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The effect of radiofrequency radiation on DNA and lipid damage in female and male infant rabbits

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Abstract

Purpose: **We aimed to design a prolonged radiofrequency (RF) radiation exposure and investigate in an animal model, possible bio-effects of RF radiation on the ongoing developmental stages of children from conception to childhood.**

Materials and methods: **A total of 72 New Zealand female and male white rabbits aged one month were used. Females were exposed to RF radiation for 15 min/day during 7 days, whereas males were exposed to the same level of radiation for 15 min/day during 14 days. Thirty-six female and 36 male infant rabbits were randomly divided into four groups: Group I [Intrauterine (IU) exposure (−); Extrauterine (EU) exposure (−)]: Sham exposure which means rabbits were exposed to 1800 MHz Global System for Mobile Telecommunication (GSM)-like RF signals neither in the IU nor in the EU periods. Group II [IU exposure (−); EU exposure (+)]: Infant rabbits were exposed to 1800 MHz GSM-like RF signals when they reached one month of age. Group III [IU exposure (+); EU exposure (−)]: Infant rabbits were exposed to 1800 MHz GSM-like RF signals in the IU period (between 15th and 22nd days of the gestational period). Group IV [IU exposure (+); EU exposure (+)]: Infant rabbits were exposed to 1800 MHz GSM-like RF signals both in the IU period (between 15th and 22nd days of the gestational period) and in the EU period when they reached one month of age. Biochemical analysis for lipid peroxidation and DNA damage were carried out in the livers of all rabbits.**

Results: **Lipid peroxidation levels in the liver tissues of female and male infant rabbits increased under RF radiation exposure. Liver 8-hydroxy-2′-deoxyguanosine (8-OHdG) levels of female rabbits exposed to RF radiation were also found to increase when compared with the levels of non-exposed infants. However, there were no changes in liver 8-OHdG levels of male rabbits under RF exposure.**

Conclusion: **Consequently, it can be concluded that GSM-like RF radiation may induce biochemical changes by increasing free radical attacks to structural biomolecules in the rabbit as an experimental animal model.**

Keywords: Radiofrequency radiation, 8-hydroxy-2'-deoxyguanosine, 8-OHdG, malondialdehyde, MDA, FOX, liver, female and male infant

Introduction

Intense exposure to radiofrequency (RF) radiation emitted from portable devices or base stations from childhood to adulthood is one of the most curious subjects in the scientific arena. In the 20th century, scientists have focused on this subject faced with an increase in the incidence of leukemia among 2-to 5-year-old pre-school children (Milham and Ossiander 2001). The International Agency for Research on Cancer (IARC) has declared extremely low frequency (ELF) magnetic (B) fields as the potential carcinogenic agent to humans mainly based on epidemiological studies on childhood leukemia (IARC 2002). Recently, the IARC Monograph Working Group has discussed and classified RF field exposure as the possible carcinogenic agent to humans (24–31 May 2011; Lyon, France). To evaluate the available information relevant to children's sensitivity to electromagnetic fields (EMF) and to identify research needs, the World Health Organization (WHO) held an expert workshop in Istanbul, Turkey, in June 2004 (Kheifets et al. 2005). Concerns about the sensitivity of children to ELF and RF fields were discussed in detail in this workshop. In the literature, residential exposure to RF radiation from broadcast transmitters has provided limited information, only on the leukemia incidence rates (Ha et al. 2007, Merzenich et al. 2008, McKinney et al. 2008, Micheli, 2010), whereas further research is required concerning individual exposure (Schüz and Ahlbom 2008). Pre-school children may also be considered to be more sensitive to any negative health effects than adults because of the greatest absorption of mobile phone radiation. Furthermore, depending on the size of a child's head, there can be the head resonance effect and greater ease in penetration of Global System for Mobile Telecommunication (GSM) radiation to

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the thinner skull of an infant (Hyland 2000). Particularly in the developmental stages (from conception to adulthood), their brain tissues, with high content of water and ions, absorb more RF radiation at mobile phone frequencies (Kheifets et al. 2005). Increased intensity and extended duration of exposure to RF radiation during these stages may lead to inherited disorders by altering the conformation of structural molecules.

The direct causal relations between RF fields and genetic disorders are still inconsistent because of their insufficient energy level to affect the macromolecular bonds (Repacholi 1997, Ahlbom et al. 2004, Zmyślony et al. 2004). However, the discovery of the Bystander Effect in radiobiology illustrates that direct quantum energy is not necessary to induce DNA strand breaks (Azzam et al. 1998, Mothersill and Seymour 1998, 2006, Brenner and Sachs 2002). It is vital to clarify whether or not RF exposure causes DNA damage and leads to chromosome breaks.

There are three possible ways in the literature in which RF fields can interact in the body and cause biological damage, including molecular bond breakage. One of these concerns natural magnetite particles in the body which could respond to RF radiation (Kirschvink 1996) and indeed to 217 Hz modulation. The other one is the so-called Radical Pair Mechanism (RPM) where low intensity magnetic fields can effectively increase the lifetime of free radicals (Brocklehurst and McLauchlan 1996, Chignell and Sik 1998, Ritz et al. 2000, 2004, 2010). This would likely affect the 217 Hz modulation rather than the RF mobile phone frequency carrier wave. There are also several studies related to the effects of RF radiation on DNA damage, gene expression and chromosomal abnormalities (Garaj-Vrhovac et al. 1992, Lai and Singh 1997, Goswami et al. 1999, Harvey and French 2000, Leszczynski et al. 2002, Belyaev et al. 2006, Remondini et al. 2006, Buttiglione et al. 2007, Friedman et al. 2007, Garaj-Vrhovac and Orescanin 2009). Some of these changes reported in the genetic material may be caused by cellular stress induced by RF radiation; in particular, free radicals or reactive oxygen/nitrogen derivatives as sources of cellular stress. An increase in the formation of highly reactive molecules under RF radiation may be one of the reasons lying behind the non-thermal effect on biological systems (Zmyślony et al. 2004, Belyaev 2005a, 2005b). There are scarce experimental data to support a biologically plausible mechanism for RF-induced free radical generation. In the biochemical approach, the most important oxygen-free radical causing damage to the basic biomolecules (proteins, membrane lipids, and DNA) is the hydroxyl radical (HO·).

Erkoç and Erkoç (2002) theoretically investigated the structural and electronic properties of guanine and guanosine by performing semi-empirical and *ab initio* molecular orbital theory calculations and concluded that guanine was a highly polar molecule; therefore, it might interact with its surroundings, especially with other polar molecules in the cell in a stronger way. Thus, this makes guanine molecule a potential threat to cellular damage. The interaction of hydroxyl radical (HO·) with nucleobases in the DNA strand, such as guanine, leads to the formation of C8-hydroxyguanine (8-OHGua) or its nucleoside form deoxyguanosine (8-hydroxy-2-deoxyguanosine) which is the predominant form of free radical-induced oxidative lesions, and has therefore been widely used as a biomarker for oxidative stress (Valavanidis et al. 2009).

Other markers formed by the interaction between unsaturated membrane phospholipids and reactive oxygen species and nitrogen species have led to lipid peroxidation end products and are determined by thiobarbituric acid reactive substances (TBARS) (malondialdehyde [MDA] assay) and ferrous oxidation in xylenol orange (FOX) assays (Esterbauer and Cheeseman 1990, Janero 1990, Bonnes-Taourel et al. 1992). Organoperoxides are early products and MDA is a later product of lipid oxidation of polyunsaturated lipids such as arachidonic acid (Esterbauer et al. 1991). There may be an increase in organoperoxides, independent of MDA (Chamblee et al. 2000), and the results are raising the question of whether measurement of organoperoxides by procedures such as the FOX assay rather than MDA might provide a more consistent/reliable index of oxidative stress.

In the present study, the principal aim was to design the continual RF exposure and investigate the possible bio-effects of RF radiation on the ongoing developmental stages of children from conception to childhood. The levels of lipid peroxidation and DNA damage based on free radical attacks were analyzed in the liver tissues of baby rabbits aged one month.

Materials and methods

Animals

A total of 72 one-month-old female and male New Zealand white rabbits were used in this study. The animals were obtained from the Laboratory Animals Breeding and Experimental Research Center of Gazi University. The experimental protocol was reviewed and approved by the Laboratory Animal Care Committee of Gazi University (G.U.ET-06.027).

Thirty-six of the infant rabbits were exposed to 1800 MHz GSM-like RF radiation for 15 min/day during a week in the intrauterine period (between 15th and 22nd days of the gestational period when the transition from embryogenesis to organogenesis takes place) whereas others were not exposed. After birth, all 72 infant rabbits were kept with their mothers until they reached one month of age. They were breastfed and their optimum growth was obtained during this one-month period.

Baby rabbits aged one month were housed under the same conditions in a temperature and humidity-controlled room (20 ± 1°C, 50 ± 10% relative humidity) and 14/16 h light/dark cycle conditions. The animals were provided with tap water and standard pelletized food *ad libitum* except during exposure periods.

Only one animal was placed in each cage during each radiofrequency radiation (RFR) exposure period because placing more than one animal in a cage could have created stress. All of these infant rabbits were placed with their mothers and remained so until they reached one month of age. They were breastfed and their optimum growth was obtained during this one-month period.

Exposure level and quality control

The GSM-like RF exposure system is illustrated in detail in Figure 1. To provide a brief summary, GSM-like signals in

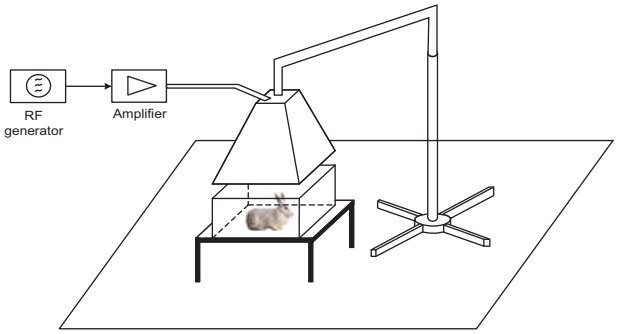

Figure 1. GSM-like radiofrequency exposure system.

1800 MHz frequency were formed by using a signal generator (Agilent Technologies 8648C, 9 kHz–3.2 GHz, Santa Clara, CA, USA) with the integrated pulse modulation unit and horn antenna (Schwarzbeck, Doppelsteg Breitband Horn antenna BBHA 9120 L3F, 0.5–2.8 GHz, Schönau, Germany) in a shielded room. The length of the cable which connects the signal generator to the antenna was 2 m long, i.e., the lateral distance between the signal generator and the antenna was approximately 2 m.

On the other hand, it should be emphasized that the distance between the midline of the rabbit's head and the directional horn antenna which generated the RF energy was 20 cm. The generated power was controlled by a spectrum analyzer (Agilent Technologies N9320A, 9 kHz–3 GHz, Santa Clara, CA, USA) integrated to the signal generator. The signals were amplitude-modulated by rectangular pulses with a repetition frequency of 217 Hz and a duty cycle of 1:8 (pulse width 0.576 ms), corresponding to the dominant modulation component of the GSM.

An RFR generator provided 20 dBm (0.1 W) power during the exposure period. The signal was controlled by means of the spectrum analyzer connected to the signal generator, and an electromagnetic radiation meter (EMR 300 meter with type 26.1 probe, NARDA Safety Test Solutions, Pfullingen, Germany) was used for measurement of output radiation. Measurements were taken during the entire experimental duration and data were stored in a computer connected to the device via fiber optic cable. Specific Absorption Rate (SAR) was calculated as 1.8 W/kg by Finite Difference Time Domain (FDTD) method by using a homogenous rabbit model

Experimental design

Groups of animals

A total of 72 one-month-old female and male New Zealand white rabbits were used. Thirty-six females were exposed to RF radiation for 15 min/day during 7 days, whereas 36 males were exposed to the same level of radiation for 15 min/day during 14 days.

Female and male infant rabbits were randomly divided into four groups:

Group I [Intrauterine exposure (−); Extrauterine exposure (−)]: Sham exposure which means rabbits were exposed to 1800 MHz GSM-like RF signals neither in the intrauterine (IU) nor in the extrauterine (EU) periods.

Group II [Intrauterine exposure (−); Extrauterine exposure (+)]: Infant rabbits were exposed to 1800 MHz GSM-like RF signals when they reached one month of age.

Group III [Intrauterine exposure (+); Extrauterine exposure (−)]: Infant rabbits were exposed to 1800 MHz GSM-like RF signals in the IU period (between 15th and 22nd days of the gestational period).

Group IV [Intrauterine exposure (+); Extrauterine exposure (+)]: Infant rabbits were exposed to 1800 MHz GSM-like RF signals both in the IU period (between 15th and 22nd days of the gestational period) and in the EU period when they reached one month of age.

The day after the last exposure, baby rabbits were anesthetized and sacrificed with ketamine (35 mg/kg, i.m.) and xylazine (5–10 mg/kg, i.m.).

Biochemical analysis

Liver tissues were removed, washed free from contaminated blood with ice-cold buffered saline. For the determination of malondialdehyde (MDA Assay), tissue MDA levels were determined by using TBARS assay (a major aldehyde species for lipid peroxidation). The difference in the absorbances of the two measurements from the butanol phase was used as the MDA value (nmol/g tissue) (Mihara and Uchiyama 1978).

For the determination of lipid peroxidation (FOX assay), the assay was intended for the quantitative determination of low levels of lipid hydroperoxides in the samples. It is based on the oxidation of ferrous ions (Fe^{2+}) to ferric ions (Fe^{3+}) by hydrogen peroxide under acidic conditions. The ferric ion binds with the indicator dye xylenol orange to form a stable colored complex which can be measured at 560 nm as an indirect measure of hydroperoxide (HP) concentration with incubation time modifications. HP equivalents (HPE, nmol/g wet weight) were calculated according to Hermes-Lima et al. (1995).

For the measurement of oxidative DNA damage (lesions/10^6 DNA nucleosides), genomic DNA of liver tissues was extracted with Roche DNA extraction kit, then denatured by heating at 95°C for 3 min and cooled on ice; 100 µl 2 mM deferoxamine mesylate (DFAM) and 20 mM acetate buffer (pH = 5) were added to the denaturated DNA. DNA content was analyzed spectrophotometrically at 260 nm and then hydrolyzed to nucleotides by incubation with 4 µl of 3.3 mg/ml suspension of nuclease P1. Tris hydrochloride (Tris-HCl) buffer (pH = 8.5) was added to the mixture and hydrolysis to corresponding nucleosides was achieved by incubation with calf intestinal alkaline phosphatase for 1 h at 37°C. After adding acetate buffer and 50 mM ethylenediaminetetraacetic acid (EDTA)/10 mM DFAM solution, the mixture was filtered through a 0.22 µm Millipore filter unit (UltraFree, Bedford, MA, USA) and centrifuged at 10,000 *g* for 20 min at 4°C. Reverse-phase High Performance Liquid Chromatography with Electrochemical Detection (HPLC-EC) was performed as described by Floyd et al. (1986). The DNA hydrolysate was injected onto a Waters C18 reverse-phase column (5 µm, 0.46 × 25 cm; Waters Assoc., Milford, MA, USA) at a flow rate of 1 ml/min. The mobile phase was 50 mmol/l phosphate

buffer (pH = 5.5) with 5% methanol (Halliwell and Dizdaroglu 1992, Hamilton et al. 1999).

The eluant was monitored at 290 nm for the ultraviolet detection of deoxyguanosine (dG) and at 0.6 V for the electrochemical detection of 8 hydroxy-2-deoxyguanosine (8-OHdG). The system was calibrated with authentic dG and 8-OHdG standards (Sigma Chemical, St Louis, MO, USA). dG had a retention time of 10–12 min and 8-OHdG had a retention time of 8.7–13.8 min. Standards were run after every fifth sample for verification, and the data were expressed as the ratio of 8-OHdG to 10^6 dG.

Statistical analysis

Data analysis was carried out using the SPSS 11.5 statistical package (SPSS, Chicago, IL, USA). The Kruskal-Wallis H test (non-parametric) was applied to evaluate differences among all groups while differences between pairs of groups were evaluated by means of the Mann-Whitney U-test. The results were expressed as median (interquartile range [IQR]) values.

Results

The results for female and male infant rabbits are shown in Tables I and II and Figures 2–4.

For infant female rabbits

MDA levels were found significant in the multiple comparison of experimental groups and controls (Kruskal-Wallis [Groups I–IV], $p = 0.002$). Intergroups comparison (Groups I–II, I–III, II–IV) of MDA level were found non-significant, except for Groups I–IV ($p = 0.001$, Mann-Whitney) and III–IV ($p = 0.003$, Mann-Whitney).

FOX levels were found non-significant in the multiple comparison of experimental groups and controls (Kruskal-Wallis [Groups I–IV], $p = 0.85$). After Mann-Whitney test, a non-significant increase was found in liver FOX levels (Groups I–II, I–III, I–IV, II–IV and III–IV).

8-OHdG levels were found significant in the multiple comparison of experimental groups and controls (Kruskal-Wallis [Groups I–IV], $p = 0.0001$). Intergroup comparisons between Groups I–II ($p = 0.001$, Mann-Whitney), Groups I–IV ($p = 0.0001$, Mann-Whitney) and Groups III–IV ($p = 0.001$, Mann-Whitney) were significant, but non-significant increases were determined for Groups I–III ($p = 0.290$, Mann-Whitney) and II–IV ($p = 0.233$, Mann-Whitney).

There was also a significant positive correlation between MDA and 8-OHdG levels ($r = 0.457$, $p = 0.008$).

For infant male rabbits

MDA levels were found significant in the multiple comparison of experimental groups and controls (Kruskal-Wallis [Groups I–IV], $p = 0.0001$). Intergroup comparisons (Groups I–II, I–IV, and II–IV) of MDA level were found to be non-significant, except for Groups I– III ($p = 0.001$, Mann-Whitney) and III–IV ($p = 0.001$, Mann-Whitney).

FOX levels were found non-significant in the multiple comparison of experimental groups and controls (Kruskal-Wallis [Groups I–IV], $p = 0.109$). Intergroup comparisons (Groups I–II, I–IV, and II–IV) of FOX level were found non-significant, except for Groups I–III ($p = 0.034$, Mann-Whitney) and III–IV ($p = 0.040$, Mann-Whitney).

8-OHdG levels were found non-significant in the multiple comparison of experimental groups and controls (Kruskal-Wallis [Groups I–IV], $p = 0.441$). After the Mann-Whitney U-test, liver 8-OHdG levels were found to increase to non-significant levels for all experimental groups (Groups I–II, I–III, I–IV, II–IV and III–IV).

A significant negative correlation was found between MDA and 8-OHdG levels ($r = -0.474$, $p = 0.009$)

Discussion

In the present study, possible biological effects of acute exposure to GSM-like signals were examined during the prenatal and postnatal stages of female and male rabbits aged one-month-old. Oxidative DNA damage and lipid peroxidation levels in the liver tissues of all animals were observed after exposure to 1800 MHz RF radiation.

In recent years, there has been increasing concern about the possible health effects of exposure to RF radiation emitted by different man-made sources including mobile phones and base stations, television and radio broadcasting facilities, radar, medical equipments, industrial heaters and electronic devices daily used in homes and offices. However, bio-effects induced by RF fields still remain uncertain. RF radiation, frequency range of 30 kHz–300 GHz, is in the non-ionized part of the electromagnetic spectrum because its photon energy is really far too low to directly influence chemical bonds of critical macromolecules (Sienkiewicz 1998). Moreover, it is well known that RF fields can transfer their energy to biological matter resulting in the increment of medium temperature due to the vibration of atoms and molecules (Polk and Postow 1996). With regard to another approach, the essential effects of EM fields can be observed in the structures of two major macromolecules: Transport proteins in membranes and nuclear DNA in charge of protein synthesis. EM fields

Table I. Effect of 1800 MHz GSM-like radiation on lipid peroxidation and oxidative DNA damage in female infant rabbits.

Groups	MDA (nmol/g tissue)	FOX (HPE, nmol/ g wet weight)	8-OHdG (8-OHdG/10^6 dG)
I	180 (53–78)	4048 (2168–3272)	0.65 (0.04–0.15)
II	246 (120–152)	4678 (797–2760)	0.81 (0.08–0.14)
III	192 (67–73)	3487 (2518–3058)	0.69 (0.07–0.09)
IV	280 (113–136)	4636 (2364–2862)	0.84 (0.04–0.05)

All values are expressed as median (IQR) values.

Table II. Effect of 1800 MHz GSM-like radiation on lipid peroxidation and oxidative DNA damage in male infant rabbits.

Groups	MDA (nmol/g tissue)	FOX (HPE, nmol/ g wet weight)	8-OHdG (8-OHdG/10^6 dG)
I	199 (63–145)	2148 (1312–3717)	0.64 (0.06–0.12)
II	223 (62–96)	2527 (2750–3898)	0.66 (0.04–0.05)
III	112 (25–88)	4112 (1775–3353)	0.68 (0.04–0.06)
IV	213 (42–72)	2522 (1198–1554)	0.68 (0.03–0.05)

All values are expressed as median (IQR) values.

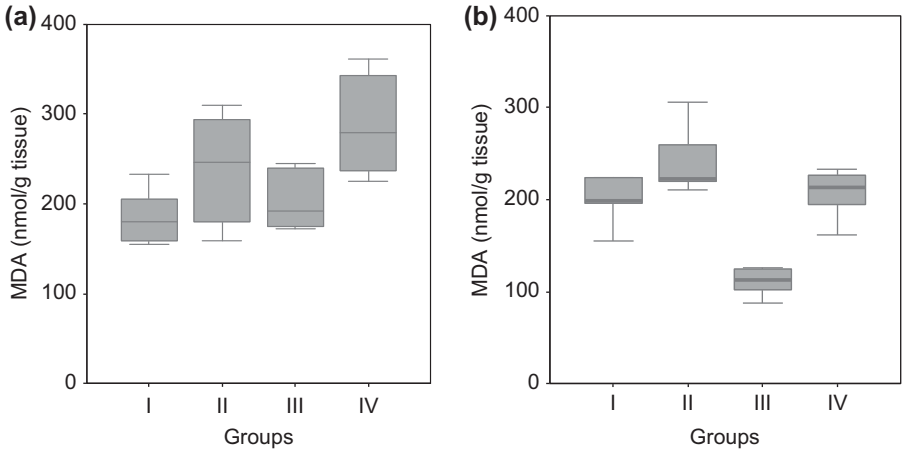

Figure 2. Effects of 1800 MHz GSM-like radiation on malondialdehyde (MDA) levels in (a) female infant rabbits ($n = 36$) and (b) male infant rabbits ($n = 36$). I. Group I [Intrauterine exposure (−); Extrauterine exposure (−]); II. Group II [Intrauterine exposure (−); Extrauterine exposure (+)]; III. Group III [Intrauterine exposure (+); Extrauterine exposure (−)]; IV. Group IV [Intrauterine exposure (+); Extrauterine exposure (+)]. All values are expressed as median (IQR) values.

can cause changes both in charge distributions and conformations of these macromolecules (Blank 2008).

Studies in the literature on RF radiation and its interaction with macromolecules show that it is possible to observe inhibition of DNA synthesis, transcription, RNA processing and translation, inhibition of cell cycle progression, protein denaturation, alterations in cellular metabolism, changes in membrane permeability and induction of chronic overexpression of heat-shock proteins (McNamee and Chauhan 2009). It should be noted that RF radiation may also induce the activation of both cellular signaling mechanisms and reactive free radicals (Belyaev 2005, 2005a). Oxidative stress associated with the augmentation of ROS generation can lead to oxidative damage in structural and functional biomolecules such as lipids, proteins and nucleic acids. DNA damage, repair and mutations are important events in carcinogenesis. In this study, the formation of 8-OHdG in liver DNA samples of female and male infant rabbits aged one month under the acute exposure of 1800 MHz GSM-like RF signals was studied to assess the risk prediction of prolonged exposure during the developmental stages. According to our results, liver 8-OHdG levels were found to have increased by almost 80% in female infant rabbits exposed to RF radiation in the intrauterinary and also extrauterinary period compared to non-exposed infant rabbits. However, no changes were obtained in 8-OHdG levels of male infant rabbits among exposed and non-exposed rabbits. This difference may be due to differential exposure of male and female rabbits. Although male rabbits were exposed more than the females, it may be hypothesized that they endured a recovery effect.

In the preliminary project of this study, the bio-effects of GSM-like signals on 8-OHdG levels of liver and brain tissues in adult non-pregnant and pregnant rabbits were analyzed. In the adults, no changes were determined in liver 8-OHdG levels of exposed rabbits compared to the non-exposed. Their newly-born rabbits (max. 2 days) were also investigated in terms of oxidative DNA damage and lipid peroxidation from the intrauterine RF exposure. Our previous results showed

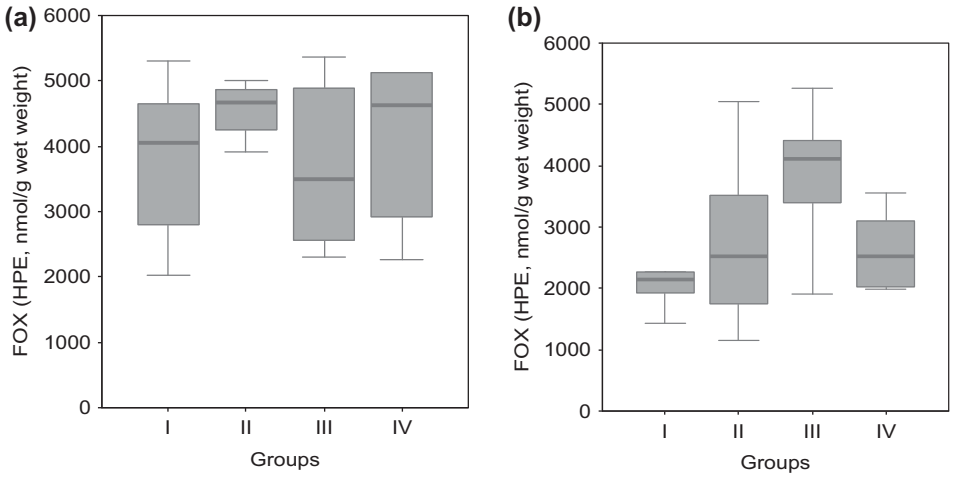

Figure 3. Effects of 1800 MHz GSM-like radiation on (FOX, nmol/g wet weight) levels in (a) female infant rabbits ($n = 6$) and (b) male infant rabbits ($n = 36$). I. Group I [Intrauterine exposure (−); Extrauterine exposure (−)]; II. Group II [Intrauterine exposure (−); Extrauterine exposure (+)]; III. Group III [Intrauterine exposure (+); Extrauterine exposure (−)]; IV. Group IV [Intrauterine exposure (+); Extrauterine exposure (+)]. All values are expressed as median (IQR) values.

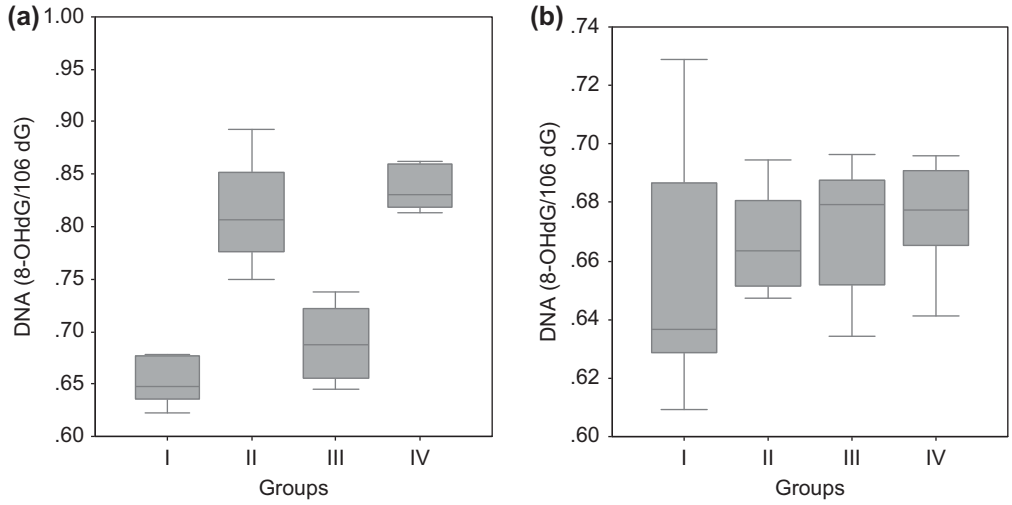

Figure 4. Effects of 1800 MHz GSM-like radiation on 8-hydroxy-2'-deoxyguanosine (8-OHdG) levels in (a) female infant rabbits ($n = 36$) and (b) male infant rabbits ($n = 36$). I. Group I [Intrauterine exposure (−); Extrauterine exposure (−)]; II. Group II [Intrauterine exposure (−); Extrauterine exposure (+)]; III. Group III [Intrauterine exposure (+); Extrauterine exposure (−)]; IV. Group IV [Intrauterine exposure (+); Extrauterine exposure (+)]. All values are expressed as median (IQR) values.

that whole-body 1800 MHz GSM-like RF exposure could affect lipid peroxidation by increasing MDA and FOX levels in both adults and their newborns (Tomruk et al. 2010). The results were very different in the brain tissues of adult rabbits. Brain tissues of pregnant and non-pregnant rabbits were affected by RF radiation, while there was no effect in their newborns. These findings may be interpreted as a result of the depth of penetration phenomenon. As RFR propagates in the tissue medium, energy is absorbed by the tissue, resulting in a progressive reduction of RFR as it advances in the tissue (Polk and Postow 1996, Güler et al. 2010). Recent studies have shown that RFR emitted from cellular phones could increase the release of free radicals. Meral et al. (2007) revealed that RF radiation generated from cellular phones (12 h/day, 30 days) may produce oxidative stress by increasing lipid peroxidation of brain tissues in guinea pigs. Ozgur et al. (2010) showed that GSM-like radiation can cause significant modification in the activities of liver antioxidant enzymes. However, the administration of an external antioxidant has a protective effect against the RF radiation by boosting the antioxidant activity. In addition to these, RFR may cause oxidative damage in the fetus; this could be caused by melatonin pathway disruption in the mother (Wakatsuki et al. 1999, 2001).

Consequently, there are few studies published in the open literature on the prolonged effects of GSM-like radiation on children (Bartsch et al. 2010). For this reason, further studies on this subject will be beneficial in order to protect children from environmental RFR exposure by drawing the attention of decision-makers and finally succeed in the establishment of international standards for the protection of children.

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Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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