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ORIGINAL ARTICLE



Theta band brainwaves in human resting EEG modulated by mobile phone radiofrequency

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ABSTRACT

Purpose: Wireless communication has become an integral part of our lives. The growing number of antennas in our environment and the expanding use of mobile phones (MPs) are increasing the population's exposure to electromagnetic fields. The present study aimed to examine the potential impact of MPs radiofrequency electromagnetic fields (RF-EMF) exposure on the brainwaves of the resting electroencephalogram (EEG) in humans.

Materials and Methods: Twenty-one healthy volunteers were exposed to Global System for Mobile communications (GSM) signal at 900 MHz MP RF-EMF. The maximum specific absorption rate (SAR) of the MP averaged on 10 g tissue and 1 g tissue were measured at 0.49 W/kg, 0.70 W/kg, respectively.

Results: Results showed that while delta and beta rhythms of resting EEG were not affected, theta brainwaves were significantly modulated during exposure to RF-EMF related to MPs. For the first time, it was shown that this modulation is dependent on the eye condition, i.e. closed or open.

Conclusions: This study strongly suggests that acute exposure to RF-EMF alters the EEG theta rhythm at rest. Long-term exposure studies are required to explore the effect of this disruption in high-risk or sensitive populations.

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1. Introduction

The intensive use of mobile phones (MPs) and the growing electromagnetic fields in the environment have dramatically increased people's exposure to radiofrequency electromagnetic fields (RF-EMF). This exposure has raised questions about the potential effects of RF-EMF on human health. In this context, the electroencephalogram (EEG) has been an interesting neuroimaging technique to study potential effects of MPs RF-EMF emission on brain activity. Indeed, the EEG is an important noninvasive approach largely used in clinical and basic research to measure the brain electrical activity by recording the electrical signals generated by the addition of synchronized postsynaptic potentials from a large number of neurons in the cerebral cortex with millisecond resolution (Cacioppo et al. 2007). To do that, the EEG uses electrodes placed on the scalp, as well as reference electrodes. Although the electrical activity of the brain can vary between individuals, age, or health state, five main periodic rhythms of electrical potential oscillation of neurons in the cortex can be recognized in adult EEGs according to their frequency and voltages: gamma (>30 Hz), beta (13–30 Hz), alpha (8–13 Hz), theta (4–8 Hz), and delta (0.5–4 Hz) bands.

Several studies investigated MPs effects on EEG resting-state of healthy volunteers—adolescents, adults, and elderly—as well as epileptic patients (Wallace and Selmaoui 2019). In particular, all of these studies focused on different MPs technologies, covering the whole range of RF-EMF signals from the 1st generation of MPs (1 G) to at least the 4 G (Wallace and Selmaoui 2019). Even though some investigations did not find any significant effect of the exposure on the EEG, the bulk of the studies on this topic reported that exposure to RF-EMF seems to affect the EEG power spectral density at rest. The main effect has been seen on the spectral power of the alpha band (for review, see (Danker-Hopfe et al. 2019; Hinrikus et al. 2021; Wallace and Selmaoui 2019). More precisely, the alpha band has been one of the most studied parameters in relation to the effect of RF-EMF. It should be noted that few other studies have examined the effects of RF-EMF exposure on other frequency bands (delta, theta, beta, and gamma) with conflicting results (Croft et al. 2002; D'Costa et al. 2003; Hinrikus, Bachmann, Lass, Karai et al. 2008; Hinrikus, Bachmann, Lass, Tomson et al. 2008; Hinrikus et al. 2011; Suhhova et al. 2013; Roggeveen et al. 2015). The aim of the present study was to examine the potential impact of Global System for Mobile communications (GSM) RF-EMF signal exposure on the delta, theta,

and beta bands of the resting EEG. Moreover, we aimed to provide additional information to fill certain gaps in our current knowledge of the effects of GSM RF-EMF exposure on control parameters, such as eye condition (closed or open).

2. Materials and methods

2.1. Participants

Twenty-one right-handed healthy volunteers with normal or corrected to normal vision (10 females and 11 males; mean age $\pm$ SD: 25.1 ± 3.6 years; mean body-mass index $\pm$ SD: 21.5 ± 1.9) entered the study. We assessed inclusion criteria with a routine clinical examination and a clinical interview to exclude subjects with any history of head injury, neurological or psychiatric disease, any chronic disease, disability, or recent acute illness during last month. Inclusion criteria also included regular sleep habits with a regular sleep-wake cycle (8 am–11 pm $\pm$ 1 h), no medication, no smoking, and no drug use. Participants were not allowed to consume alcohol and caffeine for at least 24 hours before each experimental session. They have to maintain the regular sleep-wake cycle, not to use their personal MP on the days of the experiment, and to fast for at least two and half hours before experimental sessions. All women participated in the experimental sessions only during the follicular phase of the menstrual cycle. All participants provided a written informed consent before the investigation and were compensated for their participation. All procedures were approved by the French Ethics Committee and were in accordance with the Declaration of Helsinki.

2.2. RF-EMF exposure and dosimetry

In order to follow a double-blind design, we used two identical commercial dual-band GSM MPs (Nokia 6650), which differ only in the type of RF-EMF emission. Indeed, the sham or real exposure was carried out using a ‘load’ or a ‘dummy load’ of the MP, respectively. The real MP was set to transmit a modulated 900 MHz GSM signal (peak 2 W, average 250 mW, pulse modulated with a duty cycle of 1/8) at its maximum power using a service software (Phoenix, Nokia Corp., Finland). Further details about the RF-EMF exposure setup are available elsewhere (Wallace et al. 2022).

A dosimetry analysis revealed that the maximum specific absorption rate (SAR) of the real MP averaged on 10 g tissue and 1 g tissue were measured at 0.49 W/kg, 0.70 W/kg, respectively. On the contrary, the SAR of the sham MP was below the detection level of the system (0.001 W/kg). Moreover, a polystyrene phantom head test was used to avoid any interferences between RF-EMF and EEG recordings during the exposure phase. Results showed that MPs did not affect EEG signals during the exposure. Further details about dosimetry and phantom assessment are described elsewhere (Ghosn et al. 2015; Wallace et al. 2022).

2.3. Experimental protocol

This study is part of the broad study published previously (Wallace et al. 2022). A double-blind, randomized, counter-balanced, crossover design was employed where each volunteer underwent two experimental sessions one week apart at the same time of day to avoid circadian rhythm bias. According to the protocol described previously (Wallace et al. 2021; 2022) and shown in Figure 1, each experimental session started with 12 minutes of baseline EEG recordings combined with magnetoencephalography (MEG) in a specific MEG room (from run 1 to run 2), followed by 12 minutes of baseline EEG recording in another room (from run 3 to run 4). During both baseline phases, there was no RF-EMF exposure. During the sham or real RF-EMF exposure phase, EEG was recorded for 25 minutes and 30 seconds (from run 5 to run 8). During EEG recordings from run 3 to run 8, subjects were comfortably seated in a sound-attenuating shielded room, while remaining still and staring at a central black fixation mark (black dot surrounded by a black circle) presented on a gray background at a viewing distance of 1 m from the eye. Finally, during the post-exposure phase with no RF-EMF exposure (from run 9 to run 12) EEG combined with MEG was recorded for 25 minutes and 30 seconds. Each run included a 3-minutes recording interval with eyes-open (EO) and 3-minutes interval with eyes-closed (EC) in alternation with a 30-second pause at the end.

2.4. Data acquisition

Depending on whether the EEG was combined with the MEG recordings or not, different acquisition systems were used. For this reason, we included the analyses of EEG recorded only during the baseline (runs 3 and 4) and the exposure phase (runs 5–8).

The EEG signal from run 3 to run 8 was acquired using the BrainVision BrainAmp MR plus Amplifiers (Brain Products GmbH, Germany; Figure 1), from 74 EEG-channel system with Ag/AgCl sensors placed on the participant’s scalp according to the standard 10/10 system (Figure 2), referenced to mastoids, and grounded on the left scapula. EEG data were recorded with a sampling frequency of 1 kHz and a bandwidth of 0.016–250 Hz. Ocular movements were recorded with horizontal electro-oculogram (EOG) applying two electrodes near the outer canthi of the left and right eyes, as well as two electrodes placed above and below the right eye to measure vertical EOG. EEG electrical impedances were maintained lower than 20 k Ω , while impedances of reference signal, ground signal, and EOG were maintained lower than 5 k Ω .

2.5. EEG data preprocessing and frequency analysis

EEG data from run 3 to run 8 were analyzed using MNE-Python, version 0.19.2 (Gramfort et al. 2013). First, we removed from all data analysis the electrodes too close to the EEG cap ear-slits (i.e. T9, TP9, T10, TP10) for all

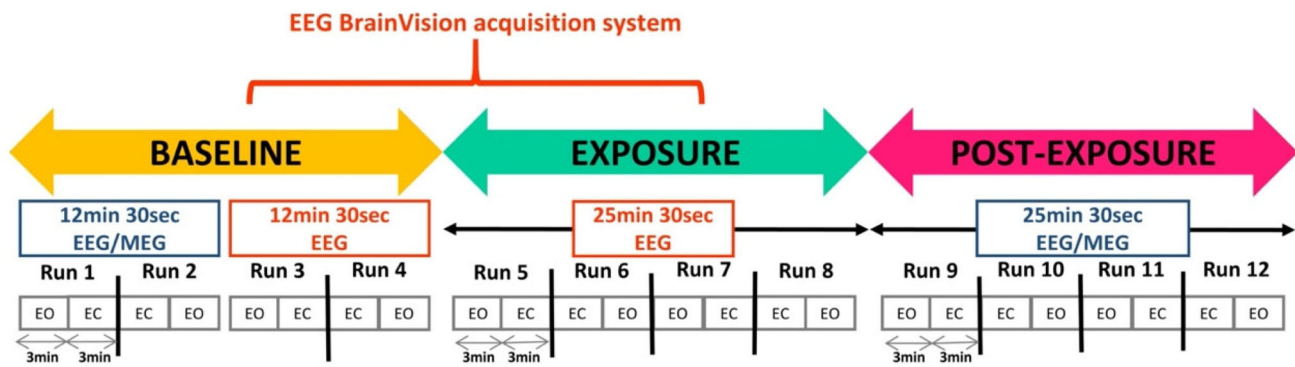


Figure 1. Experimental protocol. EEG/MEG: electroencephalography combined with magnetoencephalography; EEG: electroencephalography; EO: eyes-open; EC: eyes-closed.

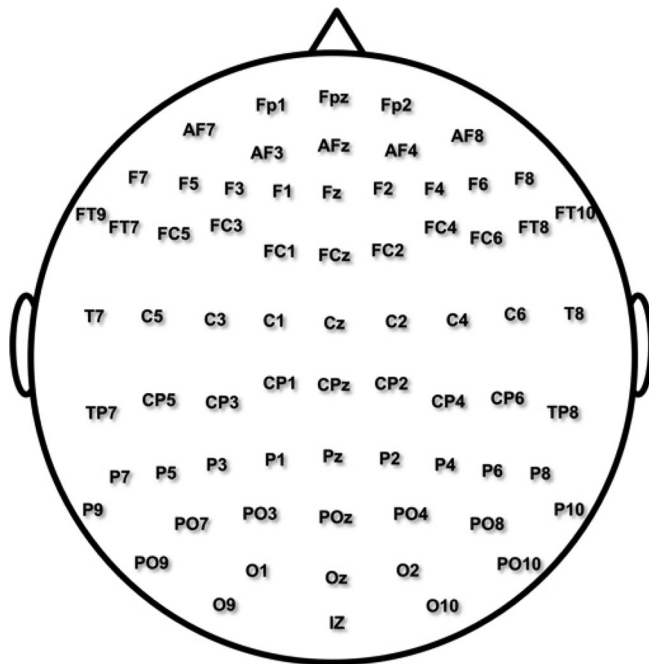


Figure 2. Electroencephalogram layout according to the standard 10/10 system.

subjects, and band-pass filtered the EEG signal between 1–20 Hz using the finite impulse response filter design with Hamming windows. Then, we applied independent component analysis (ICA) with the FastICA algorithm to decompose the signal and reject ocular artifacts. Pearson correlations were used to find vertical and horizontal EOG-related components, and bad epochs were created 500 ms before and after each eye blink, with band-pass filtering between 1 and 10 Hz.

Second, EEG ICA-corrected data were used to analyze the power spectral density for each EEG electrode considering 2 minutes and 30 seconds of EO and EC intervals of each run. We applied the fast Fourier transformation (Welch technique, Hamming windowing function, 4096 points per window, 50% of overlap between segments) to the EO and EC data, considering the beta (12–20 Hz), theta (4–8 Hz), and delta (1–4 Hz) frequency bands. The power values were log-transformed to approach a normal distribution.

Third, we averaged the log-transformed power values of each frequency band of interest over the baseline and the exposure phases with EO and EC separately, for both experimental sessions and for all subjects. Then, the baseline log-transformed data computed for each electrode were subtracted from the exposure log-transformed data computed for each electrode, considering the EO and EC conditions of each session separately. Finally, results were plotted as topographic maps showing the power values calculated for each electrode on the scalp.

2.6. Statistical analysis

Statistical analysis was performed using a custom-written MATLAB (R2019) toolbox (Yahia-Cherif 2022). First, we used a three-way repeated measures analysis of variance (ANOVA) on power values computed for each frequency band to determine a period effect (factor 1 with two levels: baseline and exposure), eyes condition effect (factor 2 with two levels: EO and EC), and exposure condition effect (factor 3 with two levels: sham exposure and real exposure), as well as their interactions. Second, we applied the one-way repeated measures ANOVA on baseline-corrected exposure power values of each frequency band comparing the two exposure conditions, and considering EO and EC data separately. F and *p*-values of both ANOVA analyses were computed for each electrode on the scalp. Statistical significance was set as $p < .05$. We further applied a *post-hoc* cluster-based permutation test to ANOVA analyses of theta band power values to correct for multiple comparisons (Maris and Oostenveld 2007). For each main effect or interaction of theta band, samples whose F-value exceeded a threshold (here, F-values corresponding to a one-tail *p*-value of .05) were clustered based on space adjacency given by an electrodes neighboring matrix. We estimated the distribution of the maximum cluster level F-statistics under the null hypothesis through 1000 randomizations for each main effect and each possible interaction, while we rejected the null hypothesis with a Monte-Carlo *p*-value $< .05$ if the observed statistic was greater than 95% of the statistic values obtained from randomized data. Finally, statistical analysis results were plotted as topographic maps showing the *p*-values calculated for each electrode on the scalp.

3. Results

3.1. Three-way repeated measures ANOVA results

As shown in Figure 3(A), results of beta band three-way repeated measures ANOVA reported that only the FT9

electrode showed a statistically significant difference ($p = .0132$) for time factor (two levels: baseline and exposure), and only one frontal electrode (F7) showed a statistically significant difference ($p = .0358$) for the exposure condition factor (two levels: real and sham). On the other

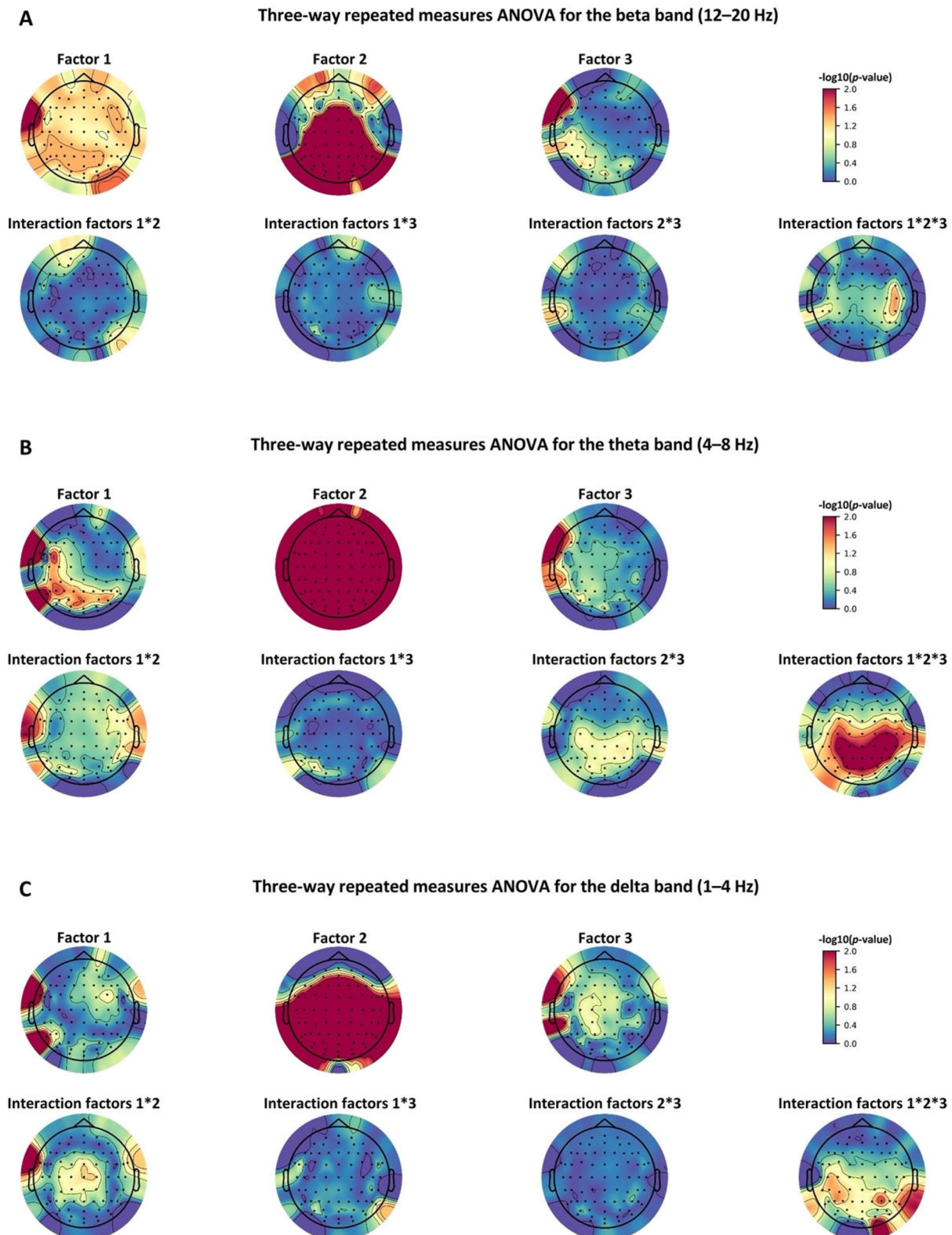


Figure 3. Results of the three-way repeated measures ANOVA for the EEG (A) beta, (B) theta, and (C) delta bands. Results are shown as p -value topography maps, where p -values were computed and plotted for each electrode. Statistical significance was set as $p < .05$. Factor 1: baseline vs. RF-EMF exposure; Factor 2: eyes-open vs. eyes-closed; Factor 3: sham RF-EMF exposure vs. real RF-EMF exposure; *: interaction between factors.

hand, the eye condition factor (two levels: EO and EC) showed statistically significant differences widespread over the central and occipital scalp regions. Considering the interactions between factors, results revealed a significant interaction between all factors at electrodes C6 ($p = .0359$) and CP6 ($p = .0469$).

Results of the theta band three-way repeated measures ANOVA are reported in Figure 3(B). The time factor showed statistically significant differences at centro-parietal and occipital electrodes (FT9, FC5, CP5, CP3, P9, P7, P5, P3, P1, Poz, PO4, PO8, and O1), while the exposure condition factor showed a statistically significant difference depicted only by the FT9 electrode ($p = .0248$). The eye condition factor showed significant differences widespread over the whole scalp. The interaction between all factors showed a statistically significant difference involving the central, parietal, and occipital electrodes (C1, Cz, C2, C4, C6, T8, CP5, CP3, CP1, Cpz, CP2, CP4, CP6, TP8, P9, P7, P5, P3, P1, Pz, P2, P4, P6, PO9, PO7, PO3, Poz, PO4, O1, and Oz).

Results of the delta band three-way repeated measures ANOVA are reported in Figure 3(C). The time factor showed a statistically significant difference between power values measured at baseline and exposure recording phases depicted by electrodes FT9 ($p = .0031$) and P9 ($p = .0045$). Similarly, only two electrodes, FT9 ($p = .0456$) and TP7 ($p = .0155$), showed a statistically significant difference between real and sham exposure phases for the exposure condition factor. On the contrary, the eye condition factor showed significant differences widespread over the scalp. As before, the interaction between all factors showed a slight statistically significant difference involving centro-parietal and posterior electrodes (CP5, CP3, P5, and O10).

In Figure 4 are shown the results of the cluster-based permutation of the three-way repeated measures ANOVA. Only time factor, eyes condition factor, and interaction between all factors reported significant clusters. Considering the time factor, results showed one significant cluster involving left centro-parietal electrodes (Monte Carlo $p < .001$),

one significant cluster involving left parietal electrodes (Monte Carlo $p < .001$), two significant clusters involving fronto-temporal electrodes (Monte Carlo $p < .001$), and one significant cluster involving right parieto-occipital electrodes (Monte Carlo $p < .001$). Considering the eyes condition, we found one significant cluster involving all electrodes over the scalp (Monte Carlo $p < .001$). The interaction between all factors showed one significant cluster involving central, temporal and parietal areas (Monte Carlo $p < .001$) and two more significant clusters at the parieto-occipital region (Monte Carlo $p = .001$ and Monte Carlo $p < .05$, respectively).

3.2. One-way repeated measures ANOVA results

As shown in Figure 5(A), the one-way repeated measures ANOVA showed that power values of the beta band significantly decreased at the C6 ($p = .0489$) and CP6 ($p = .0448$) electrodes during the real RF-EMF exposure phase relative to the sham exposure with EO. However, no statistically significant differences were found between beta band power values measured during the real and sham exposure phases with EC.

Results of the one-way repeated measures ANOVA showed a statistically significant decrease of the theta band powers computed at centro-parietal and occipital electrodes (C4, C6, CP1, CPz, CP2, CP4, P3, P1, Pz, P2, P4, P6, PO3, Poz, PO4, and O2) with EO during the real RF-EMF exposure compared to sham exposure. On the other hand, with the EC condition, the power values computed for the theta band revealed a slight statistically significant increase during the real exposure phase relative to sham exposure at parietal electrodes (P9, P7, P3, and PO7) (Figure 5(B)).

Concerning the delta band one-way repeated measures ANOVA, results reported a slight decrease of power values depicted at the P4 electrode ($p = .0418$) during the real RF-EMF exposure, relative to sham with EO, while showing an increase of power values depicted by P10 ($p = .0370$)

Cluster-based permutation three-way repeated measures ANOVA

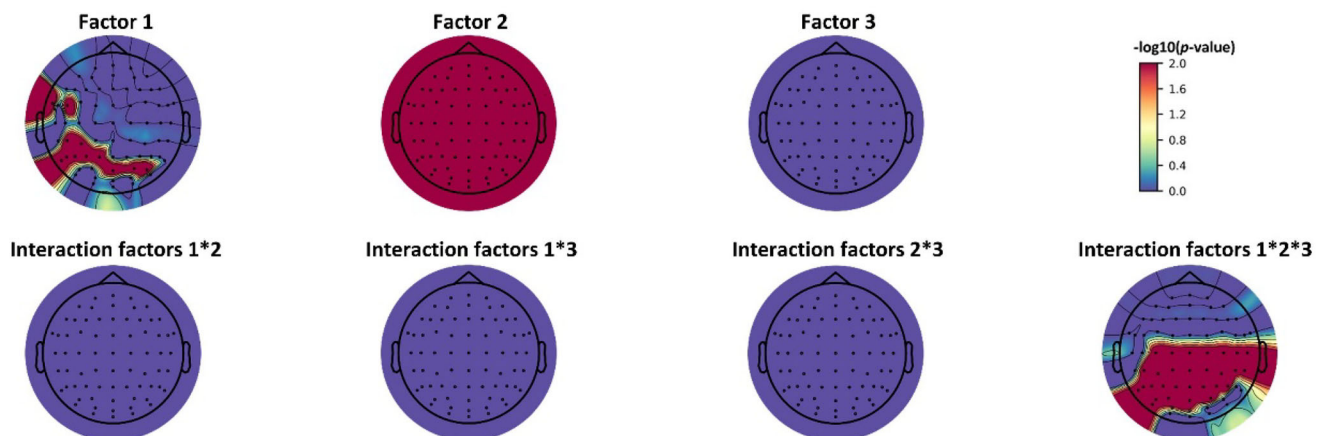


Figure 4. Results of the cluster-based permutation of three-way repeated measures ANOVA for the EEG theta band. Results are shown as p -value topography maps, where p -values were computed and plotted for each electrode. Statistical significance was set as $p < .05$. Factor 1: baseline vs. RF-EMF exposure; Factor 2: eyes-open vs. eyes-closed; Factor 3: sham RF-EMF exposure vs. real RF-EMF exposure; *interaction between factors.

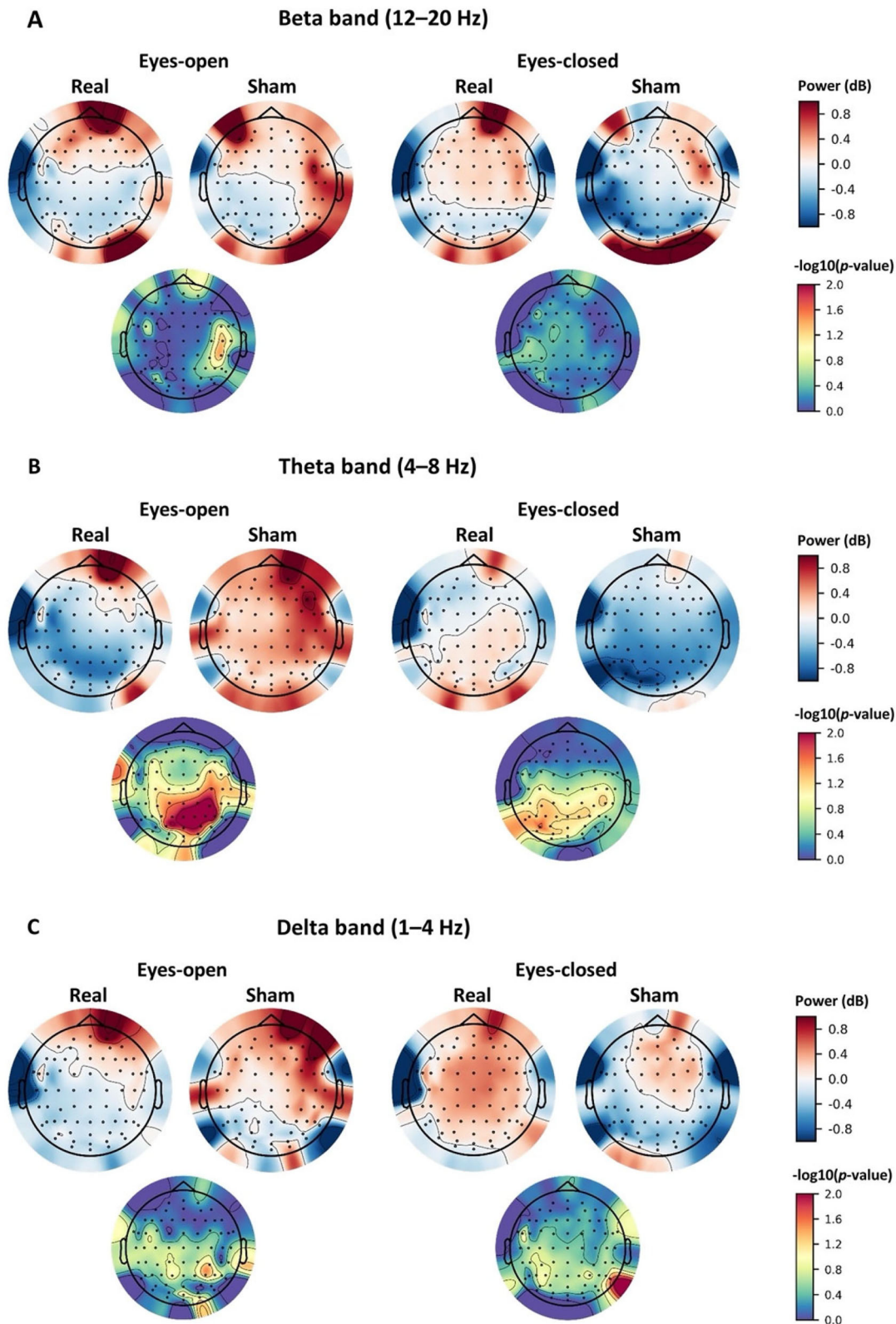


Figure 5. Topography maps for (A) beta, (B) theta, and (C) delta bands. Power topography maps show EEG baseline-corrected power values computed for each electrode during the real and sham RF-EMF exposures with eyes-open and eyes-closed. *p*-value topography maps show the results of one-way repeated measures ANOVA on the EEG baseline-corrected powers (real exposure vs. sham exposure). *p*-values were computed and plotted for each electrode. Statistical significance was set as $p < .05$.

during the real exposure relative to sham exposure with EC (Figure 5(C)).

In Figure 6 are shown the results of the cluster-based permutation of the one-way repeated measures ANOVA.

Considering the EO condition, we found one significant cluster involving centro-parietal electrodes (Monte Carlo $p < .001$) and one significant cluster at parieto-occipital region (Monte Carlo $p < .05$), while we found one significant

Cluster-based permutation one-way repeated measures ANOVA

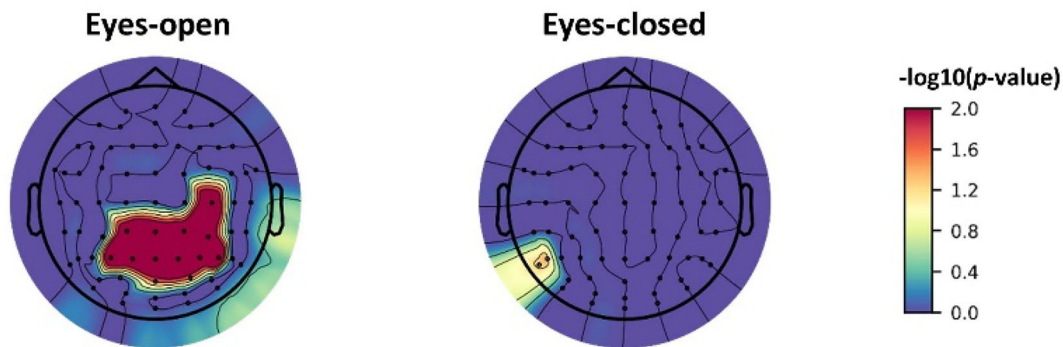


Figure 6. Results of the cluster-based permutation of one-way repeated measures ANOVA (real exposure vs. sham exposure) for the EEG baseline-corrected powers of the theta band. Results are shown as *p-value* topography maps, where *p*-values were computed and plotted for each electrode for eyes-closed and eyes-open conditions. Statistical significance was set as $p < .05$.

cluster involving left parietal electrodes (Monte Carlo $p < .05$) for the EC condition.

4. Discussion

In this study, we investigated the potential effects of the exposure to RF-EMF related to GSM 900 MHz MPs on the electrical brain activity of healthy volunteers at rest in a double-blind, randomized, counterbalanced, and crossover EEG protocol. We analyzed the EEG power spectral density of the beta (13–20 Hz), theta (4–8 Hz), and delta (1–4 Hz) bands before and during RF-EMF exposure, considering EEG recordings with EO and EC. The analysis of baseline-corrected data showed that, among the three investigated frequency bands, the theta band seemed to be significantly affected by RF-EMF exposure. Indeed, the beta and delta oscillations reported slight effects of RF-EMF exposure, as shown by both three-way and one-way ANOVA statistical analysis. We could assume that the weak effect observed on these bands is likely to represent a random occurrence rather than a real effect. On the contrary, the theta band reported a greater effect of the exposure (which survived to multiple comparison correction) with a significant decrease of the power during the real exposure compared to the sham exposure with the EO and a significant increase with the EC. This power modulation related to RF-EMF exposure was depicted by the centro-parietal and occipital electrodes. It is noteworthy that, even though the theta oscillation is generally widely distributed across the scalp (Segalowitz et al. 2010), it has shown a high degree of MEG resting-state reliability in the parietal cortex over three years (Lew et al. 2021), which could explain our findings. Moreover, we can exclude any influences on brain electrophysiology related to physiological factors known to modulate EEG activity. As previously shown elsewhere (Wallace et al. 2021), we assessed some confounding factors (salivary biomarkers of stress, heart rate, heart rate variability, and salivary caffeine concentration), which did not report any statistically significant differences related to the exposure condition.

Our EEG results are in line with previous results by (Hinrikus et al. 2011), which reported a power increase of the theta band related to 450 MHz RF-EMF exposure during EEG resting-state recording of healthy volunteers with EC; in particular, our theta band results confirm these previous results with the eyes-closed condition. On the other hand, the data obtained with EO went in the opposite direction. The theta band spectral power decreased when the eyes were open in our experiment. This suggests that the eyes-open or eyes-closed condition could affect the results of the theta band response to RF-EMF exposure. This is in agreement with our previous results (Wallace et al. 2021), which highlighted that RF-EMF exposure affected the alpha band activity differently depending on the condition of the eyes (open or closed). Furthermore, previous studies investigating RF-EMF effects on EEG reported somewhat different results for the delta and beta bands. Indeed, findings showed either a decrease in the delta (Croft et al. 2002) and beta bands powers (D'Costa et al. 2003; Yang et al. 2017), or an increase in the delta (Hinrikus et al. 2011) and beta bands powers (Hinrikus, Bachmann, Lass, Karai et al. 2008; Hinrikus, Bachmann, Lass, Tomson et al. 2008; Hinrikus et al. 2011; Suhova et al. 2013; Roggeveen et al. 2015). Other studies reported a modulation of the gamma band activity with an increase in the power spectral density related to the RF-EMF exposure (Hinrikus et al. 2011; Curcio et al. 2015; Roggeveen et al. 2015). This divergence in results may be the result of differences between studies in protocol designs as reported above (e.g. eyes condition), participant inclusion criteria, RF-EMF frequencies and intensities, EEG data acquisition, or data analysis, and statistics, as reviewed elsewhere (Danker-Hopfe et al. 2019; Wallace and Selmaoui 2019).

Since the theta band is considered a marker of memory functions, processing information and attention, and its synchronization reflects working memory performance (Klimesch 1996; Klimesch et al. 1997; Klimesch 1999; Jensen and Tesche 2002; Klimesch et al. 2005; Grover et al. 2022), a question arises spontaneously: what is the clinical implication

of its modulation related to RF-EMF exposure? Based on the results of the current study, it is too early to answer this question; however, it has been shown that the MP RF-EMF exposure modulated the event-related desynchronization of the alpha band, enhancing human cortical neural efficiency and cognitive-motor processes (Vecchio et al. 2012). Generally, previous results suggested that RF-EMF related to GSM MPs may have a slight effect on human attention and working memory (Barth et al. 2008). Moreover, it is noteworthy that some noninvasive brain stimulation techniques, such as transcranial magnetic stimulation, transcranial direct current stimulation (tDCS), oscillatory tDCS, or transcranial alternating current stimulation (tACS), employ either magnetic or electric fields in order to stimulate the cortical neuron assemblies and modulate behavioral responses. In particular, it has been shown that theta-tACS administration had a positive effect on working memory functions (Albouy et al. 2018), allowing to enhance long-lasting working memory capacities in the elderly through repetitive applications (Grover et al. 2022).

5. Conclusions

In conclusion, this work contributes to the growing literature that suggests the potential modulation of human spontaneous EEG by exposure to RF-EMF. Indeed, it showed that acute exposure to MP RF-EMF at 900 MHz modulates the spectral power of the theta band in healthy volunteers at rest. To our knowledge, it also emphasized the role of the eye condition in the effect of RF-EMF on EEG for the first time.

Clinical implications of this modulation need further investigation. In particular, chronic exposure effects should be assessed, especially in high-risk and sensitive populations.

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Disclosure statement

No potential conflict of interest was reported by the author(s).

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