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Human resting-state EEG and radiofrequency GSM mobile phone exposure: The impact of the individual alpha frequency

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ABSTRACT

Purpose. With the extensive use of mobile phone (MP) several studies have been realized to investigate the effects of radiofrequency electromagnetic fields (RF-EMF) exposure on brain activity at rest via electroencephalography (EEG), and the most consistent effect has been seen on the alpha band power spectral density (PSD). However, some studies reported an increase or a decrease of the PSD, while others showed no effect. It has been suggested that these differences might partly be due to a variability of the physiological state of the brain between subjects. So, the aim of this study was to investigate the alpha band modulation, exploring the impact of the alpha band frequency ranges applied in the PSD analysis.

Materials & Methods. Twenty-one healthy volunteers took part to the study with a double-blind, randomized and counterbalanced crossover design, during which eyes-open (EO) and eyes-closed (EC) resting-state EEG was recorded. The exposure system was a sham or a real GSM (global system for mobile) 900 MHz MP (pulse modulated at 217 Hz, mean power of 250 mW and 2 W peak, with a maximum specific absorption rate of 0.70 W/kg on 1 g tissue). The experimental protocol presented a baseline recording phase without MP exposure, an exposure phase during which the exposure system was placed against the left ear, and the post-exposure phase without MP. EEG data from baseline and exposure phases were analyzed and PSD was computed for the alpha band in the fixed range of 8–12 Hz and for the individual alpha band frequency range (IAF).

Results. Results showed a trend in decrease or increase of EEG power of both alpha oscillations during exposure in relation to EC and EO recording conditions, respectively, but not reaching statistical significance. Findings did not provide evidence for a different sensitivity to RF-EMF MP related to individual variability in the frequency of the alpha band.

Conclusion. In conclusion, these results did not show alpha band activity modulation during resting-state under RF-EMF. It might be argued the need of a delay after the exposure in order

to appreciate an EEG spectral power modulation related to RF-EMF exposure.

INTRODUCTION

Mobile phone (MP) has become an integral part of our daily life with a worldwide usage that has considerably increased in recent years (ITU 2018). Given the proximity of head to MP when hands-free devices are not used, several research focused on non-thermal biological effect of electromagnetic field of radiofrequencies (RF-EMF) related to the MP on the human brain electrophysiology. The majority of previous studies (about the 80%) which investigated spontaneous brain activity with electroencephalography (EEG) showed that RF-EMF related to MP could affect the resting EEG, with a mainly effect on the alpha band (Wallace and Selmaoui 2019).

Between cortical oscillations divided into bands by frequency, the alpha band has received great attention from scientists, whereas initially for many decades it has been considered as passive and idling state of the brain (Pfurtscheller et al. 1996). With recent studies the alpha band seems to be a pure cognitive signal with an active role in relaxed wakefulness processing (for review (Başar 2012), (Klimesch 2012)) with an amplitude of about 10 to 45 μ V (Cacioppo et al. 2007). Topographically, its greatest amplitude is localized over the posterior regions at the occipito-temporal and parietal regions (Niedermeyer and da Silva 2005) and can best be seen during resting periods in which the subjects have the eyes closed (Barry et al. 2007). Its rhythmic activity is commonly attributed in the range of about 8–12 Hz which most likely is generated in thalamo-cortical feedback loops as well as in the cortex (Klimesch 1997; da Silva 2013). The alpha rhythm is blocked or attenuated by attention processes and it was showed to play a direct role in alertness, attention and consciousness state (Palva and Palva 2007). Specifically, in cognitive tasks, the lower alpha (8–10 Hz) suppression (or desynchronization) is associated with a task-unspecific increase in attentional demands. On the other hand, event-

related changes indicate that the upper alpha (10–12 Hz) desynchronization is positively correlated with semantic long-term memory performance, processing of sensory semantic information and stimulus specific expectancy (Klimesch 1999; Minarik et al. 2018).

Our previous study (Ghosn et al. 2015) showed a decrease of the lower and upper alpha band during and after 900 MHz RF-EMF exposure in health young adults. Similar findings were reported by some authors (D'Costa et al. 2003; Maby et al. 2006; Perentos et al. 2013; Yang et al. 2017; Vecsei et al. 2018), while others claimed an increase of the alpha band spectral power related to the RF-EMF exposure (Reiser et al. 1995; Croft et al. 2002; Regel, Gottselig, et al. 2007; Croft et al. 2008; Hinrikus, Bachmann, Lass, Karai, et al. 2008; Croft et al. 2010; Roggeveen et al. 2015).

So, despite the great reliability of the alpha band reported by many EEG test-retest analysis, (Pollock et al. 1991; Burgess and Gruzelier 1993; Fingelkurts et al. 2006; Corsi-Cabrera et al. 2007; Cannon et al. 2012), results have been quite heterogeneous and hardly to reconcile.

It is noteworthy that few studies investigated the effects of the RF-EMF exposure on other frequency bands (delta, theta, beta and gamma) and they reported heterogeneous results. Indeed, as for the alpha band, some studies showed a decrease of the delta (Croft et al. 2002) and beta bands power (D'Costa et al. 2003; Yang et al. 2017) related to MP exposure, while other showed an increase of the delta band power (Hinrikus et al. 2011) and beta band power (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) as consequence of RF-EMF exposure. Moreover, only one study reported a power modulation of the theta band (EEG power increase) (Hinrikus et al. 2011), while several studies found that the gamma band power was increased with the RF-EMF exposure (Hinrikus et al. 2011; Curcio et al. 2015; Roggeveen et al. 2015).

It is argued that all these discrepancies in results could be related to differences in participant

inclusion criteria, protocol approaches, RF-EMF frequencies and intensities, data analysis and statistics, as reviewed elsewhere (Danker-Hopfe et al. 2019; Wallace and Selmaoui 2019).

Interestingly, we can also argue that EEG modulation under RF-EMF exposure could be related to inter-individual variability because of an individual sensitivity to RF-EMF, as suggested by studies on RF-EMF effects on resting EEG (Hinrikus, Bachmann, Lass, Karai, et al. 2008) and sleep (Loughran et al. 2012). This individual sensitivity should not be interpreted as hypersensitivity to RF-EMF, but rather as a variability of the physiological state of the brain (Hinrikus, Bachmann, Lass, Karai, et al. 2008). So, we could assume that the heterogeneous results reported in literature about RF-EMF exposure effects on human spontaneous EEG (decreased or increased EEG power) could be also related to electrophysiological inter-individual variability factors when measuring EEG activity.

First of all, an important EEG parameter that could vary between subjects is the frequency range of the alpha band. Indeed, even though the alpha band is commonly defined as a fixed frequency band of about 8–12 Hz, it varies to a considerable extent even in healthy and age matched subjects with a standard deviation of about 1 Hz (Klimesch 1996). This variation has been related to genetic determination (Smit et al. 2006; Malone et al. 2014). For these reasons, adjusting the alpha frequency window for each participant could avoid EEG spectral power distortion. This approach prevents not only to lose part of that alpha rhythm distribution which falls outside the fixed range, but also to include activity from theta and beta adjacent frequencies. It is suggested that applying the individual alpha frequency range (IAF), computed for each subject from the maximum spectral peak alpha frequency (PAF) or from the center of gravity (Klimesch, Schimke, et al. 1993), helps to assess the alpha rhythm activity properly (Klimesch 1999; Haegens et al. 2014).

In this framework, few previous studies about MP effects on EEG activity applied the IAF in order to compute the EEG frequency analysis. It has been reported that MP exposure could

modulate cognitive performance, decreasing the peak amplitude of event-related IAF desynchronization (Vecchio, Buffo, et al. 2012), while other studies on EEG spectral coherence reported a significant modulation of the interhemispheric functional coupling of IAF (Vecchio et al. 2007; Vecchio et al. 2010; Vecchio, Tombini, et al. 2012).

Moreover, power spectral densities of resting EEG could vary between subjects because of environmental and endogenous factors. For example, several external factors could affect the alpha band power spectral density, such as nicotine (Teneggi et al. 2004; Domino et al. 2009), alcohol (Lithari et al. 2012; Rosen et al. 2014) and caffeine consumption (Siepmann and Kirch 2002; Deslandes et al. 2005) or emotional stress behavior (Lewis et al. 2007; Al-Shargie et al. 2016) and physiological hormonal variation, such as melatonin (Cajochen et al. 1998), cortisol (Sannita et al. 1999; Tops et al. 2005) and female ovarian hormones (Becker et al. 1982; Brötzner et al. 2014). In addition, the sleep-wake cycle could determine the level of alertness, modulating the waking EEG and the alpha band amplitude (Cajochen et al. 1995; Lafrance and Dumont 2000; Regen et al. 2013; Lees et al. 2018). Some of previous studies which investigated the RF-EMF effect on the waking EEG considered abovementioned factors, in order to limit potential interaction between these confounding factors and EEG modulation (for review see (Danker-Hopfe et al. 2019; Wallace and Selmaoui 2019)).

In this study, healthy volunteers were recruited and targeted considering strict inclusion criteria to limit bias with the aim to investigate the EEG at rest before and during GSM (global system for mobile) MP RF-EMF exposure. We restricted the EEG analysis to the alpha band, using both the usual fixed range of 8–12 Hz and the IAF computed for each participant, in order to verify if the individual EEG power spectra could contribute to RF-EMF effect, partially explaining heterogeneous results in literature. So, we assume that the shift of the alpha band according to the calculated IAF could participate in the differences of detected effects mentioned above, knowing that RF-EMF exposure seems to affect in a different way between

studies (increase or decrease) the EEG alpha band power as well as EEG power of adjacent bands. To our knowledge, the IAF power spectral density has not been investigated so far with a resting-state EEG protocol as a possible individual sensitivity factor representing the variability of the physiological state of the brain between subjects when measuring EEG activity.

SUBJECTS AND METHODS

Subjects

Twenty-one healthy volunteers were included in the experiment (10 females and 11 males; mean age \pm SD: 25.1 ± 3.6 years; mean body-mass index \pm SD: 21.5 ± 1.9). They were right-handed with normal or corrected to normal vision. All volunteers underwent a clinical interview to assess inclusion criteria to exclude any history of head injury, neurological or psychiatric disease or any chronic disease, disability or recent acute illness during last month. Inclusion criteria comprehended also regular sleep habits with a regular sleep-wake cycle (8 a.m.–11 p.m. \pm 1 h), no medication, no smoking and no-drug use. No-drug use was confirmed by a urine multi-drugs test (NarcoCheck Evolutive, France), in order to detect tetrahydrocannabinol, cocaine, crack cocaine, opiates and opioids (morphine, heroin, fentanyl, methadone, buprenorphine), ketamine, lysergic acid diethylamide, alpha-methylphenethylamine, 3,4-methylenedioxymethamphetamine, benzodiazepines, barbiturate and tricyclic antidepressants. Pregnant women were not included in the study (confirmed by urine pregnancy test, NADAL hCG Pregnancy test 10 mIU/ml, nal von minden GmbH, Germany). Participants were instructed to abstain from consuming alcohol and caffeine for 24 hours before each experimental session, to maintain the regular sleep-wake and not to use their personal MP on the days of the experiment. Participants had been fasting for at least two and half hours before the recordings. To avoid potential confounding effect of cyclical ovarian

hormonal impact on cerebral electrophysiology (Becker et al. 1982; Brötzner et al. 2014; Bazanova et al. 2017), all women participated to the experimental sessions during the follicular phase of their menstrual cycle. A written informed consent was obtained from participants before the investigation. Participants were compensated for their participation. All procedures were approved by the French Ethics Committee CPP Ouest VI (ID n°: RCB 2017-A01702-51) and were in accordance with the Declaration of Helsinki.

Radiofrequency electromagnetic field exposure setup

The RF-EMF exposure system was constituted by two identical commercial dual-bands GSM MPs (Nokia 6650), which differ only for the RF-EMF emission, in order to realize a double-blind design. The sham or the real exposure was carried out using a “load” or a “dummy load” of the MP, respectively. For this purpose, an external power load was connected to the remote antenna connector of the phone. A 50-ohm resistive load and an open-circuit dummy load were developed for sham/exposed conditions with the same shape and structure in order to provide the double-blind protocol of the study.

A specific experimenter, different from the experimenter who carried out the protocol with participants, was in charge of planning sham or real exposure and setting the frequency and the RF-EMF power of MPs, with a counterbalanced and randomized order. With this purpose, before starting the exposure phase, and in order to set the standard exposure parameters, the phone was connected to a personal computer to control the required frequency and RF power by service software (Phoenix, Nokia Corp., Finland). 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. After setting, they were disconnected from the personal computer. Then, for the exposure phase the real or the sham MP was placed against the left ear using a tubular tissue bandage, according to a counterbalanced and randomized order.

Radiofrequency electromagnetic field dosimetry

Measurements of the RF-EMF power absorbed by the head, defined by the specific absorption rate (SAR), were performed in a standard SAM phantom (Specific Anthropomorphic Mannequin Phantom, Antennessa, France) filled with standard liquid equivalent to brain tissue (Satimo, France) according to the IEC standard document (IEEE/IEC62209-1528-2020). Dosimetry was performed by using an experimental approach, by measuring the electric field in the phantom at the WHIST laboratory (Mines-Télécom Institute, Paris, France). A small electric field probe (O6-EP64, Satimo, France) connected to a microvoltmeter (Keithley, Cleveland, OH, USA) was used to measure the electric field intensity inside the liquid. The liquid was characterized using an open coaxial cable immersed in the phantom liquid and connected to a vector network analyzer (Wiltron 360B, USA). The phone was positioned in contact with the phantom in the same way as the study protocol. The displacement of the electric field probe was performed by an XYZ positioning system slaved to a motor of the 3D "step-by-step" robot (Charlyrobot SA, France). The maximum SAR of the real MP averaged on 10 g tissue, 1 g tissue, and the peak value were measured at 0.49 W/kg, 0.70 W/kg, and 0.93 W/kg, respectively. The SAR of the sham phone was below the detection level of the system (0.001 W/kg) at any position of the phantom, neither any electric field was detected on the surface of the sham MP. Standard errors were less than 20%, comparable to the work of Beard et al. (Beard et al. 2006).

A polystyrene phantom head test was realized, with the purpose of avoiding interferences between RF-EMF and EEG recordings during the exposure phase. Results showed that MPs did not affect EEG signals during exposure (for more details see (Ghosn et al. 2015)).

Experimental protocol

Subjects underwent to two experimental sessions at weekly interval at the same time of the day, to avoid confounding effects related to circadian rhythms, with a crossover, randomized,

double-blind and counterbalanced experimental design. Recordings started at about 11.30 a.m. or 3.30 p.m. for each experimental session scheduled in the morning or in the afternoon, respectively. Participants were instructed to stay still and to fixate a common fixation point during recordings. Recording sessions 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. Then, the EEG recordings continued in the same room for 25 minutes and 30 seconds of exposure phase with sham or real RF-EMF exposure (from Run 5 to Run 8). EEG recordings were realized in a sound attenuating shielded room, where subjects were comfortably seated staring a central black fixation mark (black dot surrounded by a black circle) presented on a gray background at a viewing distance of 1 m. Finally, EEG recordings combined with MEG were carried out for 25 minutes and 30 seconds of post-exposure with no RF-EMF exposure (from Run 9 to Run 12). EEG recordings combined with MEG during baseline and post-exposure phases will not be analyzed and not reported in the current paper. All runs were constituted by 3 minutes of eyes-open (EO) and 3 minutes of eyes-closed (EC) resting-state recordings in alternation and at the end of each run there was a pause of 30 seconds. During the whole duration of the protocol, participants were monitored in real-time via a camera in order to verify their vigilance and adherence to vocal instructions. All participants respected vocal instructions during both experimental sessions.

Electrophysiological data acquisition

EEG data were recorded using 74 EEG-channel system with Ag/AgCl sensors (Easycap Products GmbH, Germany) placed according to the standard 10/10 system. The EEG signal was referenced to mastoid signal and one ground electrode was placed on the left scapula. EEG data were recorded with a sampling frequency of 1 kHz and a bandwidth of 0.016-250 Hz. The

electrical impedances were kept lower than 20 k Ω and were verified before starting recording for each run. To monitor ocular movements, the horizontal electro-oculogram (EOG) was recorded with two electrodes positioned near the outer canthi of each eye, while the vertical EOG was recorded with electrodes positioned above and below the right eye. The electrical impedances of mastoid signal, ground signal and EOG were kept below 5 k Ω and were verified before starting each run recording. All data were acquired with BrainVision BrainAmp MR plus Amplifiers (Brain Products GmbH, Germany).

EEG data preprocessing

Resting EEG data of baseline phases (Run 3 and Run 4) and exposure phases (from Run 5 to Run 8) were analyzed using MNE-Python, Version 0.19.2 (Gramfort et al. 2013). Electrodes near to ear-slits of the EEG cap (i.e. T9, TP9, T10, TP10) were removed from all data analysis for all subjects. The signal was band-pass filtered between 1–20 Hz using Finite Impulse Response (FIR) filter design with Hamming window. Independent component analysis (ICA) with FastICA algorithm was performed to decompose EEG signal and reject ocular artefacts. Pearson correlations were used to find vertical and horizontal EOG related components and bad epochs were created 500 ms before and after each blink, with band pass filtering between 1 and 10 Hz.

Individual alpha frequency quantification

The IAF was determined for each subject, as follow. The PAF was computed using a non-parametric automated technique (Corcoran et al. 2018). The PAF was obtained from 2 minutes of artifact-free baseline resting-state recordings with closed eyes (Run 3 and Run 4) for each experimental session. It was computed from occipital electrodes (PO9, PO7, PO3, POz, PO4, PO8, PO10, O1, Oz, O2, O9, Iz, O10) between 6 and 14 Hz with a resolution of 0.10 Hz. For each subject the PAF was averaged across baseline runs and sessions. Then, according to

(Vecchio, Buffo, et al. 2012), the IAF range was defined for each subject as $PAF \pm 2$ Hz.

EEG power spectral density analysis

Resting EEG artefact-free data were analyzed for 2 minutes and 30 seconds of recording periods with EO and EC, separately. A Fast Fourier Transformation (Welch technique, Hamming windowing function, 4096 points per window, 50 % of overlap between segments) was applied to EO and EC periods to obtain spectral power values for each EEG channel. Log-transformed power values were calculated to approach a normal distribution for the alpha band with usual frequency range (8–12 Hz) and for the IAF. Finally, log-transformed power values of the alpha band (8–12 Hz) and IAF were averaged over the baseline period and the exposure period for all subjects, considering EO and EC conditions separately. To obtain baseline-corrected exposure data for all subjects, baseline log-transformed data computed for each electrode were subtracted from exposure log-transformed data computed for each electrode, considering EO and EC conditions separately.

STATISTICAL ANALYSIS

Firstly, a three-way repeated-measures ANOVA was applied to power values computed for the alpha band (8–12 Hz) and the IAF, separately, in order to determine the effect of the period (two levels: baseline, exposure), eyes condition (two levels: EO, EC) and exposure condition (two levels: sham exposure, real exposure). Then, to further characterize RF-EMF exposure effects, we carried out a one-way repeated measures ANOVA on baseline-corrected exposure powers computed for each electrode for the two exposure conditions. The analysis was applied on alpha band (8–12 Hz) and IAF powers for EO and EC data, separately. We compute F and p-values for each electrode on the scalp using a custom MATLAB (R2017b) software toolbox (Maris and Oostenveld 2007; Ducorps et al. 2010). Statistical significance was set for $p < 0.05$.

RESULTS

Individual alpha frequency interval

The mean of PAF values computed individually during Run 3 and Run 4 of both experimental sessions at rest with eyes closed was 9.78 Hz (± 1.06 Hz SD). So, the IAF interval mean was 7.78–11.78 Hz (± 1.06 Hz SD). The PAF values computed for each participant are shown in **Table 1**.

Alpha spectral power results

Results of three-way repeated measure ANOVA showed statistically significant differences of both alpha band (8–12 Hz) and IAF for period (two levels: baseline, exposure) and eyes condition (two levels: EO, EC) factors. In particular, for both frequency ranges there were statistically significant differences between eye conditions on the whole scalp ($p < 0.001$), while the period factor showed a statistically significant difference between the baseline and the exposure on all centroparietal, temporoparietal, parietal, occipitoparietal and occipital electrodes ($p < 0.01$). A statistically significant interaction between the period factor (two levels: baseline, exposure) and eyes condition factor (two levels: EO, EC) was found for all prefrontal and frontal electrodes ($p < 0.01$).

The baseline-corrected alpha band (8–12 Hz) power computed in the EO condition showed higher values at prefrontal, frontal, parietal, parieto-occipital and occipital electrodes during the real exposure session relative to the sham exposure session (**Figure 1A**). However, results showed no statistically significant difference between sham and real exposure conditions. In the EC condition, the baseline-corrected alpha band (8–12 Hz) power showed lower values mainly at frontal and parieto-occipital electrodes during the real exposure compared to the sham exposure (**Figure 1B**), without statistically significant difference between RF-EMF exposure conditions.

The baseline-corrected power computed for the IAF during EO recordings showed similar results to the alpha band (8–12 Hz) power, with no statistically significant difference between exposure condition (**Figure 2A**). With the EC condition, the power values were lower at frontal and parieto-occipital electrodes during the real exposure session relative to the sham (**Figure 2B**), but no statistically significant difference between sham and the real exposure was found.

DISCUSSION

The present study evaluated the effect of MP exposure on the electrical activity of the human brain. We studied the effect of 900 MHz RF-EMF exposure related to MP on the alpha oscillations of healthy volunteers at rest in a double-blind, randomized, counterbalanced, crossover design. As reported elsewhere, the alpha band seems to be the most impacted oscillations of spontaneous EEG by MP exposure at rest but results remain elusive (Danker-Hopfe et al. 2019; Wallace and Selmaoui 2019). Since previous studies argued that MP could have different effect on subjects, depending on the physiological state of the brain (Hinrikus, Bachmann, Lass, Karai, et al. 2008; Loughran et al. 2013), the purpose of this study was to investigate RF-EMF effect on spontaneous EEG alpha rhythm activity, analyzing not only the usual fixed range of 8–12 Hz but also the IAF, as possible inter-individual EEG variability factor. Indeed, even though the alpha oscillation is generally analyzed in the fixed range, the individual dominant peak or gravity of EEG alpha frequency is characterized by variability at rest or during cognitive performance (Chiang et al. 2011; Haegens et al. 2014).

Participants were exposed for about 26 minutes to GSM MP signal at 900 MHz, which represent the second-generation technology signal of cellular mobile telecommunication system (2G). Even though further generation of MP technology signals have been introduced, up to the fifth-generation (5G), nowadays 2G technology is still largely used worldwide (GSMA 2018). It is noteworthy that according to the INTERPHONE study (INTERPHONE-

Study-Group 2010) only a group of users reporting to have phoned more than 1640 hours had a significant risk of glioma. The cutoff point for the top 10% of users (1640 cumulative hours of use over the lifetime), spread over 10 years, is about half an hour per day. We deemed then that the duration of about 26 minutes that we applied, when considering the heavy experiment for recording EEG, is close to half hour that might have an effect on brain physiology and electrical activity.

Our findings showed a statistically significant difference between session periods (baseline, exposure), between eyes condition (EO, EC) and a significant interaction between the period and the eyes condition, for spectral power of both alpha frequency ranges. These findings could reflect a modification in the attention and fluctuations of vigilance during experimental recording periods. So, to further characterize the exposure effect, we compared the baseline-corrected power of the two exposure conditions. Powers computed for both alpha frequency intervals showed higher values over the scalp during the real RF-EMF exposure relative to the sham when subjects had their eyes open, while we observed lower power density values at frontal and posterior regions when the eyes were closed. In general, these results together could suggest a dissimilar reaction of alpha power to RF-EMF exposure depending on EO or EC recording conditions, but there were no statistically significant differences between real and sham exposures for both alpha frequency intervals. So, findings showed that the RF-EMF exposure of about 26 minutes did not determine a statistically significant effect on the alpha power with EO or EC, neither on the usual frequency range (8–12 Hz) nor on the IAF. It is worth noting that the eyes condition applied in previous EEG studies on RF-EMF effects is an important parameter that differs between protocols. Moreover, most of previous studies applied just one eyes condition, EO or EC, for the whole duration of the experimental EEG recording session (Danker-Hopf et al. 2019; Wallace and Selmaoui 2019). However, since modifications on EEG and alpha rhythm could be related to the eyes condition (Chen et al. 2008), it is recommended to include periods with EO and EC as an important means of

assessment (Sinha et al. 2016).

Further studies should be realized to measure different effects of RF-EMF on EO and EC EEG resting-state, considering also the post-exposure period. Indeed, even if we did not find any significant effect of the MP during the exposure, several previous studies reported an effect detectable after the RF-EMF exposure (Reiser et al. 1995; Von Klitzing 1995; Lebedeva et al. 2000; Huber et al. 2002; Regel, Gottselig, et al. 2007; Vecchio et al. 2007; Vecchio et al. 2010; Vecchio, Buffo, et al. 2012; Ghosn et al. 2015; Hinrikus et al. 2017; Vecsei et al. 2018) and a delay in the onset of EEG modulation after exposure has been suggested (Reiser et al. 1995). Moreover, the persisted effect of RF-EMF on EEG was also observed during sleep, with EEG power modulation following the active period of exposure (Huber et al. 2002; Loughran et al. 2005; Regel, Tinguely, et al. 2007).

It should be recalled that use of different EEG references, different numbers of electrodes, and different topographical clustering may contribute to the heterogeneous results of previous studies (Danker-Hopfe et al. 2019), making difficult to highlight a conclusive effect of RF-EMF exposure on a specific brain region. Normally, the temporal lobe presents the highest average relative SAR with RF-EMF frequencies between 900 MHz and 1800 MHz when the MP is positioned near the ipsilateral phantom cheek position, as showed by dosimetry measurements (Cardis et al. 2008). These studies further revealed that the average relative SAR decrease with increasing depth in brain anatomical layers, particularly at higher frequencies (Cardis et al. 2008). Indeed, the penetration depth depends on the dielectric properties of the medium, as well as frequency. As frequency increases, the penetration depth decreases, and is predominantly within the surface tissues at frequencies higher than about 6 GHz (ICNIRP 2020). The lowest level of SAR at which the effect in EEG related to RF-EMF exposure was detected is 0.003 W/kg, with a microwave exposure at 450 MHz and a field strength of 2.45 V/m (Hinrikus et al. 2021). On the other hand, the use of Bluetooth hands-free kits has

been shown to reduce the level of exposure with a GSM mobile phone (Wall et al. 2019). Based on these dosimetry studies, we could assume a mainly effect of the MP on the electrical brain activity localized on the ipsilateral temporal lobe (e.g., temporal gyrus), when the exposure system is placed near the ear. In accordance with the CENELEC standard EN 50361 (CENELEC 2002), in our study the MP was placed so that its longitudinal axis followed an imaginary line from the entrance to the ear canal to the corner of the mouth during the exposure phase (with a SAR peak value of 0.93 W/kg), but our findings did not show any modulation of the EEG alpha band activity depicted by temporal electrodes, neither by electrodes on other scalp regions. However, previous study on healthy volunteers found a modulation of the interhemispheric frontal and temporal functional coupling of EEG alpha band related to the exposure to GSM MP, which was positioned near the head in the parieto-temporal areas miming the MP normal position for use (Vecchio et al. 2007). We could assume that any EEG activity modulation on a specific brain region (as temporal region) related to the RF-EMF exposure could be highlighted using also other technical approaches. Indeed, on one hand EEG allows to measure neuronal processes in the time frame in which these processes occur, but on the other it suffers from poor spatial resolution making difficult the location of the brain areas generating the neuronal activity measured on the scalp (Michel and Brunet 2019). For example, a functional magnetic resonance imaging study revealed that exposure to fourth-generation Long-Term Evolution Electromagnetic field seems to induce changes in the amplitude of spontaneous low frequency fluctuations in the temporal gyrus and modulate brain functional intra- and inter-regional connectivity, including temporal brain regions (Wei et al. 2019). Furthermore, the high-density EEG systems (Michel and Brunet 2019) or magnetoencephalography (da Silva 2013) combined with a precise information of the head anatomy could help to obtain accurate source localization to estimate the distribution of the underlying neuronal sources.

In order to explore the impact of the alpha band frequency range in the study of MP exposure

effects, we assessed the alpha band power spectral density in the fixed range of 8–12 Hz, as well as the IAF power spectral density. In general, peak frequency and center of gravity frequency are the most commonly used measures to calculate alpha frequency (Klimesch, Pfurtscheller, et al. 1993). The first one reflects the frequency at which the power value is the greatest with no notions about the shape of the alpha peak. The second one, reflects the entire shape of the alpha peak within a selected frequency window. According to Klimesch et al. (Klimesch, Pfurtscheller, et al. 1993), we measured the IAF from baseline data for each participant considering the peak frequency, since during a resting period a pronounced alpha peak can be expected with no drastic amplitude modulation correlated to mental tasks. Results of IAF showed that the alpha band could varies between subjects with a standard deviation of about 1 Hz, as expected (Klimesch 1996). Anyway, since power of both alpha frequency intervals showed no differences between real and sham exposure recordings, we could exclude that IAF could contribute to RF-EMF effect as electrophysiological variability factor. It should be noted that volunteers were recruited according to strict inclusion criteria and participated to two experimental sessions following protocol instructions to limit confounding factors.

Recently it has been described that the overuse of MPs (i.e. smartphone) could affect the sleep quality and determine anxiety, stress and addiction behaviors (for review see (De-Sola Gutiérrez et al. 2016; Vahedi and Saiphoo 2018), with a great impact on brain pathways, memory performance and cognitive control (Gardner 2011). MPs addiction analysis could be an interesting tool for inter-variability assessment for further studies about MP effects on human brain activity and the RF-EMF individual sensitivity hypothesis.

Even though several studies suggested that MP can modulate spontaneous EEG, there are apparent inconsistencies between studies that limit conclusive statements in this area (Zhang et al. 2017) and the mechanism behind biological effects induced by the RF-EMF non-thermal exposure need further investigations.

CONCLUSION

In conclusion the current study reported no alpha band activity modulation during resting-state under RF-EMF and did not provide evidence for a different sensitivity to MP related to inter-individual variability of the frequency of the alpha rhythm. Since the reason why the alpha band reacts differently between studies is still unclear, further research on EO-EC resting EEG should be carried out in order to investigate the hypothesis of individual differences to MP exposure.

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Disclosure statement

The authors declare no competing interests.

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Table 1.

The PAF values computed individually from occipital electrodes (PO9, PO7, PO3, POz, PO4, PO8, PO10, O1, Oz, O2, O9, Iz, O10) for twenty-one participants during Run 3 and Run 4 of both experimental sessions at rest with eyes closed.

SUBJECT	PEAK ALPHA FREQUENCY (Hz)			
	Session 1		Session 2	
	Run 3	Run 4	Run 3	Run 4
1	11.70	11.20	11.60	11.80
2	7.40	8.00	6.60	9.50
3	10.20	10.80	10.50	7.80
4	9.50	9.80	8.90	9.00
5	9.30	9.40	9.30	9.40
6	10.50	10.70	10.80	10.80
7	9.70	9.80	9.50	9.50
8	11.30	11.20	11.20	11.40
9	10.10	10.50	10.50	10.40
10	7.40	8.10	7.00	10.70
11	10.10	10.40	9.50	10.10
12	12.30	11.50	12.60	10.50
13	9.60	10.00	8.70	9.60
14	10.80	11.00	10.80	11.40
15	9.60	10.60	9.50	10.00
16	8.30	8.90	8.90	9.10
17	10.10	10.00	10.40	10.50
18	8.60	8.40	9.20	9.10
19	10.20	9.90	8.50	6.10
20	9.10	8.60	9.40	8.80
21	6.00	11.30	10.50	10.50

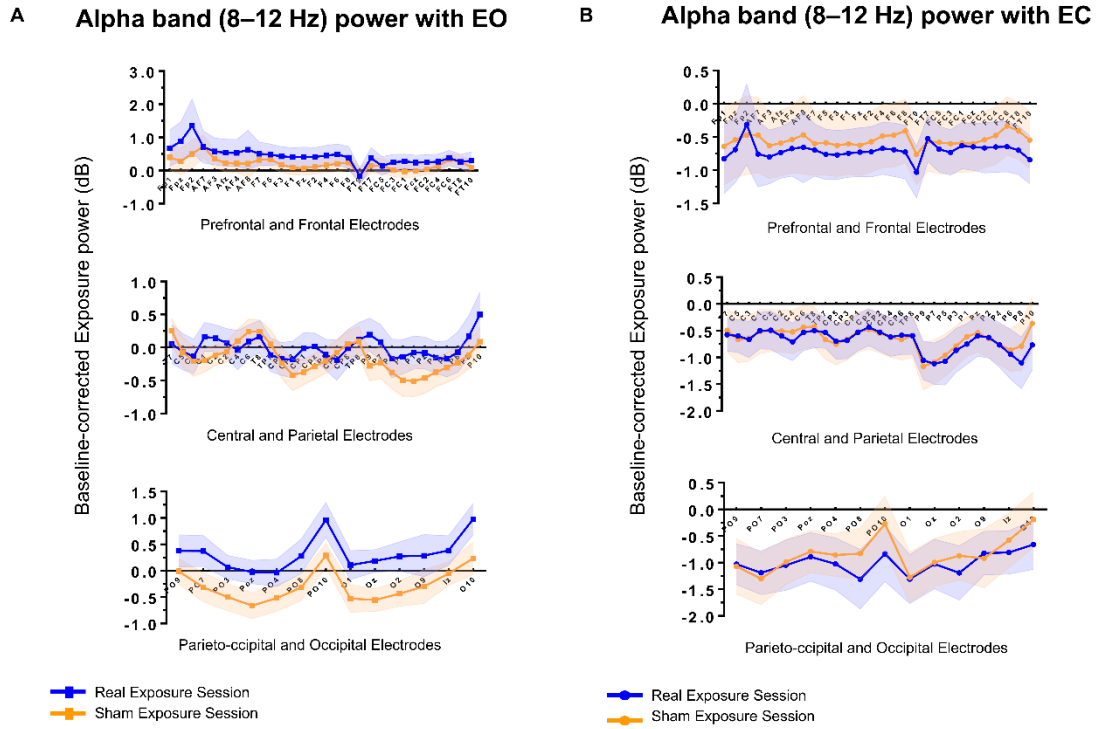
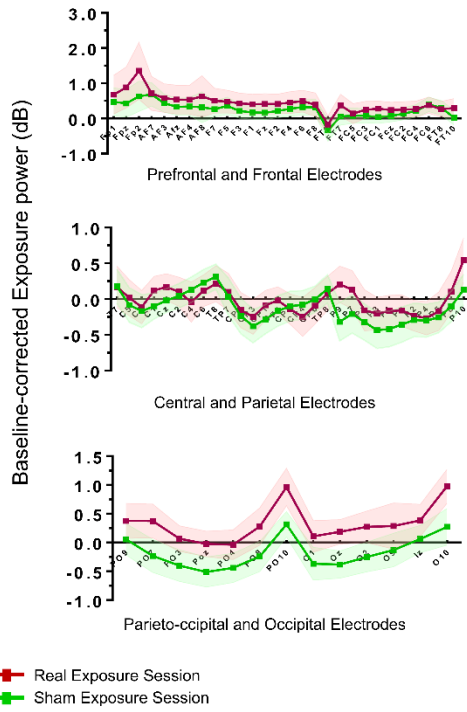


Figure 1. Baseline-corrected powers computed for the alpha band (8–12 Hz) during EEG recording of real and sham exposure sessions with eyes open (**A**) and eyes closed (**B**). Powers were computed for each electrode and averaged across subjects and across exposure runs. Results are shown as mean \pm SEM.

EO: eyes open; EC: eyes closed

A IAF power with EO



B IAF power with EC

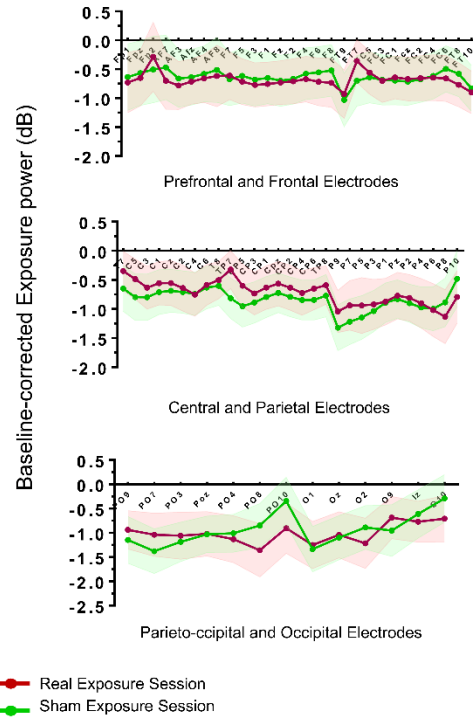


Figure 2. Baseline-corrected powers computed for the individual alpha frequency range (IAF) during EEG recording of real and sham exposure sessions with eyes open (A) and eyes closed (B). Powers were computed for each electrode and averaged across subjects and across exposure runs. Results are shown as mean \pm SEM.

EO: eyes open; EC: eyes closed