30], [129].

Numerous studies have provided evidence linking interoceptive dysfunction to various conditions such as depression, anxiety disorders, eating disorders, and chronic pain, among others [130], [131], [132], [133]. Interoceptive signals are not inherently categorized as positive or harmful and the way individuals attend to these signals can distinguish between adaptive and maladaptive responses to interoceptive cues [59]. For instance, anxiety may arise from the conditioned perception of body sensations as threatening signals, in comparison to a more mindful or decentred attentional approach [134]. Embracing a mindful attitude toward feelings, sensations, and thoughts, particularly through practices rooted in acceptance, appears to instigate significant changes in coping mechanisms and strategies for emotion regulation [135].

The findings of this study concerning dispositional mindfulness and interoceptive sensibility are notably significant, as they suggest that the cultivation of mindfulness, coupled with enhancements in interoceptive sensibility, reflects a heightened awareness of both mental and bodily signals. This increased awareness is indicative of a more adaptive and attuned response to these signals, underscoring the potential positive impact of mindfulness practices on the integration of mind and body awareness. Moreover, both dispositional mindfulness and interoception are linked to improved functioning and psychological well-being [17], [42], [136], [137].

In contrast to our initial hypotheses, the analysis did not reveal any significant intervention effects on state and trait anxiety scales, as well as the Heartbeat Tracking Task. While mindfulness training is often associated with reductions in anxiety [47], [48], [50], our study suggests that the IM might not be directly associated with improvements in anxiety in a healthy sample, as we did not find statistically significant differences in the pre-post anxiety scores between groups. However, it is essential to highlight that despite the lack of statistical significance, the pre-post mean scores in both state and trait anxiety scales within the intervention group suggest a discernible decrease in both state and trait anxiety; the pre-post intervention differences for state anxiety are smaller in the control group compared to the intervention group while for trait anxiety an increase in the mean score is observed. The lack of significance may be attributed to the specific measures employed, which might not have captured the subtleties of anxiety-related changes. Additionally, the intervention's impact on anxiety may not have clearly emerged in this study due to the small sample size and the lack of compliance with the intervention by many participants, reducing possible effects. In our previous study among medical students, we did not find significant changes in state anxiety after the IM training [110] and we can conclude that IM did not prove to have a direct effect on anxiety among healthy young population, whilst it enhances psychological factors that are inversely associated with anxiety, such as dispositional mindfulness.

The null findings regarding interoceptive accuracy align with findings from other studies. A study [129] revealed that mindfulness training enhanced in interoceptive sensibility but did not show a

corresponding improvement in accuracy. Investigations comparing meditators to non-meditators reported no notable differences in heartbeat perception between the two groups [36], [138]. Contrarily to interoceptive sensibility, there is a general lack of consistent correlation between interoceptive accuracy and mindfulness [31], [32] and the association between interoceptive accuracy and well-being remains inconclusive [23], [139].

In the literature, one of the most consistently observed electro-neurophysiological effects associated with mindfulness meditation involves increases in the amplitude of EEG alpha oscillations during meditation practice, as well as during resting-state condition [68], [70], [140]. The alpha rhythm is recognized for its role in attentional processes, particularly internalized attention and top-down attentional control [141], [142]. Consequently, these findings align with the proposed role of mindfulness meditation in the regulation of attentional function.

Our analysis of neurophysiological measures uncovered a noteworthy interaction effect in the evaluation of the IAP power. The observed significant interaction between the intervention and baseline scores indicates that the intervention's effect is not uniform across all participants. Specifically, the negative coefficient implies that as baseline scores increase, the intervention's effect on alpha peak power diminishes. In practical terms, participants with higher initial levels IAP power exhibited a more subdued response to the intervention, suggesting a potential threshold effect. This finding emphasizes the importance of tailoring mindfulness interventions to individual baseline characteristics, acknowledging that one-size-fits-all approaches may not capture the diverse responses within a participant cohort.

The examination of the IAP frequency did not show a shift post-intervention. The observed changes in alpha relative power despite not reaching conventional levels of statistical significance, hinted at a potential increase. This subtle alteration in brain activity suggests that mindfulness practice may exert influences on alpha oscillations, even if these effects do not manifest prominently. The intricacies of these neural changes led us to perform further exploration to assess the impact of the intervention across the different part of the brain.

The exploratory analysis of alpha activity provided a deeper understanding of the changes across electrodes placed over different brain regions. In the electrodes placed above the left hemisphere, the intervention effect on IAP power varied with baseline levels, with higher baseline power being associated with reduced changes in IAP power. Similarly, in the electrodes placed over the right hemisphere, the changes in IAP power were smaller when the baseline IAP power values were higher. We also found that in the right hemisphere the changes in the IAP frequency varied according to baseline values; in this case a higher baseline IAP frequency was associated with a greater increase in the IAP frequency. Lastly, among the midline electrodes, an increase in baseline alpha relative power was associated with a greater increase in post-intervention alpha relative power compared to pre-intervention levels. These results suggest that the intervention might modulate the alpha frequency band activity differently across brain regions, and that the effect of the intervention is associated with individual characteristics. It emerges that the IM induces fewer changes in IAP power among individuals with higher baseline IAP power, while those with higher baseline IAP frequency and alpha relative power are more likely to see an increase in these measures postintervention. To elucidate the significance of alpha activity changes, we conducted a partial correlation analysis, probing the relationship between shifts in behavioural and neuropsychological

domains. Here, we will discuss the strongest correlations identified in the intervention group, focusing on measures that exhibited statistically significant improvement.

In the intervention group, we observed moderate bidirectional correlations between changes in dispositional mindfulness, interoceptive sensibility, and alterations in alpha activity across multiple electrodes. Changes in dispositional mindfulness demonstrated a trend of positive correlation with the power changes in the IAP frequency, particularly in left-hemisphere electrodes such as F3, C3, and T3. Conversely, the frontal midline electrodes Fz and Cz exhibited a trend of negative correlations with dispositional mindfulness and interoceptive sensibility. Notably, the most prominent positive associations for interoceptive sensibility were found in parietal electrodes, while the sub-domain of attention regulation and self-regulation shows a positive association also with the temporal electrodes.

Regarding changes in IAP frequency, an increase in dispositional mindfulness correlated most strongly with an increase in P3. Notably, the non-reactivity to inner experience domain exhibited a positive correlation with IAP frequency, while demonstrating a negative correlation with IAP power for electrode Fz. Additionally, for interoceptive sensibility, an increase in post-intervention scores was associated with an increase in IAP frequency in the O2 electrode.

For alpha relative power, dispositional mindfulness consistently showed negative correlations with changes in several electrodes, with the strongest correlation found in Cz and in the temporal electrodes. Positive correlations were observed between the frontal electrodes, in particular F3 and F4, and sub-domains of dispositional mindfulness, as well as with interoceptive sensibility and its sub-domain attention regulation.

The correlation analysis in the control group indicates in general a more negative association between the changes in behavioural measures and the changes in IAP power. Dispositional mindfulness showed the strongest negative correlation with the IAP power in occipital regions, with a few positive correlations found between the 'observing' sub-domain and midline central and parietal electrodes (Cz and Pz), as well as temporal electrodes (T3 and T4). Conversely to the intervention group, in the control group the interoceptive sensibility showed a trend of negative correlation with changes in the IAP power in the parietal and occipital areas. Another main difference between the correlation results among the two groups is the trend of negative correlation between the interoceptive sensibility and IAP frequency in parietal and occipital electrodes. For the correlation between behavioural measures and alpha relative power, a clear trend of negative correlation emerged for both dispositional mindfulness and interoceptive sensibility across several electrodes.

These results underscore the complex relationship between changes in behavioural and neurophysiological measures and set the stage for future investigations. The position in the brain found to correlate more strongly with the changes in dispositional mindfulness and interoceptive sensibility in the intervention group can be used as a reference to formulate and test specific hypotheses, using more advanced methodological and statistical approaches, such as the use of high-density EEG data and to perform source reconstruction or to combine EEG with other neuroimaging techniques, to elucidate the role of mindfulness training-induced changes in particular brain regions in promoting adaptive psychological processes. Moreover, it appears that different measures to describe the alpha oscillatory activity can be taken into count to describe the relation

with behavioural measures. In this study for example, an increase in dispositional mindfulness is negatively correlated with an increase in the relative power obtained when considering the whole range of alpha frequency band. Contrarily, when considering the IAP power, which describes the intensity of the alpha activity around the peak frequency, a trend of more positive association emerges between dispositional mindfulness and alpha activity, with the exception for the frontal mid-line electrodes, which shows a negative association both with the IAP power and the alpha relative power. These results might be explained by considering that the classic 8-12 Hz range for the alpha band may not adequately capture individual variability. Meanwhile, the power of the IAP, measured at different frequencies for each individual, might better reflect changes in individual alpha band oscillations. Additionally, as pointed out by Klimesch [142], [143], the alpha band can be divided into lower (8-10 Hz) and upper (10-12 Hz) sub-bands, reflecting the activities of distinct neuronal populations and accounting for different neurocognitive functions. Indeed, previous research on mindfulness found different results regarding low and high alpha sub bands [94], [144], indicating that considering the activity in the whole alpha frequency range might not fully capture the effects of a mindfulness training. Following studies on IM should therefore explore the effects on different alpha frequency sub bands.

5.1 Limitations and future directions

This study is subject to several limitations that warrant consideration when interpreting the findings. Firstly, participation was voluntary, and no compensation was provided, potentially introducing selection bias. Individuals self-selected, therefore, participants of the study might have had a strong interest in meditation, potentially limiting the representativeness of the sample among young, healthy adults. Another source of bias emerged during the study, as a significant number of participants dropped out. Consequently, the generalizability of the results may be limited. Additionally, it is important to note that due to the high dropout rate, the final sample size was smaller than the originally calculated sample size, rendering our analysis underpowered to detect the intended effects. Further studies with larger and more diverse samples are warranted to strengthen the robustness of the findings.

The low attendance in the intervention group may have also influenced the results. While the doseresponse effect in meditation research remains unclear, fewer meditation classes could diminish the impact on psychological and neurophysiological parameters Consequently, such an effect might be less detectable when participants are not fully compliant with the intervention. To better understand the dose-response relationship with Integrative Meditation (IM), future research could explore intermediate data points throughout the intervention. This consideration is especially pertinent given the structured and sequential nature of the IM program, where each cycle builds on the abilities developed in the previous one. The absence from one or more sessions could significantly affect participants' ability to fully engage with and benefit from subsequent sessions, potentially skewing the observed effects of the intervention. Additionally, although participants were encouraged to practice meditation individually, this study did not document the extent of each participant's individual practice. As a result, relying solely on class attendance may not fully capture the overall extent of meditation practice that each participant has engaged in. Addressing these aspects in future studies would provide a more comprehensive understanding of the relationship between meditation dosage and its effects.

The significant dropout rate and low attendance among participants in this study prompt a discussion on the reasons for non-compliance with the intervention, a common challenge in other MBI studies. MBIs typically require a substantial time commitment from participants, involving attendance at mindfulness meditation classes and regular home practice over several weeks. In this study, participants, primarily university students, faced logistical challenges such as reaching the meditation class location and balancing academic responsibilities, particularly during the busy exam period. Additionally, the intervention occurred during December and January, coinciding with exams and holiday breaks, which may have further hindered participation. Despite the aim of IM being to support students during exams by enhancing focus and reducing stress, some participants may have perceived attending meditation classes as detracting from their academic obligations.

Individual differences play a significant role in the practice of mindfulness; personality traits like neuroticism and conscientiousness have been reported to impact the efficacy of mindfulness training among university students [145] while conscientiousness and openness foster adherence to out-of-class meditation practice [146]. A study indicates that individuals often favour techniques that align with their personalities, implying that assessing individual differences and customizing MBI to meet individual needs could enhance intervention effectiveness and subsequent outcomes [147]. Future research should explore individual differences and personality traits that correlate with adherence to IM training and its efficacy.

The difficulties in practicing meditation should be contextualized in the Western society; cultural variations in self-construal between the Eastern interdependence and Western independence may influence the efficacy of MBPs, as mindfulness, rooted in Buddhist doctrine, addresses the concept of self [148]. These cultural differences might result in difficulties for individuals in Western cultures to familiarize themselves with the practice of mindfulness. Also, adhering to formal mindfulness practices can pose challenges for many individuals, as lengthy and silent meditation may not suit everyone. Developing a mindful mind is the result of active and purposeful attentional training, requiring frequency and effort [149], [150] as well as a willingness to confront challenges such as self-judgment and aversion [151]. Therefore, it is necessary to explore adaptations that enhance engagement and help overcome obstacles to consistent practice.

In this study, we customized the Integral Meditation (IM) to better suit the needs of young adults, primarily students, by modifying the duration of meditation classes and emphasizing specific types of exercises. While these adjustments may have been beneficial for some participants, they might not have resonated with others. Despite many participants initially showing interest during recruitment and early classes, the challenges presented by the meditation training may have affected their subsequent motivation and engagement. Given that none of the participants completed more than ten classes, the duration of the IM training, consisting of twelve classes over three months, would result in limited exposure to meditation exercises for those who practiced during the classes. Although participants were given audio-guided meditation exercises to practice at home and encouraged to engage in individual meditation sessions, we posit that fostering active participation in meditation practice, both within and outside of classes, would be crucial in improving the effectiveness of IM.

Informal mindfulness practice (IMP) involves integrating moments of mindfulness into daily routines without dedicated time for formal practice. It focuses on engaging in mindful moments during everyday activities, such as noticing distractions and refocusing attention. Studies among elder adults showed that IMP is preferred over the formal practice [152], [153] and IMP has higher adherence in both frequency [154] and practice consistency in the long-term [155] compared to formal practice.

Offering participants guidance on integrating mindfulness into their daily routines, along with teaching brief meditation exercises, could enhance the amount of mindfulness practice and adherence to the IM training. This approach also fosters the cultivation of mindfulness as a habitual practice, rather than solely as an exercise confined to class sessions. These insights should inform future development of the IM program, and subsequent research should consider implementing such strategies to enhance adherence to MBIs, particularly in contexts where adherence may be challenging. Monetary incentive is another policy effective for improving physical health and wellbeing and it also works for MBPs [156], but it does not suit for research with scarce funding.

Another significant limitation of this study is the absence of an inactive control group, a constraint also observed in previous research on IM. In future studies, it will be essential to evaluate the efficacy of IM against an active control group. This will allow researchers to differentiate global effects induced by IM training from those attributable to its components, such as participating in group activities or receiving respiratory training.

The use of self-report measures introduces inherent limitations that merit consideration in this study. Firstly, self-report data relies on participants' subjective perceptions, memories, and interpretations, which may be influenced by factors such as social desirability or a recall bias, leading to inaccuracies in the collected data. Additionally, self-report measures may be susceptible to individual differences in comprehension and interpretation of the questions, potentially compromising the reliability and validity of the gathered information. Moreover, the use of self-report measures might be influenced by participants' emotional states at the time of assessment, impacting the accuracy of their responses. While self-report measures offered valuable insights into participants' experiences, emotions, and behaviours, we should acknowledge these limitations and consider incorporating complementary objective measures to enhance the robustness and comprehensiveness of the findings in future research.

The absence of significant effects on state and trait anxiety warrants further exploration. Future research could delve deeper into understanding the conditions under which meditation interventions may influence anxiety, possibly considering the role of intervention duration, intensity, or participant characteristics.

Relying solely on alpha activity, rather than on the entire EEG frequency spectrum, poses notable limitations for the study. By focusing exclusively on alpha oscillations, the study may overlook potential contributions and nuances within other frequency bands, like theta, beta, or gamma, which play distinct roles in cognitive and emotional processing and have been reported in other mindfulness meditation studies. EEG is a complex signal that reflects multifaceted brain dynamics, and restricting the analysis to alpha activity alone may provide an incomplete picture of the impact of mindfulness. Furthermore, the exclusive reliance on EEG data for assessing mindfulness effects may not capture the broader spectrum of physiological changes associated with mindfulness

practices. To enhance the study's validity and depth, future research could consider a more comprehensive analysis of multiple EEG frequency bands and complement the EEG data with other physiological measures such as Heart Rate Variability.

A notable limitation of our EEG activity examination was adhering to the traditional alpha frequency range of 8-12 Hz, a common practice in meditation literature [157]. However, it is important to acknowledge that various studies exploring alpha activity have reported different frequency ranges, suggesting a broader definition, such as 7-13 Hz. An alternative and intriguing approach is the use of the Individual Alpha Frequency (IAF), which has gained widespread use as a metric for assessing alpha activity [124], [158]. Relying solely on a fixed frequency band, like 8–12 Hz, carries the risk of introducing bias, particularly against individuals with an IAF beyond this range [159]. Embracing the IAF offers a more personalized and accurate evaluation of alpha activity, accommodating individual variations and enhancing the precision of neurophysiological assessments. Moreover, the IAF can be utilized to estimate other frequency band ranges and alpha sub-bands. In our study, we treated the alpha frequency band as a single entity, not differentiating between low-alpha and high-alpha frequencies. Additionally, the calculation of the IAP frequency was based on the frequency with the highest power in the 8-12 Hz range, which did not specifically address cases with multiple alpha peaks, split alpha peaks, or unclear alpha peaks.

Given the numerous limitations, the results of this study should therefore be interpreted cautiously and should not be considered conclusive for investigating the effects of IM training. The limitations described here should be addressed in future studies to expand the understanding of IM effects.

5.2 Conclusion

In summary, this study investigated the effectiveness of the IM intervention among healthy young adults, employing a two-group pretest-posttest experimental design. This study represents the first attempt to assess the neural correlates of the IM alongside behavioral measures. The results demonstrated significant improvements in dispositional mindfulness and interoceptive sensibility, as measured by the FFMQ and MAIA questionnaires, indicating a heightened awareness of both mental and bodily signals. Contrary to initial hypotheses, however, there were no significant intervention effects on state and trait anxiety scales or interoceptive accuracy. The neurophysiological analysis revealed a noteworthy effect in the evaluation of IAP power, emphasizing the importance of individual differences in response to the IM. Exploratory analyses across different brain regions suggested that the intervention might modulate alpha frequency band activity differently, with associations between baseline measures and post-intervention changes. The partial correlation analysis highlighted complex relationships between changes in behavioural and neurophysiological measures, pointing towards specific electrode locations that correlated more strongly with improvements in dispositional mindfulness and interoceptive sensibility. These findings underscore the intricate interplay between mindfulness practice, psychological well-being, and neurophysiological changes, providing valuable insights for future research and the refinement of mindfulness interventions tailored to individual needs.

Our results support a shift toward personalized approaches in mindfulness interventions, acknowledging that participants with diverse baseline characteristics may exhibit varied responses to the same intervention. This not only raises questions about the generalizability of mindfulness

interventions but also emphasizes the importance of tailoring these practices to individual needs and characteristics. As the field of mindfulness research progresses, acknowledging and addressing these individual differences will be essential in refining and optimizing the effectiveness of mindfulness interventions for diverse populations.

Ethics approval and consent to participate

The study was approved by the Ethics Committee of the Department of Brain and Behavioral Sciences of the University of Pavia (Prot. n 129/23). The study was performed in accordance with the ethical standards as laid down in the 1964 Declaration of Helsinki and its later amendments. Participants were informed that their personal data would be treated in accordance with the privacy policy and all participants signed informed consent.

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