Bioelectromagnetics

Heart Rate Variability Affected by Radiofrequency Electromagnetic Field in Adolescent Students

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This study examines the possible effect of radiofrequency (RF) electromagnetic fields (EMF) on the autonomic nervous system (ANS). The effect of RF EMF on ANS activity was studied by measuring heart rate variability (HRV) during ortho-clinostatic test (i.e., transition from lying to standing and back) in 46 healthy grammar school students. A 1788 MHz pulsed wave with intensity of 54 ± 1.6 V/m was applied intermittently for 18 min in each trial. Maximum specific absorption rate (SAR_{10}) value was determined to 0.405 W/kg. We also measured the respiration rate and estimated a subjective perception of EMF exposure. RF exposure decreased heart rate of subjects in a lying position, while no such change was seen in standing students. After exposure while lying, a rise in high frequency band of HRV and root Mean Square of the Successive Differences was observed, which indicated an increase in parasympathetic nerve activity. Tympanic temperature and skin temperature were measured showing no heating under RF exposure. No RF effect on respiration rate was observed. None of the tested subjects were able to distinguish real exposure from sham exposure when queried at the end of the trial. In conclusion, short-term RF EMF exposure of students in a lying position during the ortho-clinostatic test affected ANS with significant increase in parasympathetic nerve activity compared to sham exposed group. Bioelectromagnetics. © 2018 Wiley Periodicals, Inc.

Keywords: autonomic nervous system; cell phone; RF EMF exposure; mobile communication; ortho-clinostatic test

INTRODUCTION

Given the steep rise in the use of cell phones, WiFi transmitters, and other microwave devices, the biological effects of radiofrequency (RF) electromagnetic fields (EMF) have been widely discussed. Nowadays, every adolescent student seems to be an active cell phone user. Thus, the RF EMF source is often held very close to the head, enabling possible biological interactions.

There is still limited knowledge on the effects of RF EMF on the autonomic nervous system (ANS), heart rate (HR), blood pressure (BP), and other cardiovascular functions [Parazzini et al., 2007]. Some studies provided evidence that RF EMF might

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have a negative effect on brain function in animals [Paulraj and Behari, 2002] and isolated hearts' cardiac cycle [Yee et al., 1984]. It has been reported that RF EMF can influence HR [Havas et al., 2010], BP [Szmigielski et al., 1998], and heart rate variability (HRV) in humans [Bortkiewicz et al., 2006; Nam et al., 2006; Wilen et al., 2007; Andrzejak et al., 2008]. However, these results are still controversial. While HR was not affected by exposure while sleeping [Mann et al., 1998; Huber et al., 2003], diverse effects were found in conscious subjects: an increase in sympathetic nerve tone associated with an increase in BP [Braune et al., 1998]. In contrast, other authors did not observe these effects [Braune et al., 2002; Tahvanainen et al., 2004]. This variability in study outcomes may be accounted for in various conditions of exposure, given that RF effects were reported to be dependent on multiple physical and biological variables [IARC, 2013].

HRV is defined as oscillations around HR mean value. These oscillations are directly influenced by the sympathetic or parasympathetic parts of ANS. Thus, HRV can reveal gentle functional changes in sympathetic-parasympathetic ANS control. In particular, linear analysis of HRV in both time and frequency domain (spectral analysis) may provide valuable information about sympathetic and parasympathetic nerve activity (sympathovagal balance).

Ortho-clinostatic test (a slow transition from lying to standing and back to lying position) may provide a better possibility to reveal eventual effects on HRV. Indeed, transition from standing to lying stimulates ANS activity, which may be affected by RF EMF exposure. For example, Parazzini et al. [2007, 2013] exposed volunteers to RF and found effects only in standing position using orthostatic test. Thus, we aimed to study the effects of RF EMF on ANS through monitoring HRV online in ortho-clinostatic test. To this aim, 46 grammar school students were exposed and sham exposed to RF EMF. The exposure setup was developed to provide conditions similar to real exposures to near-field uplink signal.

MATERIALS AND METHODS

Study Group

Forty-six adolescent students (16 male and 30 female) in good health without acute or chronic diseases (e.g., flu, arterial hypertension, or menstrual period in females) and not using any medication were enrolled in this study. The participants were asked to avoid smoking and drinking alcoholic and caffeinated beverages at least 24 h before the trial. They were also instructed to have just a light breakfast on the day of the trial. Twenty-three participants were both sham exposed and exposed to RF EMF while the other 23 participants were sham–sham exposed (Table 1). To avoid any cumulative effects that might occur, these groups did not include the same people. The strength of the study group was that it was homogeneous in both age and body mass index (BMI) (Table 1). After the trial, the participants were asked to describe their subjective feelings to distinguish between sham and real exposure to RF EMF.

All procedures on human subjects were conducted in accordance with the Declaration of Helsinki and were permitted by the Ethical Board of Jessenius Faculty of Medicine in Martin, Comenius University in Bratislava, and by the laws of the Slovak Republic and European Union under permissions covered by the project APVV-0189-11 (Prof. Jakus).

RF EMF Exposure System

The 1788.0 MHz pulse modulated wave with a duty cycle of 50% and pulse width of 100 μs (Fig. 1) was generated by functional signal generator Agilent N9310A (Agilent Technologies, Santa Clara, CA) and then amplified with a 5 W amplifier 5S1G4 (Amplifier Research, Souderton, PA) set to 2 W. The frequency of this signal was chosen because it is close to and does not disturb Digital Cellular System (DCS-1800) prepaid uplink bandwidth, which is set at the 1710.1–1776.1 MHz band by the regulatory authority for electronic communications and postal services of the Slovak Republic for Slovak mobile network providers [TELE-OFF, 2014]. Intensity of the electric field (E) was monitored by Narda 550NBM high-frequency broadband meter (Narda Safety Test Solutions, Pfullingen, Germany). The International Commission for Non-Ionizing Radiation Protection (ICNIRP) has set an E field value of 58.34 V/m rms (root mean square) as a safety standard for the 1800 MHz band [ICNIRP, 1998]. In our study, the E field was slightly lower at 54 ± 1.6 V/m (mean ± SD; averaged through all trials).

We used two identical loop antennas to expose students at different positions during ortho-clinostatic test (Fig. 2) [Galuscak and Hazdra, 2008]. Each antenna was comprised of one λ long dipole coiled into a circular shape (Fig. 1). The antenna's base was manufactured from aluminum alloy and represented a rugged cup-shaped reflector, which helps to improve the front-to-back radiation ratio (Fig. 3). The outer radius of base was 16 cm. Each antenna was fixed on a tripod stand 30 cm (approximately 2λ) from the right side of the student's face (Fig. 2) in the middle of a main radiation lobe (Fig. 3).

TABLE 1. Physiological Characteristics Of 23 Students Subjected to Sham-Real (A) and Sham-sham (B) Exposures (Mean ± SD)

Physiological parameter	Before the trial	After the trial	*P*-value
A			
Men:women	8:15		
Age [years]	18.2 ± 1.1		
Age (range) [years]	16.7–23.3		
BMI [kg/m^2]	21.5 ± 2.3		
Head diameter[a] [cm]	18.5 ± 1.8		
Tympanic temperature [°C]	36.1 ± 0.4	35.9 ± 0.6	0.3
Blood pressure (Sys.) [mmHg]	118.9 ± 13.9	113.7 ± 11.2	0.2
Blood pressure (Dias.) [mmHg]	66.8 ± 8.2	63.3 ± 8.5	0.2
B			
Men:women	8:15		
Age [years]	18.3 ± 1.6		
Age (range) [years]	16.8–23.3		
BMI [kg/m^2]	20.2 ± 1.7		
Head diameter[a] [cm]	18.4 ± 0.4		
Tympanic temperature [°C]	36.6 ± 0.4	36.6 ± 0.6	0.7
Blood pressure (Sys.) [mmHg]	119.5 ± 11.6	118.7 ± 10.3	0.6
Blood pressure (Dias.) [mmHg]	70.4 ± 9.2	69.0 ± 8.9	0.4

[a] Head diameter calculated from perimeter.

During sham–sham exposure, the RF field was off while all RF EMF equipment (antennas, EMF generator, and amplifier with wiring) were at the same position as during EMF exposure. MW switching relay was used to choose the output antenna in each position.

Previous studies have suggested that extremely low frequency (ELF) magnetic background field, usually in the μT–mT range, may interfere with the effects of RF EMF [Belyaev, 2010]. At the location of exposure/sham-exposure, background ELF magnetic flux density did not exceed 81 nT for the frequency band of 5–100 Hz, 58 nT for 100–10 kHz, or 39 nT for 10–100 kHz as measured with Narda EHP50-D (Narda Safety Test Solution, Pfullingen, Germany). RF EMF effects were also reported to be dependent on static magnetic field (SMF) [Belyaev, 2010]. Hence, the horizontal and vertical components of SMF were 12 ± 3 μT and 41 ± 2 μT, respectively, at the location of exposure/sham-exposure.

SAR Determination

he experimental setup was numerically simulated. The antenna was modeled based on measurements of the disassembled unit and consisted

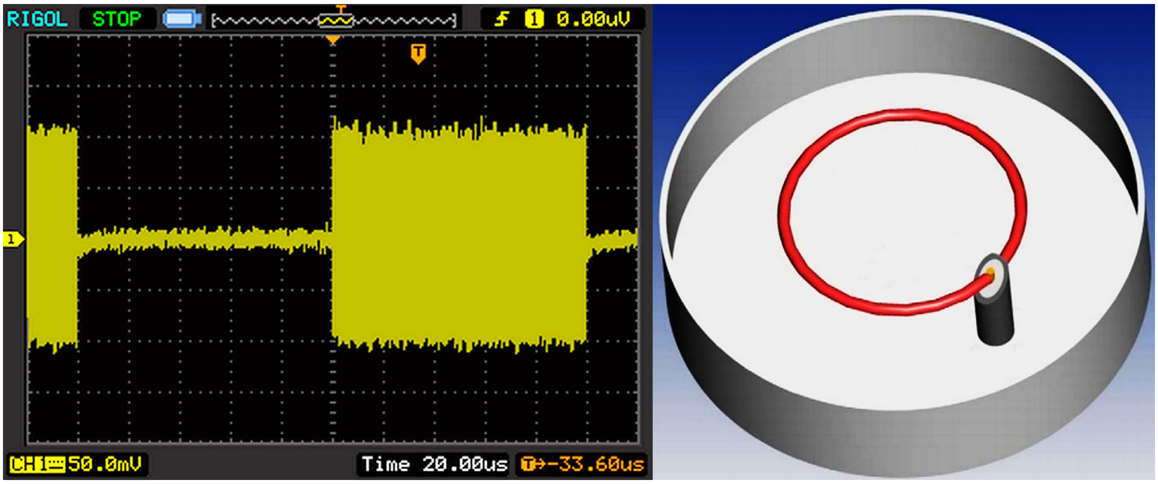

Fig. 1. Left panel: Exposure signal: 1788 MHz, 100 μs pulse width, 50% duty cycle. Right panel: one lambda (λ, refers to wavelength) long radiator coiled into circular shape with rugged cup-shaped reflector (with outer diameter 16 cm) was used as antenna in exposure system [Galuscak and Hazdra, 2008].

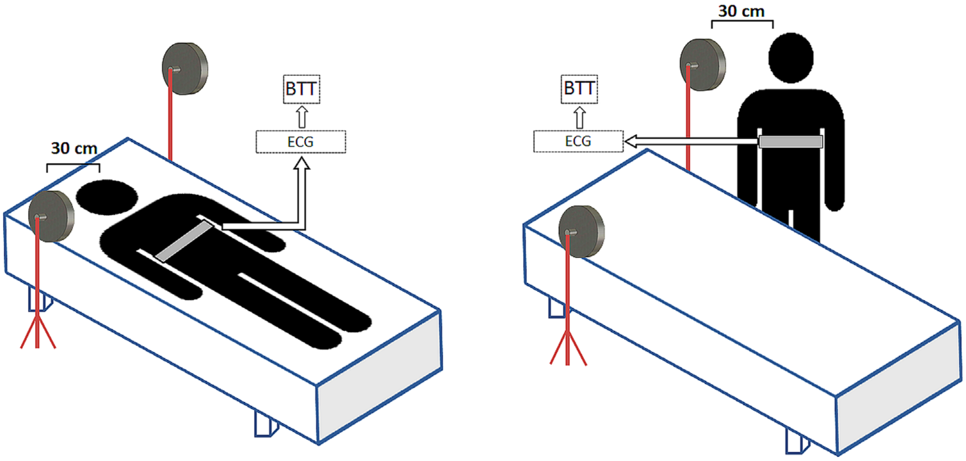

Fig. 2. Each student was exposed to RF with antenna fixed by a tripod stand at 30 cm distance from student's right face while lying (left side) and standing (right side). ECG belt was attached on student's chest and wired to Bluetooth transducer.

principally of a circular radiating element with a diameter of one λ and a circular reflector element with a diameter of 16 cm. We used 2 mm resolution AustinMan Electromagnetic Voxel v2.5 phantom [Massey and Yilmaz, 2016] to evaluate both E-field distribution and SAR_{10g} value in areas of interest (sagittal and coronal plane of the head). The antenna was placed 30 cm away from the right face of the phantom, and reference power was set to 2 W. According to numerical simulation, maximal SAR_{10g} value reached 0.405 W/kg (Fig. 4). This method provided comprehensive information about E-field and SAR spatial distribution.

Temperature Measurements

While RF thermal effects are well known, RF exposure could also result in so-called non-thermal effects, which are not related to heating [Adair and Black, 2003; Blackman, 2009; Belyaev, 2015].

We measured temperature in two ways: (i) on the skin using a Flir i3 thermal imaging camera (Flir Systems, Wilsonville, OR); and (ii) in the exposed/sham exposed ear using a GentleTemp 510 tympanic ear thermometer (Omron, Kyoto, Japan). The thermal imaging camera had a spatial resolution of 72 dpi (dots per inch) on the horizontal and vertical axes. Thus, we were able to detect potential hotspots larger than 1 mm^2. Both methods provided an accuracy of 0.1 °C.

Protocol for RF EMF Exposure and Measurements

All exposures and sham exposures were performed blind under identical conditions. All records and results were verified blind to exposure conditions by Professor I. Tonhajzerova PhD, MD, who is a skilled physiologist and expert in HRV analysis. HRV was constantly measured during sham and real exposures. The protocol for exposure

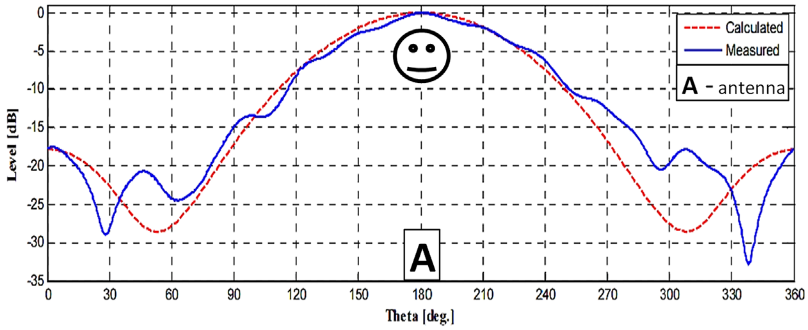

Fig. 3. Radiation pattern from plane circular waveguide feed. E was both measured (solid) and calculated (dashed) [Galuscak and Hazdra, 2008]. Head was located in main radiation lobe. *Theta* determines measurement angle while Level shows strength of electric field in dB.

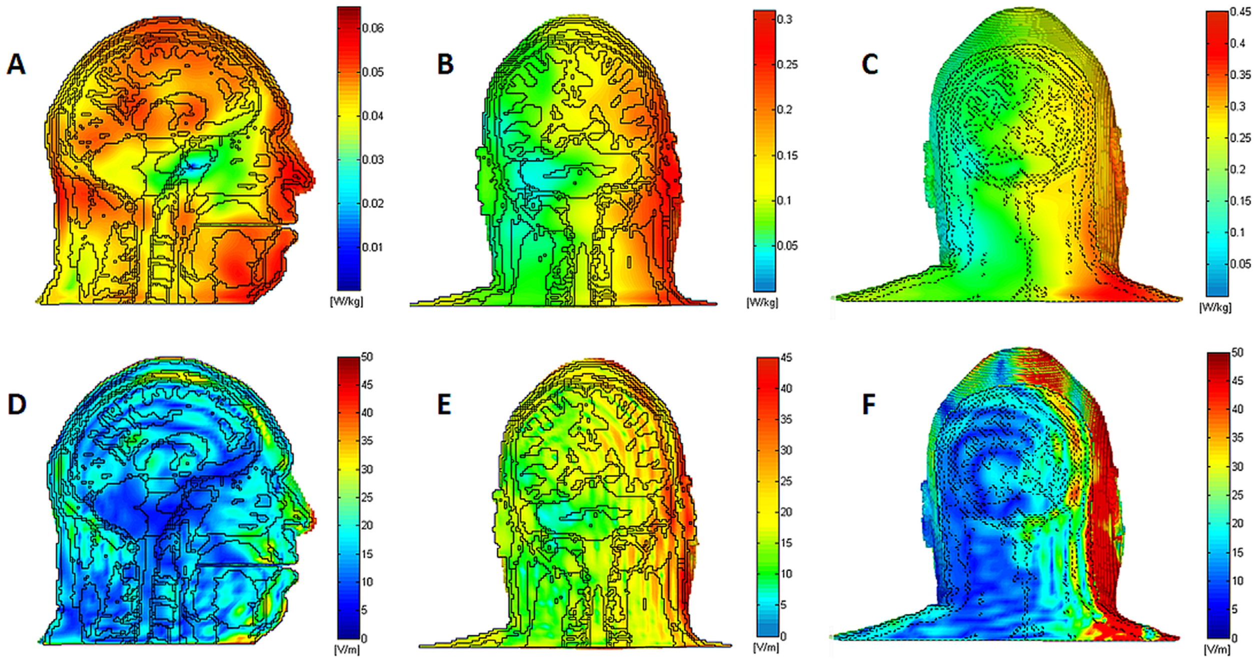

Fig. 4. Numerical simulation showing SAR_{10} spatial distribution in sagittal (A) and coronal (B, C) plane and E- field distribution in sagittal (D) and coronal (E, F) plane of human head phantom with the same signal and experimental setup as used for measurements.

and measurements is presented in Figure 5. The field was applied intermittently for 18 min in each trial (5 min on and 1 min off for period IV., V., and VI. in the exposed group). The participants were asked to rest in a sitting position for 15 min before the trial. After the rest period, we measured the perimeter of the head (followed by a calculation of head diameter), temperature, and BP using an Omron 705 IT sphygmomanometer (Omron, Kyoto, Japan). We applied ortho-clinostatic test to reveal functional changes in ANS. Thus, all exposures/sham-exposures were performed during standing-to-lying periods of the trial (Periods II.–III. and V.–VI. (Fig. 5). Periods I. and IV. were additional resting periods in a lying position. After the trial, temperature and BP were measured again.

All investigations were performed under well-controlled conditions at the same location in the same laboratory. The laboratory was semi-darkened and without any sound, visual, or smell disturbances, at a room temperature of 24.1 ± 0.8 °C (mean ± SD). Trials were always performed in the morning between 8:00–11:00 am in order to avoid any possible effects of circadian rhythms. HRV is very sensitive to any excitement that can modify HRV components, HR, and respiration rate. Therefore, no speaking, reading, or talking was permitted during measurement periods [Bernardi et al., 2000]. Prior to start of the trial,

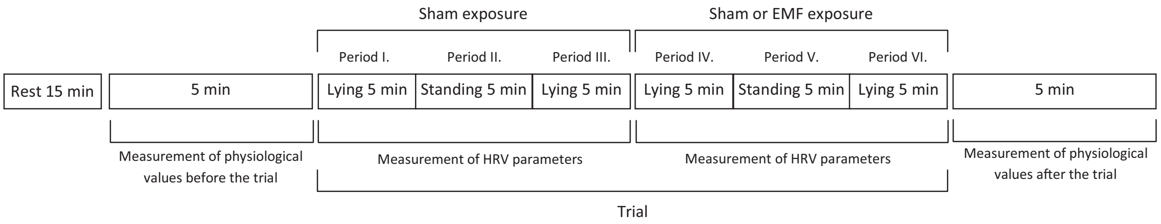

Fig. 5. Experimental protocol included a rest period, measurements of physiological parameters, and EMF exposure/sham-exposure trial ($n = 46$). Each trial included measurements of HRV during sham and real EMF exposure under conditions of ortho-clinostatic test (lying-to-standing-to-lying position). Periods I.–III.–sham-exposure, periods IV.–VI–either sham or real EMF exposure ($n = 23$).

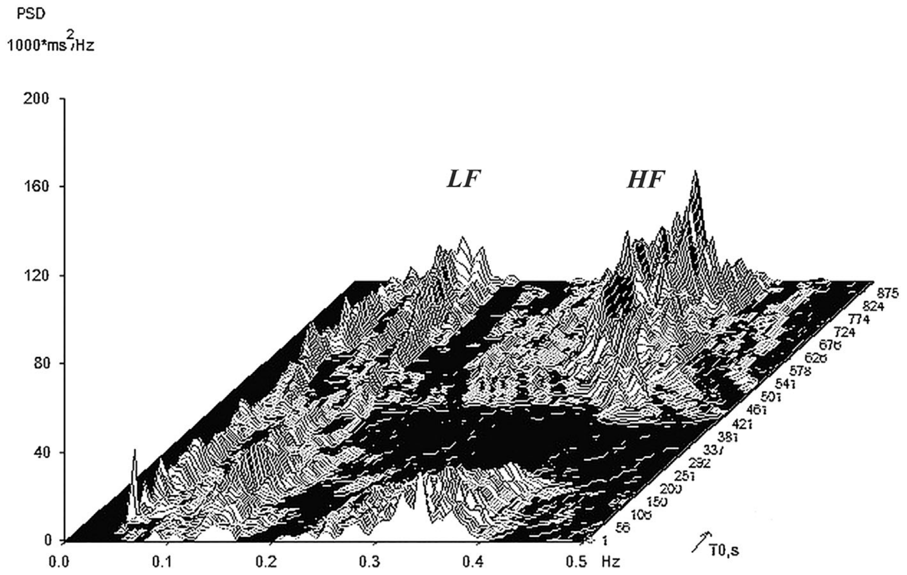

Fig. 6. Representative short spectral analysis of cardiac time (T0) series. Figure depicts power spectral density (PSD) with two distinct regions: low frequency (LF) and high frequency (HF) band for sham exposed student.

students were informed about experimental protocol and ortho-clinostatic test but not about sham- or EMF-exposure periods. A one-word command for position change was ordered by experimenter between recording periods, and thus did not disturb the HRV recordings.

Heart Rate Variability

All students underwent one-lead electrocardiograph (ECG) recording during the whole trial (Periods I.–VI., Fig. 5). A chest belt with two electrodes was situated on the thoracic segment of midclavicular lines (Fig. 2). The belt was wired to a Bluetooth device (DiANS PF8 HRV, Dimea Group, Olomouc, Czech Republic) that transmitted the recorded ECG signals online to a laptop. The Bluetooth transmitter emitted 2 mW in the 2.4 GHz band, the E-field being 0.87 V/m at a distance of 5 cm. To maximally reduce this field, the transducer was placed 0.5 m away from a tested student. Under physiological conditions, the QRS complex of ECG consists of Q, R, and S oscillations. It represents contraction (depolarization) of left and right ventricles detectable on the ECG waveform. The interval between R oscillations, the so-called RR interval, expresses the time that is inversely proportional to the HR frequency. We used a complex automated DiANS PF8 HRV system (Dimea Group, Olomouc, Czech Republic) to detect basic QRS points, RR intervals, and respiration rate. All recordings were subjected to HRV manual filtration to identify and eliminate artifacts and extra-systoles

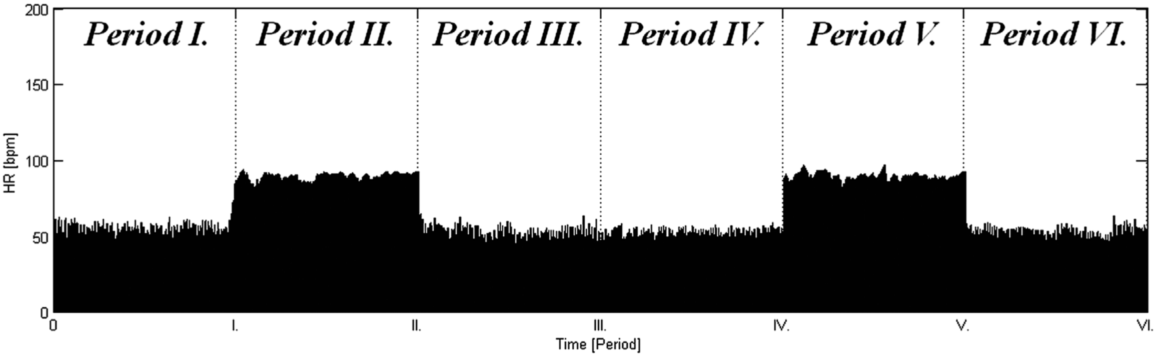

Fig. 7. Representative time domain analysis showing heart rate (HR) and heart rate variability (HRV) in each period of the trial for sham exposed student.

TABLE 2. No Statistically Significant Difference Was Revealed Between Sham-Exposed—Exposed (23 Students) and Sham–Sham-Exposed (23 Students) Groups in Their Physiological Parameters Before the Trial

Physiological parameter	P-value
Age [years]	0.5
BMI [kg/m^2]	0.5
Tympanic temperature [°C]	0.4
Blood pressure (Sys.) [mmHg]	0.9
Blood pressure (Dias.) [mmHg]	0.6
LF Spectral power [ln(ms^2)]	0.9
HF Spectral power [ln(ms^2)]	1.3
LF/HF	0.6
Total spectral power [ln(ms^2)]	1.0
RR [s]	1.0
rMSSD [ln(s)]	1.0
Respiration rate [fpm]	1.3

of heart's electric activity according to generally accepted recommendations [Camm et al., 1996].

Our experimental design included two types of standardized periods: a 5-min interval was used for the time domain parameters in order to take into account their dependence on time, and 300 heartbeats/interval for frequency (spectral) domain parameters.

Spectral (frequency) analysis of cardiac time series (Fig. 6) was used to determine two distinct regions: the low frequency (LF) and high frequency (HF) band. Parasympathetic activity contributes mainly to the HF component (0.15–0.5 Hz), whereas the LF component (0.04–0.15 Hz) mostly mirrors sympathetic and baroreceptor's activities. Some authors have suggested that parasympathetic nerve activity also contributes to the very low frequency band (VLF) HRV band below 0.04 Hz [Taylor et al., 1998]. Other authors have considered that the VLF band is under control of both parasympathetic and sympathetic activities [Bortkiewicz et al., 1996]. Due to unclear physiological interpretations (thermoregulation, renin-angiotensin-aldosterone system, etc.) [Bernardi et al., 1996; Mortara et al., 1997; Javorka, 2008], we decided to exclude VLF from evaluation.

Frequency and time domain analyses of the LF and HF bands were performed. The following parameters were calculated for the spectral domain: LF band spectral power [ln(ms^2)], HF band spectral power [ln(ms^2)], total spectral power (LF + HF) [ln(ms^2)] reflecting ANS activity [Javorka, 2008], and the LF/HF ratio, which is an indicator of sympatho-vagal balance. In the time domain (Fig. 7), we determined the RR intervals, respiration rate, and root Mean Square of the Successive Differences (rMSSD) [ln(s)] indicating parasympathetic nerve activity.

Statistics

Data processing was carried out using MS Office Excel and GraphPad InStat (La Jolla, CA) software. All data were analyzed for normal distribution with the Kolmogorov–Smirnov test. The non-normally distributed parameters (LF, HF, and total spectral powers, LF/HF ratio, and rMSSD) were log-transformed to obtain parameters with normal distribution. We set the statistical power to 0.80 for detection of expected 5% effect. The evaluation of differences was made by either Student's paired t-test or analysis of variance followed by adjustment of multiple comparisons using False Discovery Rate method. The confidence interval was set at 95% ($P < 0.05$).

RESULTS

Table 1 shows physiological characteristics of students from both groups. No difference in the physiological parameters was revealed between these groups (Table 2). Respiration rate and systolic and diastolic BPs were calculated in both groups. No significant changes were found after the trial when compared with measurements taken before the trial. Measurements of tympanic and skin temperature revealed no heating under RF exposure. On the

TABLE 3. HRV of Students Subjected to RF EMF Exposure (Period VI.) and Sham Exposure (Period III.) in a Lying Position (Mean ± SD, $n = 23$)

HRV parameter	Period III.	Period VI.	P-value
LF spectral power [ln(ms^2)]	5.64 ± 0.86	5.86 ± 1.19	0.3
HF spectral power [ln(ms^2)]	6.76 ± 1.42	7.06 ± 1.47	0.02*
LF/HF	0.85 ± 0.14	0.85 ± 0.16	0.8
Total spectral power [ln(ms^2)]	12.40 ± 2.06	12.92 ± 2.34	0.07
RR [s]	0.92 ± 0.12	0.97 ± 0.12	0.007**
rMSSD [ln(s)]	4.02 ± 0.72	4.17 ± 0.70	0.007**
Respiration rate [fpm]	12.89 ± 3.47	12.78 ± 3.40	0.9

*Statistical significance at $P < 0.05$; **Statistical significance at $P < 0.005$.

TABLE 4. HRV of Students Subjected to Sham Exposures in a Lying Position During Period III. and Period VI. (Mean ± SD, $n = 23$)

HRV parameter	Period III.	Period VI.	*P*-value
LF spectral power [ln(ms^2)]	5.54 ± 0.98	5.79 ± 0.76	1.4
HF spectral power [ln(ms^2)]	6.87 ± 0.96	6.86 ± 0.92	0.9
LF/HF	0.82 ± 0.15	0.86 ± 0.13	0.4
Total spectral power [ln(ms^2)]	12.41 ± 1.56	12.65 ± 1.33	1.0
RR [s]	0.93 ± 0.16	0.94 ± 0.16	1.1
rMSSD [ln(s)]	4.00 ± 0.48	4.02 ± 0.42	1.1
Respiration rate [fpm]	13.99 ± 3.08	13.43 ± 2.89	1.2

contrary, we noticed a slight decrease, although statistically insignificant, in tympanic temperature after exposure (Table 1A). We did not observe any pathological ECG changes or arrhythmias during or after the trials. However, RF exposure while lying (Period VI.; Table 3) resulted in statistically significant depression of HR (represented by RR interval; $P < 0.005$) in comparison to lying position (Period III.; Table 4) during sham-exposure.

The HF HRV band rose during RF EMF exposure in lying Period VI. (HF power: $P < 0.05$, rMSSD: $P < 0.005$), indicating an increase in parasympathetic nerve activity (Table 3). The tendency towards a rise in the total spectral power (Table 3; $P = 0.07$) in lying Position VI. was in line with these changes. The observed increase in HF band spectral power was unlikely caused by breathing, as the respiration rate did not change (Table 3; $P = 0.9$).

RF EMF exposure while standing, Period V. (Table 5), did not change any HRV parameters in comparison to sham-exposure, Period II. (Table 6). We did not find any significant HRV changes in the sham–sham exposed group during both lying and standing. This result indicated a feasibility of the study design.

None of the tested subjects were able to distinguish real exposure from sham exposure when queried at the end of the trial.

DISCUSSION

This study demonstrates that short-term RF EMF exposure may influence ANS ortho-clinostatic test. RF field at an intensity slightly lower than the ICNIRP guideline suppressed HR and increased both HF spectral power and rMSSD, showing elevation in parasympathetic nerve tone. An increase in parasympathetic nerve tone may be beneficial for people with excessively elevated HR or BP. However, none of the tested students had such an excessive increase. Thus, our study did not demonstrate any beneficial effect.

There was no measurable change in temperature during sham–sham exposure while we found a slight decrease, but not significant, in tympanic temperature during RF exposure. These data are in line with a previous study, which revealed that intermittent exposure to GSM900 uplink signal lowered the tympanic temperature compared to sham-exposure [Bortkiewicz et al., 2012].

Ortho-clinostatic test includes two positions that may increase the sensitivity of detecting RF EMF effects. Of note, RF exposure affected ANS only in the lying position while no effects were observed when standing. Prolonged recumbence can increase HF HRV stimulating parasympathetic nerve activity. Conversely, the standing position evokes sympathetic excitation associated with inhibition of parasympathetic nerve activity [Javorka, 2008]. The combination of these positions in the ortho-clinostatic test resulted in dynamic changes in parasympathetic nerve activity that may be more sensitive to RF EMF exposure than the static status of ANS in a standing position. Thus, our data suggest that ortho-clinostatic maneuver represents a

TABLE 5. HRV of Students Subjected to RF EMF Exposure (Period V.) and Sham Exposure (Period II.) in a Standing Position (Mean ± SD, $n = 23$)

HRV parameter	Period II.	Period V.	*P*-value
LF spectral power [ln(ms^2)]	5.93 ± 1.06	6.20 ± 1.02	0.08
HF spectral power [ln(ms^2)]	4.66 ± 1.34	4.72 ± 1.15	0.7
LF/HF	1.36 ± 0.35	1.36 ± 0.23	0.7
Total spectral power [ln(ms^2)]	10.59 ± 2.25	10.91 ± 2.10	0.4
RR [s]	0.64 ± 0.09	0.63 ± 0.08	0.3
rMSSD [ln(s)]	2.75 ± 0.65	2.74 ± 0.62	0.8
Respiration rate [fpm]	14.84 ± 2.80	14.42 ± 2.93	0.6

TABLE 6. HRV of Students Subjected to Sham Exposures in a Standing Position During Period II. and Period V. (Mean ± SD, $n = 23$)

HRV parameter	Period II.	Period V.	*P*-value
LF Spectral Power [ln(ms^2)]	6.42 ± 0.79	6.82 ± 0.71	0.6
HF Spectral Power [ln(ms^2)]	5.23 ± 0.91	5.31 ± 0.78	0.9
LF/HF	1.25 ± 0.16	1.30 ± 0.15	0.7
Total spectral power [ln(ms^2)]	11.64 ± 1.55	12.13 ± 1.36	1.0
RR [s]	0.64 ± 0.09	0.64 ± 0.10	1.0
rMSSD [ln(s)]	2.94 ± 0.47	3.00 ± 0.46	1.1
Respiration rate [fpm]	14.38 ± 2.40	14.62 ± 2.22	1.0

feasible experimental design for revealing possible RF effects on HRV.

The vagal nerve visceromotorically affects chronotropic function of the heart, which causes a marked decrease in HR (negative chronotropic effect) [Javorka, 2008]. Results of simulations revealed greater RF absorption in the neck area, while in the middle of the spinal cord and *medulla oblongata,* where the vagal nerve origins are located, SAR was about 0.1 W/kg (Fig. 4B). Thus, we may hypothesize that the E-field intensity of approximately 15 V/m in the area of the spinal cord and *medulla oblongata* (Fig. 4D and E) is sufficient for influencing HRV in a non-thermal way. Biophysical mechanisms, which have previously been proposed for non-thermal RF effects on cell membrane, voltage-gated ion channels, or change in action potential threshold values, may underlie the reported effects here [Belyaev, 2015].

Assuming that physiological interventions influence HRV in lying position, posture change from standing to lying (clinostatic test) is associated with "vagal rebound," that is, regulatory mechanism characterized by increased cardiac-linked parasympathetic efferents to sinoatrial node, resulting in lower HR and increased HRV. Thus, the reflection of physiological activation of parasympathetic subdivision in dynamic sympathovagal balance in lying position could occur. The same effect can be observed during prolonged recumbence. Therefore, we utilized ortho-clinostatic test to prevent this physiological reaction. In this context, other interventions can produce a change of sympathovagal balance towards parasympathetic dominance, for example, nonpharmacological noninvasive interventions, such as HRV training (biofeedback) or autogenic training [Taylor et al., 2010], or invasive direct vagal stimulation used in depressive disorders [Howland, 2014]. Also, listening to some specific frequency beats may increase parasympathetic activation and sympathetic withdrawal [McConnell et al., 2014]. Factors such as smoking [Karakaya et al., 2007], coffee and alcohol drinking [Spaak et al., 2010; Turnbull et al., 2017], sauna [Gayda et al., 2012], lack of sleep [Hall et al., 2004], or other sensory and motor interventions are commonly associated with stress reactions in humans. Changes in HRV associated with stress are represented by a decrease in parasympathetic nerve activity, increase in HR, and a higher level of sympathovagal balance compared to our results with a higher level of parasympathetic activity. Our study also included sham-exposed group, which prevented misinterpretation of the results.

Despite the fact that higher parasympathetic activity is associated with a lower risk of cardiovascular complications [Taylor et al., 2010], several studies referred to the association between high cardiovagal activity indexed by HRV parameters and acute cardiovascular complications [Mokra et al., 2013].

Our results are partially in line with those obtained by Andrzejak et al. [2008]. Those authors reported an increase in the HF component after volunteers were exposed to 1800 MHz GSM for 20 min while cell phones were in speaking mode, indicating cardiac vagal overactivity. However, these data were obtained from volunteers in a sitting position, while we observed the effects in supine (lying) position during the ortho-clinostatic test. A similar effect on the reduction in HR was obtained by Hietanen et al. [2002]. In their double-blind provocation study, subjects with reported idiopathic environmental intolerance attributed to EMF were tested during sham or real exposure to NMT 900 MHz, GSM 900, and 1800 MHz cell phones for 30 min. In line with our study, those authors also reported that none of the tested subjects were able to correctly distinguish between real and sham exposures. Wilen et al. [2007] showed a significant increase in total HRV power, lower HR, and an increase in parasympathetic nerve activity due to long-term exposure to low-level RF from plastic sealers. In contrast, Mohamed et al. [2011] exposed rats to RF from 1800 MHz cell phones for 4–8 weeks and observed activation of the sympathetic part of ANS with an increase in HR and systolic and diastolic BP, which increase the liability of hypertension.

Other authors did not identify any effect of RF EMF on HRV, HR, or BP; for example, Tahvanainen et al. [2004] (35 min of calling, 2 W 900 MHz, 1 W 1800 MHz), Nam et al. [2006] (CDMA, 30 min exposure, 835 MHz, SAR 1.6 W/kg), or Choi et al. [2014] (WCDMA, 35 min exposure with SAR 1.58 W/kg for 900 MHz and 0.70 W/kg for 1800 MHz). In all available studies, a number of methodological conditions, types of exposure signals, frequency bands, and other factors were often not the same and at least partially account for various outcomes. In addition, some exposure parameters such as SMF and ELF stray fields at the location of exposure, which are considered to be important for replication studies [IARC, 2013], were not described in these studies. Thus, the results of the available studies are not directly comparable.

Parazzini et al. [2013] reported no HRV effects or acute changes in the central regulation of ANS after exposure to 900 MHz from cell phone during 13 min. Of note, those authors found statistically significant increase in the LF component while in standing position, whereas no effects were observed in the lying position. HRV is predominantly influenced by parasympathetic nerve activity; thus, in our study the lying position was sensitive to detecting potential discrete abnormalities in cardiovagal function. We assume that continuous BP monitoring is important for studying sympathetic regulation of the cardiovascular system, while measurements of BP are missing in the Parazzini et al. [2013] study. Further research using advanced monitoring techniques such as continuous BP or skin conductance measurements are needed to resolve this question. As was mentioned above, the difference in exposure system (cell phone), exposure signal (GSM 900 MHz), and different study group (age range 21–28 years) might also cause miscellaneous effects in comparison to our study.

In this study, we observed statistically significant effect of RF exposure on selected HRV parameters. Thus, this study may indicate a new strategy for analyzing mechanisms and the significance of the observed effect in future trials. It is also possible to modify some parameters (e.g., time, signal type, modulation, incidence area, etc.) to explore and exploit new findings on HRV analysis that might provide an interesting avenue for further research. It is important to note that HRV provides information regarding autonomic effects of cardiac chronotropic function. In particular, analysis of nonlinear features in beta-adrenergic activity may yield complementary information on sympathetic nerve activity related to interaction of RF EMF with cardiac autonomic regulation.

One limitation of our study is the linear data evaluation, while calculating nonlinear HRV parameters may reveal new outcomes [Parazzini et al., 2013]. Of note, larger antenna dimensions were used in this study compared to standard dimensions of cell phones, resulting in differences of SAR distribution. However, even the maximal SAR was below the thermal values in our experiments. The authors are aware that only short-term exposure was evaluated, and chronic intermittent RF EMF exposure warrants further study.

CONCLUSION

Our results showed that RF EMF has a noticeable effect on HRV parameters, which can be revealed with an ortho-clinostatic test. Short-term intermittent RF EMF exposure affected ANS, leading to a significant increase in HRV indicators such as HF band spectral power and rMSSD, and a decrease in HR (measured by the RR interval). The obtained data indicate that short-term exposure to RF EMF under given conditions increases parasympathetic nerve activity. Closer attention should be dedicated to long-term chronic exposure from widespread use of cell phones.

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